

Bulletin of

**Veterinary Science &
Animal Husbandry**

Nepal



Official Organ of Nepal Veterinary Association

BULLETIN OF VETERINARY SCIENCE & ANIMAL HUSBANDRY, NEPAL

Vol. 26

2000

Regd. ZCBA 7/2025/26

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NEPAL VETERINARY ASSOCIATION

Veterinary Complex
Tripureshwor, Kathmandu, Nepal
Tel: 257496
Fax: 242867, 261521

Annual Subscription:

SAARC Countries: Rs. 40.00

Other Countries: US\$ 10.00

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EDITORIAL

Livestock sector since time immemorial has significant role in Nepalese farming households for their family income and as an indispensable part of our agriculture system. It is also an important source of animal protein in human diet. Furthermore improvement in livestock production is amongst the few alternatives available to the rural communities to improve their standards of living. Realising its tremendous potential for growth, national planners have given due importance to the livestock sector in the Agriculture Perspective Plan, a long-term policy of the government for agriculture development. Accordingly, the ninth five year plan has envisaged several activities to improve the production and productivity of the Nepalese livestock. Apart from inherent characters of our indigenous livestock population, animals health problems related to different infectious diseases, development of resistance in disease causing organisms, insects and pests are the major constraints to increase livestock production and productivity.

With restoration of democracy in the country, the role of the government is redefined mainly as a motivator, facilitator, promoter and regulator instead of so called "Contractor of Development" of the past. In this changed context, privatisation of veterinary services could play a complementary role towards providing effective veterinary services, which will be a milestone for flourishing a viable livestock industry of the country. This on the other hand will ample opportunities for employment in private sector reducing the excessive dependence upon the government for employment. To ensure quality veterinary services, production of qualified man power is imperative and establishment of Veterinary college at Institute of Agriculture and Animal Sciences at Rampur, is another step towards fulfilling the vacuum that was created.

The theme of this conference was timely and the issues discussed were relevant to the national goal. The conference has been an excellent platform offered to its participants to interact and share technical knowledge and experiences for their scientific advancement.

As decided by the executive committee and announced in our earlier volume of Bulletin of Veterinary Sciences and Animal Husbandry, will be published as "NEPALESE VETERINARY JOURNAL" this issue onward. We hope to receive cordial help and support from all colleagues working in this field, in our endeavour to establish Nepalese Veterinary Journal as a reputed journal.

**PROCEEDINGS OF
THE SIXTH NATIONAL CONFERENCE OF
NEPAL VETERINARY ASSOCIATION**

(Vetcon 99)

Kathmandu

February 22-24, 1999

The sixth national conference of Nepal Veterinary Association was held from February 22 to 24, 1999, with the theme of "*Effective Veterinary Services for Poverty Alleviation*", at International Convention Hall, Baneshwor and Administrative Staff College, Jawalakhel, Kathmandu.

The conference was inaugurated by Honourable Minister for Education, Mr. Kul Bahadur Gurung, as chief guest and chaired by Honourable Vice-chairman of National Planning Commission Mr. Prithvi Raj Ligal. The chief guest pointed out the role of livestock in the national economy and its potential contribution in poverty alleviation. The welcome speech was delivered by Dr. Pralhad Sapkota, President of NVA and the objectives and activities of the association were highlighted by the General Secretary, Dr. Narayan Prasad Ghimire.

Dr. Mukti Narayan Shresth, acting Secretary, Ministry of Agriculture stated the importance of effective veterinary services towards achieving the national goal of poverty alleviation and also stressed upon the role of veterinarians to be played to achieve the livestock production target set by Agriculture Perspective Plan. Honourable member of National Planning Commission, Mr. Hari Shankar Tripathi also addressed and highlighted the need of veterinary services to boost the agriculture based national economy and in particular, the socio-economic growth of the rural livestock holders of the country.

Awards, Plaques and Certificates were distributed to the various distinguished participants by the chief guest for their significant contribution on strengthening of veterinary services and livestock development of the country.

As concluding remarks, the chairman, Honourable Mr. Prithvi Raj Ligal, NPC vice chairman, stated the need of the veterinary services to the nation and was hopeful that the house would consider the national goal to achieve the poverty alleviation and make its valuable recommendations.

At last the vote of thanks was presented by Dr. Mahendra Nath Lohani, Vice President of NVA.

The conference was attended by 238 member veterinarians from all over the nation and some foreign participants from England, Japan and SAARC countries. A total of 37 scientific papers on various aspects of the profession were presented in seven different scientific sessions.

In a closed session, the general assembly elected the new executive committee for next two years' period. The executive body elected were as follows:

Dr. Nil Prakash Singh Karki	→	President
Dr. I. P. Dhakal	→	Vice President
Dr. N. P. Ghimire	→	General Secretary
Dr. K. P. Premy	→	Joint Secretary
Dr. Rajesh Yadav	→	Treasurer
Dr. R. R. Shrestha	→	Central Member
Dr. B. K. Dahal	→	Central Member
Dr. S. N. Deo	→	Regional Member (Eastern)
Dr. K. B. Kayastha	→	Regional Member (Central)
Dr. M. D. Lekhak	→	Regional Member (Western)
Dr. P. S. Kushwaha	→	Regional Member (M. Western)
Dr. H. C. Karki	→	Regional Member (F. Western)
Dr. S. N. Mahato	→	Editor in Chief

The closing ceremony was held on 24 February at Administrative Staff College hall, Jawalkhel.

INFECTION AND EFFECTS OF GASTROINTESTINAL NEMATODES IN GOATS RAISED UNDER THE SEDENTARY AND MIGRATORY MANAGERMENTS IN THE HILLS AND MOUNTAINS OF NEPAL

B.R. Joshi¹

ABSTRACT

A study was conducted for a period of one year on the epidemiology and clinical significance of gastrointestinal nematodes on the health and production of goats raised under the sedentary and migratory managements in the hills and mountains in Nepal. The findings of the study showed that the worm burden in the migratory goats was considerably higher than that in the sedentary goats throughout the year. However, this difference was not reflected on the faecal egg counts of the animals as this value was always lower in the migratory goats. The clinical significance of this sub-clinical infection was considerable in both systems as the growth rate in worm controlled kids was 160 and 93 percent higher than that of the untreated animals in the sedentary and migratory systems respectively. These findings are discussed and the approach for strategic control is suggested in the paper.

INTRODUCTION

Goat production is an important component of subsistence agriculture system in Nepal and the estimated 5.4 million goats in the country are raised under the sedentary or migratory managements throughout the country (LMP, 1993). The four main breeds of goats in the country, namely Chhyangra, Sinhal, Khari and Terai are raised respectively in the high mountainous, mountainous, hills and terai regions of the country. Goats are raised by all communities and classes of Nepalese society for their diverse use but principally for meat production, which is relished by all sectors of the society. The demand of goat meat in the country is ever increasing and to fulfil the daily needs of the urban population, there is a regular import of live animals from the neighbouring countries as it could not be fulfilled from the production within the country. However, despite the constant import of live animals, the supply is still well below the demand, causing the steady rise in the market price of goat meat.

The local estimated population of 5.4 million goats in the country contribute very little to the market needs of the country and is perhaps only able to supply to the necessities of the rural population. The production systems are traditional and the losses in the system are very high to affect the productivity of the system. Shrestha (1994) reported 27.5% mortality in kids and 5.4% in adult animals in the sedentary management with poor growth rate of the surviving kids. The productivity of Sinhal goats in the migratory system is not well studied but is not expected to be satisfactory due to various influencing factors.

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Among the various factors influencing goat productivity, infection of gastrointestinal nematodes is regarded as one of the important factor (Shrestha, 1994) and in some isolated studies the response to anthelmintic treatment has been reported to be encouraging (Shrestha *et al*, 1990, Kadariya and Joshi, 1994). However, these studies provided little information on disease epidemiology and long term production responses. Hence, the present studies were aimed to understand the infection and effects of gastrointestinal nematodes in the goat population raised under sedentary and migratory managements in the hills and mountains of the country.

MATERIAL AND METHODS

Study area

These studies were conducted in the sedentary flocks of Bharat Pokhari village of Kaski district located at an altitude of 850 metre above sea level (masl) and in a migratory flock of Ghandruk village of Kaski district located at an altitude of 2100 masl. The migratory flock moved from 5000 masl during the summer months to 1000 masl in the low valleys during the winter months.

Epidemiological Studies

Indirect measurement of parasite burden of the animals was evaluated by faecal egg counts of the adult and young animals. Faecal egg counts were carried out according to MAFF (1986) from the representative population of animals (adult and young) from both systems at monthly intervals. The number of faecal samples examined each month is presented in Table 1.

Table 1. Number of faecal samples examined each month

System	Age	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan
Migr.	Adu.	47	48	56	54	56	55	56	56	54	49	50	50
	Kids	16	33	29	23	29	29	10	14	21	37	25	16
Sed.	Adu.	60	44	53	49	50	47	51	50	52	51	51	49
	Kids	21	20	20	34	22	25	24	31	22	27	26	35

(Note: Migr.= Migratory, Sed.=Sedentary, Adu.= Adult)

The faecal egg count data were supplemented by monitoring the worm burdens in the adult animals during different seasons of the year, for which 11 adult goats from the migratory and 12 goats from the sedentary flocks were purchased and worm burdens were assessed according to the methods described in MAFF (1986). Species identification of the parasites was carried out according to Soulsby (1965) and the hypobiotic larvae according to Thomas and Probert (1993).

Production Response studies

The effects of gastrointestinal nematodes on productivity was evaluated by monitoring the weight gain and faecal egg counts of sub-sets of kids pair matched for body weight, age and sex. The treatment group was maintained worm free by regular anthelmintic treatment (ivermectin at 300 mcg/kg, s/c at monthly intervals in the migratory goats and fenbendazole 5 mg/kg, orally, fortnightly to the sedentary goats) and the weight gain responses was compared to that with the animals managed under farmers management (no treatment). The faecal egg counts and body weight were measured at monthly intervals for a period of one year in migratory system and for 6 months in the sedentary system. A total of 14 and 10 kids were monitored in the migratory and sedentary systems respectively.

RESULTS

Faecal Egg Counts

The mean faecal egg counts of adult and young animals during different months of the year in both system is presented in Figures 1 and 2.

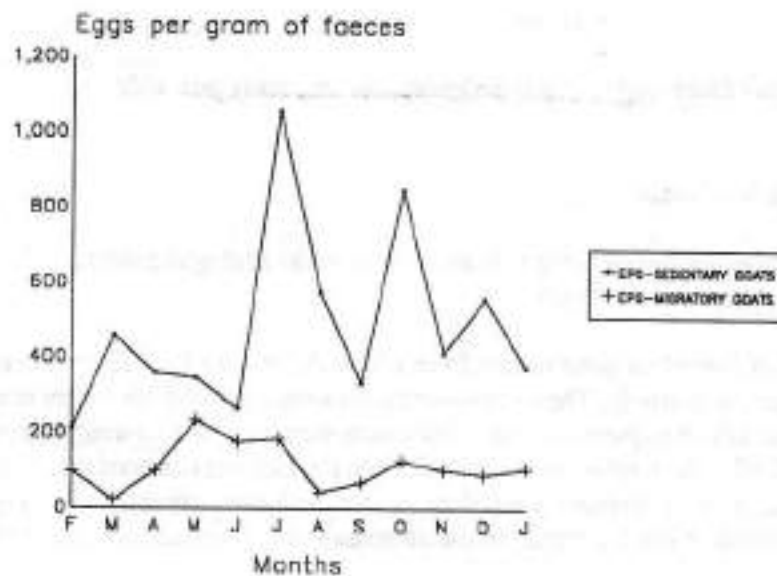


Figure 1. Mean faecal egg counts in sedentary and migratory adult goats

The mean faecal egg counts of sedentary adult goats was significantly higher than that of the migratory goats throughout the year with a peak of 1060 epg during the summer monsoon months. The egg counts of migratory adult goats on the other hand, remained at a low level during all the year, the highest value recorded being 235 epg during the month of May. The mean faecal egg count of kids followed a similar pattern to that of the adult goats. The faecal egg counts of sedentary kids increased after May, remained elevated until November and declined thereafter. The mean count of

the migratory kids showed a slight rise during May but was consistently low, never exceeding 200 epg. Thus an increase was observed in the egg counts of both adult goats and kids during the rainy season in the sedentary system, but no definite trend was recorded in the migratory system.

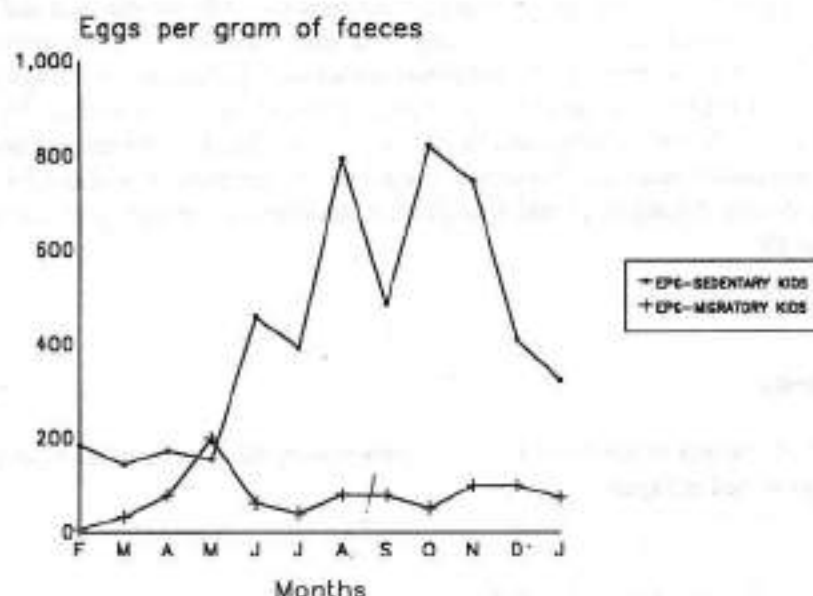


Figure 2. Mean faecal egg counts in sedentary and migratory goat kids

Worm Burden of adult Goats

The mean worm burden (including the hypobiotic larvae) in the adult goats during different seasons of the year is presented in Figure 3.

Total worm counts of individual goats ranged from 1,300-38,990 and 30-2,250 in the migratory and sedentary systems, respectively. The mean worm burdens were considerably higher in migratory than in sedentary animals throughout the year. Differences were significant during the autumn and spring seasons ($P < 0.05$). The proportion of the total worm population in arrested development was considerably higher in the migratory goats than in their sedentary counterparts. (Figure 3). Hypobiosis was recorded more frequently in the abomasal worm species and only rarely in the intestinal worms.

Seasonal changes in the worm composition are presented in Table 2.

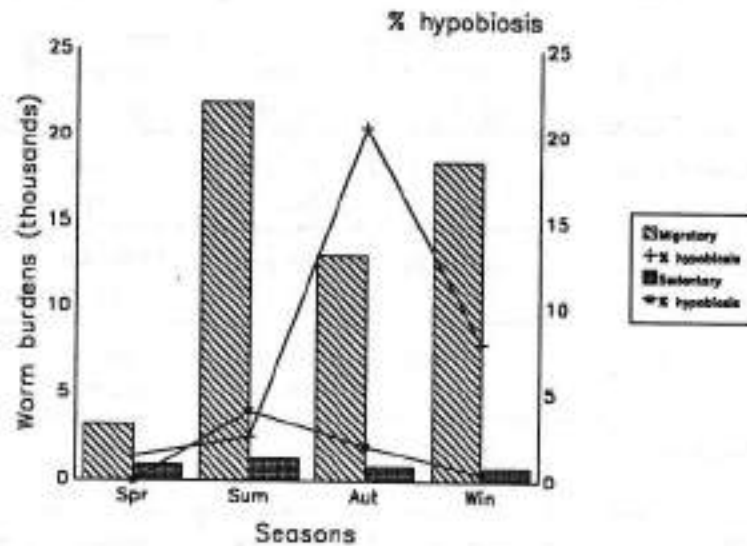


Figure 3. Mean worm burden and percent hypobiosis in adult goats in the migratory and sedentary systems

The mean worm burden in adult goats was higher in the migratory system than in the sedentary system in all four seasons. While the total worm burden of the sedentary goats rarely exceeded thousands, it was always higher than three thousands with the highest recorded burden of more than eighteen thousands in the migratory goats.

Seasonal changes in the worm composition were recorded in both management systems. *Ostertagia* was the predominant nematode genus present in the migratory system followed by *Trichostrongylus* spp with a low proportion of *H. contortus*. A significant increase in *Ostertagia* spp and a reduction in *Trichostrongylus* spp was recorded during the autumn season. In the sedentary system, however, the predominant genus was *Trichostrongylus* except during the summer season, when the highest proportion was comprised of *H. contortus*.

Effects on Production

The nematode control regime adopted in these studies did not always protect the animals completely. It was particularly true for the monthly ivermectin treatment during the wet summer months for the migratory animals. Whereas, during the dry winter months, faecal egg counts of the treated animals were always zero. Similarly, faecal egg counts were always zero in the groups treated with fenbendazole at fortnightly intervals. The faecal egg counts of the animals in control group in all studies, remained significantly higher than that of treated animals but fluctuated according to the seasons.

Table 2. Seasonal changes in worm composition of parasite species in adult goats.

Systems	Nematode genera	Spring (M-M)	Summer (J-A)	Autumn (S-N)	Winter (D-F)
Migratory	<i>Haemonchus</i>	268±174 (8.2)	240±230 (2.0)	270±150 (2.0)	200±100 (1.0)
	<i>Ostertagia</i> *	1300±376 (39.7)	5480±1940 (45.7)	11230±530 (86.4)	7275±1835 (39.2)
	<i>Trichostrongylus</i>	1383±883 (42.2)	5630±566 (46.9)	1390±750 (10.7)	2210±1210 (11.9)
	<i>Bunostomum</i>	25±25 (0.8)	0	0	50±50 (0.3)
	Others**	298±108 (9.1)	640±440 (5.3)	100±0 (0.7)	8800±8800 (47.4)
	Mean Total	3274	11990	12990	18550
Sedentary	<i>Haemonchus</i>	78±50 (8.9)	640±276 (51.7)	70±50 (8.7)	0
	<i>Ostertagia</i> *	145±53 (16.6)	15±7 (1.2)	60±40 (7.4)	0
	<i>Trichostrongylus</i>	250±93 (28.6)	480±168 (38.8)	425±275 (52.8)	495±375 (69.7)
	<i>Bunostomum</i>	0	0	50±50 (6.2)	55±55 (7.7)
	Others**	400±400 (45.7)	102±38 (8.2)	200±100 (24.8)	160±160 (22.5)
	Mean Total	873	1237	805	710

(Note:*= *Ostertagia* includes both *Ostertagia* and *Teladorsagia* species together, Others**= *Oesophagostomum*, *Cooperia*, *Strongyloides*, *Trichuris*, *Chabertia*; Figures in parenthesis represents percentage of the species in the population.

In both systems, the weight gain in parasite controlled animals was significantly higher than in the managed under farmer's management (untreated). The weight gain varied in both systems. In the migratory kids, at the end of 350 day experimental period, the cumulative weight gain in treated kids was 4.0 kg higher than the untreated animals. This difference was significant statistically ($P<0.05$). The mean weight gain per day in treated kids was 26gm/day as compared to 14.3gm/day

in control kids. While, in the sedentary system, the mean total body weight gain in the treated goat kids was 8.9 kg as compared to 3.7 kg in control animals during the study period of 6 months. The difference in weight gain is highly significant statistically ($P < 0.005$). During the study period, mean daily weight gain of treated kids was 51.4 gm/day as compared to 23 gm/day in the control animals. The summary of the weight gain responses in the treated kids in both systems is presented as Table 3.

Table 3. Body weight gain in migratory and sedentary goat kids following gastrointestinal nematode control.

System	Groups	Total weight gain (kg)	% weight gain on initial body weight	Weight gain per day (gm/day)
Migratory	Treated	9.1	56.4	26
	Control	5.0	29.2	14.3
Sedentary	Treated	9.0	60.5	51.4
	Control	3.7	23.2	23.0

Despite the fact that, anthelmintic treatments did not protect the animals completely, the body weight gain of the treated groups managed in both systems was significantly higher than that managed under farmers traditional management (untreated). It was also noticed that body weight gain in the treated animals was higher while the nutritional status of the animals was better.

DISCUSSION

Although, the browsing habit of goats reduces their intake of infective larvae, the higher worm burden in the migratory goats than in the sedentary animals reflects the grazing environment of the animals in both systems as the absence of browsing vegetation above certain altitudes forced the animals to graze. Consequently, their worm burdens were considerably higher than the sedentary goats. However, this difference was not reflected in the faecal egg counts of the animals, possibly because, the worm species (*O. nianquingtanggulaensis*) recorded in high numbers in the migratory animals might have low fecundity than the worms recorded in the sedentary goats. This speculation however, needs further investigation. The lower egg counts and relatively lower body weight loss in migratory animals than the sedentary goats, however also suggests the possibility that the better nutritional status of the migratory goats might have resulted the higher immunological status in these animals, which would reduce the fecundity of the parasites and their effects on the animals. These findings however, support the view that the faecal egg counts are poor indicator of the worm burdens in the animals. The faecal egg counts of the animals in both systems was generally low, never exceeding 200 epg in the migratory and 800 epg in the sedentary animals. Nevertheless, despite the low faecal egg counts, the response to anthelmintic treatment was highly significant in both systems. Hence, critical level of faecal egg counts in goats should be much lower than that considered for sheep. This is particularly true if the decisions on control programmes are based on

faecal egg counts. It further stresses the fact that goats are more susceptible to gastrointestinal nematode infections as suggested by Rickard (1990).

The species composition of the worm burdens in both systems varied considerably. While, *Trichostrongylus* spp was the predominant worm species in the sedentary system, it was *Ostertagia* spp. (mostly *O. nianquingtangulaensis*), which was recorded in high numbers in the migratory goats. This parasite has not been reported previously outside of Tibet (K'ung and Li, 1965). The proportion of this parasite increased while the animals were at the alpine pastures and reached to 82% of the total worm burdens during the month of August (Joshi *et al*, 1997). This parasite is smaller and slender than other *Ostertagia* or *Teladorsagia* spp. commonly found in small ruminants. Nothing is known about the pathogenicity of this species and further studies are needed to understand this aspect. The abomasal worms showed a much greater propensity for hypobiosis at higher altitudes. This may again be related to the greater proportion of *Ostertagia* in the worm population and to the exposure of the free living developmental stages of the parasite to the cold climate at the higher altitudes. In contrast, the selection pressure for hypobiosis on the *Haemonchus contortus* population at the lower altitudes would not be so strong as temperature is not a limiting factor and there is generally sufficient moisture.

The epidemiological studies show that, the main period for pasture larval challenge are the wet spring and summer months, from April/May to September/October. In the migratory system, this is the period for upward migration and grazing at the alpine pastures. Survival of infective larvae at the alpine pastures has been reported from the Swiss Alps (Eckert and Hertzberg, 1994).

These studies confirm that parasitic gastroenteritis is the cause of considerable loss in goat production in Nepal. This is true of animals reared under both the sedentary and migratory management systems as the control of nematode parasites resulted substantial improvements in body weight gain over pair matched animals reared under farmer's managements. This difference was about 93% in migratory to 160% in sedentary goats. These estimates of sub-clinical losses are probably conservative as it was apparent that the full benefit of parasite control could only be expressed in animals on an adequate plane of nutrition.

It is also important to note that although the level of infection was sub-clinical, and the animals were under sub-optimal nutritional status during some periods of the year, the body weight gain was significantly higher in treated kids in both management systems. Thus it can be anticipated that the responses to nematode control would be considerably higher if the nutrition of the animals could be improved during lean seasons. This aspect needs further investigation in the field conditions.

Possible Control Strategy

Considering the undefined and uncontrollable grazing areas and the seasonal pattern of infection, the possible strategy for the control of gastrointestinal nematodes in the goat population in the hills and mountains of Nepal would be the rational and strategic application of anthelmintics. The most important aspect of this strategy would be to prevent the animals from the high level of infection during the wet summer months (June-September). From October onward, it is expected that the low level of infection from the pasture would not affect the productivity of the animals. It will be also important to minimise the use of anthelmintics not only from the economic point of view but also

to avoid the development of anthelmintic resistance. Under these circumstances, the nematode control programmes would only be significant, if the nutritional status of the animals is also improved simultaneously. Anthelmintic application could thus be incorporated in a feed based or mineral block based formulation.

No cost benefit analysis were carried out for the control strategies suggested in the present study, hence the potential benefits of one or the other schedule can not be estimated. It is therefore, suggested that a cost benefit analysis for the different options should be evaluated under the farmer's conditions so that the strategy with highest economic return could be adopted. More over any technological possibility would need to be evaluated with the prevailing economics of the farming conditions and the potential return from the investment.

ACKNOWLEDGEMENTS

The cooperation obtained from the farmers and shepherds for this study is gratefully acknowledged. The assistance of the staff members of Veterinary Investigation Laboratory of Agriculture Research Station, Lumle for laboratory and field investigations is also highly acknowledged. The financial support of DFID (ODA, UK) for the study is also gratefully acknowledged.

REFERENCES

- Eckert, J. and Hertzberg, H. (1994). Parasite control in transhumant situations. *Veterinary Parasitology*, **54**: 103-125.
- Joshi, B. R., Gibbons, L.M. and Jacobs, D. E. (1997). *Ostertagia nianquingtangulaensis* K'ung & Li, 1965 (Nematoda: Trichostrongyloidea) from sheep and goats at high altitudes in Nepal. *Journal of Helminthology*, **71**: 21-27.
- Kadariya, R.K. and Joshi, B. R. (1994). Effect of strategic anthelmintic drenching, mineral supplementation and concentrate feeding on the performance of Nepalese (Khari) hill goats. Lumle Agricultural Research Centre Working paper no. 94/14.
- K'ung, Fan-yao and Li, Chien-fu, (1965). *Ostertagia nianquingtangulaensis* n. sp. (Nematoda: Trichostrongylidae) from domestic sheep in Tibet. *Acta Veterinaria et Zootechnica Sinica*, **8** (3): 213-216 (In Chinese, abstract in English).
- LMP (1993). Livestock Master Plan, Vol. I, II, III. Asian Development Bank, ANZDEC Limited, Agriculture Project Service Centre.
- MAFF (1986). Ministry of Agriculture, Fisheries and Food, Manual of Veterinary Parasitological Techniques, Reference Book no 418.

- Rickard, J.M. (1990). Current research on anthelmintics in goats. *Goat Health and Production. Proceedings of refresher course for Veterinarian*. no 134, 11-15 June, 1990, University of Sydney, 287-299.
- Shrestha, H.R. (1994). Final Technical Report, Goat Production System, Nepal. Nepal Agriculture Research Council and International Development Research Centre, Canada.
- Shrestha, N.P., Neupane, S.P. and Gurung, H.B. (1990). Effect of anthelmintic treatment and feeding regimes on growth rate of local goats. Pakhribas Agriculture Centre, Technical Paper no 127, Pakhribas, Dhankuta Nepal, 1-7.
- Soulsby, E.J.L. (1965). *Text book of Veterinary Clinical Parasitology. 1. Helminths*. Blackwell Scientific Publications, Oxford.
- Thomas, D.Rh. and Probert, A.J. (1993). A key to the identification of arrested gastrointestinal nematode larvae of sheep in Britain. *Veterinary Parasitology*, 47: 77-80.

MANAGEMENT OF VIRULENT FOOTROT IN NEPAL

S.C. Ghimire¹

ABSTRACT

Virulent footrot introduced into the country in the 1960s was endemic in migratory flocks of Kaski, Lamjung and Manang districts of Gandaki zone. Continuous efforts to control and possibly eradicate the disease from this area prevented its further spread, and controlled the severity and prevalence of the disease in endemic flocks. The disease, however, proved very difficult to be eradicated from some flocks due to its continuous recurrence during migration. To overcome management complications associated with annual migration, *Dichelobacter nodosus*, the essential transmitting agent of footrot, involved in these outbreaks were extensively isolated and characterised. Only isolates of two serogroups i.e. serogroup B and E, were identified to be associated with virulent footrot in these flocks. As these isolates were different from the prototypes used in vaccine production, and as the multivalent footrot vaccines are less potent than the mono or bivalent footrot vaccines, specific bivalent vaccine against footrot in Nepal was produced and used to control footrot in these flocks. Specific bivalent vaccine provided protective immunity for more than six months spent on migration. Use of this vaccine together with other management plans aimed at eradicating virulent footrot from this area has resulted in disappearance of virulent footrot in these flocks since 1994. Results of extensive clinical and serological surveillance of the disease so far suggest the elimination of virulent footrot from Nepal. Disease surveillance activities are still ongoing.

INTRODUCTION

Footrot is a contagious disease of the feet of ruminants which causes lameness. It is a mixed bacterial infection of which *Dichelobacter nodosus* is the essential transmitting agent (Beveridge, 1941). Two forms of the disease, virulent and benign, are generally recognised, though a third, intermediate form, of the disease is often described (Egerton and Parsonson, 1969). Virulent form of the disease is regarded as one of the economically important diseases of sheep as it causes a significant economic loss to the sheep industry. Virulent footrot is therefore subjected to control and eradication programme (Egerton and Raadsma, 1991). Being a mild and transient disease, benign footrot does not warrant institution of a disease control programme (Egerton and Raadsma, 1991).

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Conventionally, control of footrot is either achieved by prophylactic vaccination or by regular footbathing (5-10% formalin or 10% zinc sulphate) during disease transmission season. Eradication can be achieved by one of the three methods, namely, a. depopulation and restocking with disease free animals, b. identification and culling of all affected animals, and c. identification and treatment of all affected animals and culling of the non-responders (Egerton, 1986). Adoption of one method or the other depends on several factors including financial and practical aspects of disease management. However, one of the prerequisites for disease eradication is a low prevalence of the disease before it is attempted. In other word, effective control is required before disease eradication.

Virulent footrot is suspected to have been introduced into Nepal through imported rams in the nineteen sixties, and has become endemic in parts of Kaski, Lamjung and Manang districts of Gandaki zone (Karki, 1983). The disease proved to be very devastating in these flocks. Before the disease was diagnosed and control programmes instituted, two thirds of sheep in footrot affected flocks were reported to be lame, of which one third were walking on their knees (Flatt, 1971, cited by Karki, 1983). The mortality in acutely affected flocks was 52% higher than that observed in disease free flocks within a period of two years even though all flocks were managed under similar husbandry practices (Karki, 1994). The disease therefore not only threatened the continuity of migratory husbandry, but also posed a serious threat to the hill agriculture as a whole. Because of its adverse effects to the hill agriculture, institutional control programmes were implemented since 1975.

NEPALESE FOOTROT CONTROL PROGRAMME

The initial phase (1975-1978):

Initial footrot control programme was implemented jointly by the Lumle Agricultural Research Centre (LARC) and Sheep and Wool development Project of His Majesty's Government of Nepal (HMGN). The disease was then prevalent only in parts of Kaski and Lamjung District. LARC was responsible for controlling the disease in Kaski whereas HMGN was responsible for implementing the programme in Lamjung district. The method used for treatment was paring and footbathing of affected animals in 10% formalin during the dry winter months when migratory flocks were easily accessible. This joint venture continued until 1978, when Sheep and Wool Development Project was winded up and HMGN's direct involvement was suspended. This control programme reduced the severity of the infection in affected flocks, but was not able to eliminate the infection from the affected flocks. This programme, however, was not able to prevent the spread of the infection to disease free areas, and by 1979 the disease spread further affecting Manang flocks and several flocks of Lamjung east of Marsyangdi river.

The second phase (1978-1992):

Between 1978 and 1992, LARC was solely responsible for implementing the programme. HMGN provided support to the programme by seconding manpower and indirect support through facilitating programme implementation by administrative and moral support. Extensive surveys of adjoining districts were conducted to determine the extent of the disease spread. By 1979, it was evident that footrot had not spread west of Seti river, the

western boundary. However, it did spread eastward from the previously defined eastern boundary, the Marsyangdi river, affecting flocks from east Lamjung and Manang (Foulds, 1979). However flocks of east Lamjung east of Dordi river and those of Gorkha were not affected (Karki et al., 1982). The Dordi river was therefore identified as the eastern boundary. Thus the footrot area encompassed approximately 250 migratory flocks from parts of Kaski, Lamjung and Manang representing approximately 30000 sheep and goats between the Seti river in the west and the Dordi river in the east (Fig. 1).

By early eighties it was evident that treatment of affected animals during the dry winter months alone was neither going to reduce the flock prevalence nor help to achieve the goal of eliminating the infection from the area. A focus of infection would always pose the risk of further spread of the disease to other parts of the country, and had the potential devastate the country's small ruminant husbandry. Intensive disease management programme was therefore formulated and implemented with the ultimate aim of eradicating the disease from the country. The approach adopted was intensive treatment of affected animals (footbathing and antibiotics) during the dry winter period and culling of non-responders before next migration. Farmers received partial compensation for animals culled. Efforts were also made to reduce the disease prevalence by introducing footbathing practices during the disease transmission period of migration, but were not effective due to practical constraints. Protection of healthy animals during the risk period of migration by vaccination was attempted. Two types of vaccines were used, initially a commercial bivalent vaccine and later a multivalent (8 serogroups) commercial vaccine. However, none of the vaccine provided sufficient protection.

The programme implemented between 1978 and 1992 reduced the severity of infection in affected flocks, prevented the further spread of the disease to new areas and eradicated the virulent disease from most of the flocks. However, the disease consistently recurred in a small number of flocks. Inability to identify and cull all infected animals (including subclinical infections) before migration and to protect healthy animals while on migration were the major obstacles for the success of the programme. Frequent mixing of several flocks during migration meant that the disease that recurred in some of the flocks spread to many other flocks before they returned to the villages and were amenable to inspection and treatment. This often nullified the success achieved in the previous seasons. Furthermore, this always left a focus of infection from where the disease could easily spread to other parts should any relaxation be made on the control programme.

The third phase (1992 to date):

Due to the limitations of conventional control programmes to prevent the recurrence of virulent footrot during the risk period of migration, and to protect healthy animals from being reinfected either by footbathing or vaccination using commercial multivalent vaccines, alternate approaches of footrot management were considered. Limitation of conventional footrot vaccine is the short duration of immunity which probably was not enough to protect animals for the six months spent on migration. In contrast, mono or bivalent vaccines provide a much longer duration of immunity (O'meara et al., 1993). For this reason, the possibility of using specific vaccination for management of footrot in these flocks was explored. This investigation was jointly implemented by LARC and the University of

Sydney, Australia since 1992 with the financial assistance of the Australian Centre for International Agricultural Research (ACIAR).

Basis for specific vaccine and vaccination:

Extensive isolation and characterisation of *Dichelobacter nodosus*, the essential transmitting agent of footrot, from migratory flocks revealed the presence of virulent isolates of only two serogroups, i.e. B and E (Ghimire et al., 1996). Detailed antigenic characterisation of these isolates indicated that Nepalese isolates were not identical with the serogroup prototypes used for commercial vaccine production. It was therefore decided to formulate bivalent vaccine using the local antigens. Initially, conventional bacterin vaccine was formulated which was replaced later by recombinant DNA fimbrial vaccine. This vaccine was used in migratory flocks of Nepal. In addition to prophylactic usage, this vaccine was also used for its curative action in affected animals. Animals treated by immunisation and recovered after treatment were allowed to migrate together with other animals, but regularly received booster immunisation before next migration.

To provide the maximum protection, prophylactic vaccination was timed just before the risk period of disease transmission. Field investigations indicated that the most risky period of footrot transmission in Nepal was during the upward migration through the forest, i.e. between May and June (Ghimire and Egerton, 1906). Though to a limited extent, footrot also transmitted during the descending migration i.e. between September and October (Ghimire, 1997a). Prophylactic vaccination was done in April, just before the flocks left the villages for alpine pasture, which was also practically convenient.

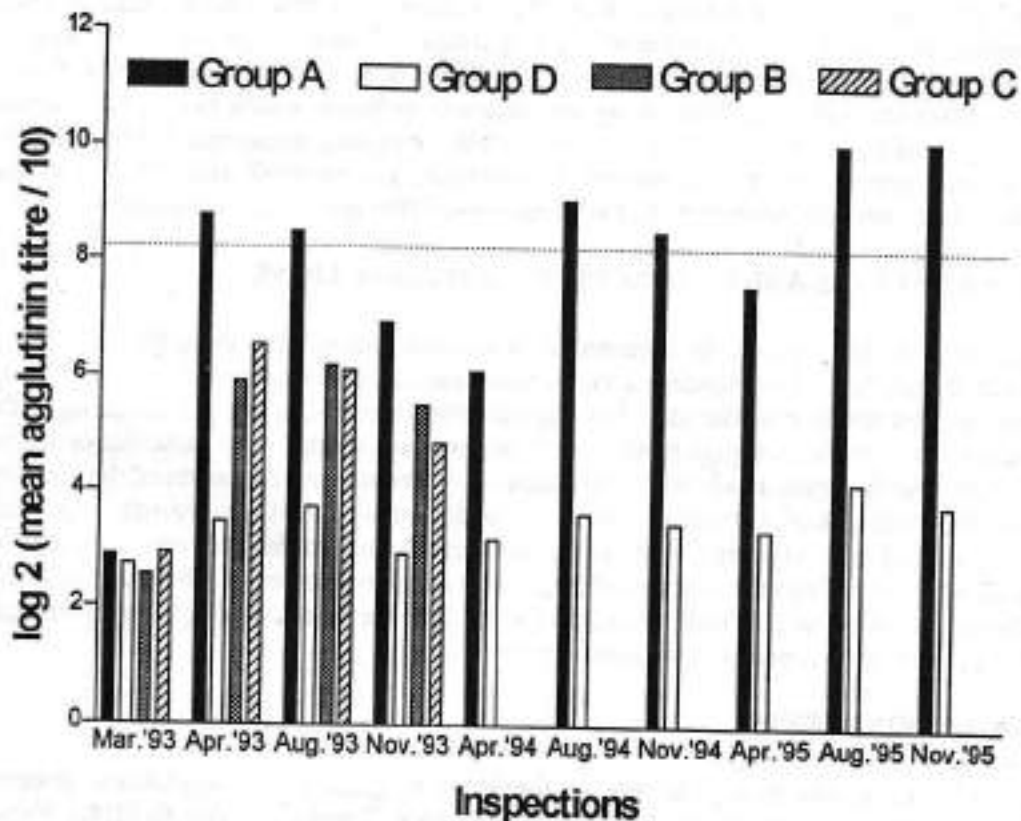
Integration of specific vaccination to field disease management programme:

The evaluation and use of specific vaccine was added to a footrot management system which consisted of inspection of all sheep and goats several times during the winter months (November-April). At these inspections animals affected with footrot were identified and treated with antibiotics and the specific vaccine. All those animals which failed to respond to the treatment were culled before the flocks went on migration i.e. before April. The aim of this rigorous inspection, treatment and culling was to ensure that only apparently disease free animals should migrate. Animals in the vaccination group received the booster dose of specific vaccine in April.

Virulent footrot did recur in some animals in the alpine pastures in the first year of vaccination. However, rigorous inspection programme for identification and treatment (antibiotics and specific vaccine) of affected animals together with culling of non-responders in subsequent inspection, and prophylactic vaccination before next migration was successful in preventing the recurrence of the disease in the following years.

No virulent footrot has been observed since the second year of the vaccination programme. Virulent isolates of serogroup B and E have not been isolated either, despite the intensive clinical and bacteriological follow up. What has been observed regularly in each migration is a low prevalence of benign footrot in both the vaccinated and unvaccinated flocks. It seems likely that animals which have been immunised annually against the two virulent

serogroups are so resistant to infection that no carriers among them have relapsed. Serological studies of representative animals have also indicated that animals vaccinated



with specific vaccine have maintained the group mean serum antibody titres above that considered protective (Thorley and Egerton, 1981) for more than six months (Fig. 2). The failure of unvaccinated animals to acquire or manifest virulent footrot may be due to the elimination of subclinical infection from that population of animals because of the low prevalence of disease occurring in Group D in the first two years of the investigation.

Figure 2. Serum agglutinin titre against serogroup E antigens in sheep of different vaccination groups during various inspections between March 1993 and November 1995 (Source: Ghimire, 1997b). The dotted line indicates the group mean agglutinin level considered being protective in sheep (Thorley and Egerton, 1991)

Note: Vaccination regime: Group A - Specific vaccine, Group B - Multivalent and Specific vaccine, Group C - Multivalent vaccine, and Group D - Nil.

Withdrawal and vaccination and intensive clinical and serological surveillance:

Virulent footrot was not recorded in any flock continuously for two migrations i.e. 1994 and 1995. To ensure that the absence of the disease was due to elimination of virulent strains of

D. nodosus from these flocks rather than due to suppression of their expression in vaccinated animals due to a strong vaccinal immunity, vaccination was withdrawn from these flocks. Initially in 1996, vaccination was withdrawn from 15% of vaccinated flocks. However, there was no outbreak of virulent footrot in those flocks during that year. As a result, in 1997, vaccination was withdrawn from the whole population. Extensive clinical monitoring of these flocks has so far failed to reveal the reoccurrence of virulent disease in those flocks. Samples from any suspected foot lesions are regularly cultured, which has so far revealed only benign isolates, if any. Furthermore, serological monitoring by anamnestic challenge of unvaccinated animals for two consecutive migrations, i.e. in 1997 and 1998, had also indicated the absence of virulent footrot in these flocks (Dhungyel et al., submitted).

CURRENT STATUS AND FUTURE RESEARCH DIRECTIONS

The evidence so far supports the elimination of virulent footrot from these flocks, and has provided the evidence that specific immunisation assists elimination of the strains of *D. nodosus* against which it is directed. Disease surveillance activities are still on going in the endemic areas. However, benign isolates of *D. nodosus* are isolated from these flocks almost every year. Furthermore, to elucidate the status of the disease in other parts of the country, clinical and bacteriological surveys are being conducted by LARC in districts other than Kaski, Lamjung and Manang. Limited surveys conducted so far had not revealed the presence of virulent footrot in any part of Nepal. An extensive survey of other parts of Nepal is required to collect scientifically valid data to declare the whole country free of virulent footrot and should be considered for future research.

ACKNOWLEDGMENTS

The author would like to express sincere thanks to all persons and institutions involved directly or indirectly over the years in this programme. Thanks are due to HMG Nepal, ACIAR, Australian Government and DFID, British Government for their support in Nepalese footrot management programme, and to Prof. J.R. Egerton, University of Sydney, Australia for his idea on specific vaccination and support to the research programme.

REFERENCES

- Beveridge, W.I.B. (1941). *Foot-rot in Sheep: a Transmissible Disease due to Infection with Fusiformis nodosus (n.sp.). Studies on its Cause, Epidemiology and Control*. Council for Scientific and Industrial Research, Melbourne, Bulletin No. 140.
- Dhungyel, O.P.; Ghimire, S.C.; Whittington, R.J. and Egerton, J.R. Confirmation of the specificity and sensitivity of fimbrial antigen for diagnosis of ovine virulent footrot by ELISA. Submitted to *Veterinary Microbiology*.
- Egerton, J.R. (1986). Control and eradication of ovine footrot. In: Stewart, D.J.; Peterson, J.E.; McKern, N.M. and Emery, D.L. (Eds.), *Footrot in Ruminants. Proceedings of a Workshop, Melbourne 1985*. CSIRO Division of Animal Health and Australian Wool Corporation, Glebe, Sydney, pp. 35-42.

- Egerton, J.R. and Parsonson, I.M. (1969). Benign foot-rot - a specific interdigital dermatitis of sheep associated with infection by less proteolytic strains of *Fusiformis nodosus*. *Australian Veterinary Journal*, **45**: 345-349.
- Egerton, J.R. and Raadsma, H.W. (1991). Breeding sheep for resistance to footrot. In: Owen, J.B. and Axford, A.F.E. (Eds.) *Breeding for Disease Resistance in Farm Animals*. CAB International, Oxon, pp. 347-370.
- Foulds, A.B. (1979). Footrot campaign report, Apr/May 1979. Lumle Agricultural Centre, P.O.Box 1, Pokhara, Nepal.
- Ghimire, S.C. (1997a). Footrot transmission in migratory flocks of western Nepal during descending migration through the forest. *Bull. Vet. Sci. Anim.Husb., Nepal* **25**: 38-45.
- Ghimire, S.C. (1997b). *Footrot in Nepal - Management by specific vaccination*. Ph.D. Thesis. The University of Sydney, Australia.
- Ghimire, S.C. and Egerton, J.R. (1996). Transmission of footrot in migratory sheep and goats of Nepal. *Small Ruminant Research*, **22**: 231-240.
- Ghimire, S.C.; Egerton, J.R. and Dhungyel, O.P. (1996). Characterisation of *Dichelobacter nodosus* isolated from footrot in sheep and goats in Nepal. *Small Ruminant Research*, **23**: 59-67.
- Karki, N.P.S. (1983). Report on eradication of footrot disease at Lumle Agricultural Centre. *Bulletin of Veterinary Science and Animal Husbandry, Nepal*, **12**: 42-52.
- Karki, N.P.S. (1994). Footrot eradication programme - problems and achievements in Nepal. In: Rasali, D.P. (Ed.), *Proceedings of the seminar on footrot eradication programme in sheep and goats in Nepal. Occasional Paper No. 94/1*. Lumle Agricultural Research Centre, P.O. Box 1, Pokhara, Nepal, pp. 7-12.
- Karki, N.P.S.; Parajuli, B.; Adhikari, D.R.; Gurung, I.B. and Gurung, C.B. (1982). Report on footrot survey in Gorkha district, 29 Sep-20 Oct, 1982. Lumle Agricultural Centre, P.O.Box 1, Pokhara, Nepal, pp 1-4.
- O'mcara, T.J.; Egerton, J.R. and Raadsma, H.W. (1993). Recombinant vaccines against ovine footrot. *Immunology and Cell Biology*, **71**: 473-488.
- Thorley, C.M. and Egerton, J.R. (1981). Comparison of alum-absorbed or non-alum-absorbed oil emulsion vaccines containing either pilate or non-pilate *Bacteroides nodosus* cells in inducing and maintaining resistance of sheep to experimental foot rot. *Research in Veterinary Science*, **30**: 32-37.

NATIONAL STRATEGY FOR FOOT AND MOUTH DISEASE CONTROL AND ERADICATION IN NEPAL

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ABSTRACT

Foot and Mouth Disease (FMD) is endemic in Nepal and is of significant economic importance in the agricultural production system of the country. The disease is a major constraint on effort designed to upgrade the quality of livestock through the introduction of high yielding improved breed of animal. Out of 75 districts, serotypes of FMD virus has been identified in 61 districts of Nepal. Of the seven standard serotypes, O, A, C and Asia-1 have been isolated. The socio-economic situation in the country is not conducive to the adoption of a slaughter policy in the control of FMD. Vaccination is likely to be the cornerstone around which any future policy will be built.

The epidemiology of FMD and national strategies for FMD control and eradication are outlined. Zonal concept for FMD vaccination campaign and efficient disease diagnostic services are discussed in details.

INTRODUCTION

Livestock plays an important and integral role in the agricultural farming system of Nepal. Livestock population of Nepal in relation to arable land and human population is large by Asian standards, but livestock productivity is low. Low livestock productivity is attributable mainly to wide range of endemic livestock diseases where FMD accounts to be the top most. Other factors include poor nutrition, low genetic potential and traditional management systems. There has been some development towards commercialisation of livestock production, but subsistence level of production still prevails in majority of rural areas. Cattle and buffaloes provide the majority of the draught power for cultivation and harvesting and the main method of transportation in rural and sub-urban areas. Buffaloes are particularly important to the people of Nepal because they are kept for multiple use, i.e. milk, meat, and manure production, draft power and bio-fuel. Live animals are imported as well as exported in large numbers from neighbouring countries and *vice-versa*.

In recent years, there is a sharp increase in the number of crossbred cattle population as a result of continuous efforts of Department of Livestock Services to improve productivity of local cattle through cross breeding and extension activities and commercialisation of dairy farming. The improved breed of cattle and buffaloes are highly susceptible to a devastating disease like FMD, which may produce fatal outcome and heavy economic losses to the

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farmers. The presence of FMD is a major constraint on effort designed to upgrade the quality of livestock through the introduction of high yielding, improved breed of animal.

ECONOMIC IMPORTANCE

The economic importance of FMD due to the loss of productivity following infection is enormous. Infection in draft cattle causes a serious problem in tillage on which the preparation of agricultural land is fully dependent. It virtually paralyses the cultivation work and thus causes a great shortfall in agricultural output. In dairy cows sharp drop in milk production, abortion and chronic mastitis is very common. In suckling calves the mortality rate is often very high. Nepal is trying to get World Trade Organisation membership in the near future and the prevalence of FMD could be a major barrier. Various study reports clearly indicated that FMD is the major livestock disease that causes enormous economic losses in agricultural production system.

EPIDEMIOLOGY OF FMD

FMD is endemic in Nepal since time immemorial. Epidemics of FMD have been reported from time to time in cattle, buffaloes, sheep, goats and pigs. It is reported in all parts of the country irrespective of altitude and climatic variation. Although the outbreak of FMD is reported all the year round, high incidence of FMD is noticed during monsoon and post-monsoon period. The reason could be extensive movement of animals for agricultural activities during the rainy season and unrestricted importation of large number of food animals across the border during religious festivals in October and November. In fact, movement of livestock takes place across all the borders of Nepal. It is seasonal in the north whereas it is all the year round in the south. Most probably, the single most important factor in the continued and high frequency of occurrence of FMD in Nepal are the movement of livestock. Constant movement of livestock across national frontiers provides the mechanism of introduction of different serotypes of viruses into the country.

The laboratory based FMD diagnosis in Nepal started in 1965 when specimens from suspected cases of FMD were sent to the World Reference Laboratory (WRL) for confirmatory diagnosis. Serotyping of FMD virus is done on regular basis in the National FMD Laboratory since 1983. The detailed record of FMD virus serotyping done so far is presented in Table 1. No virus was detected in almost 60% of the samples submitted. Of the seven standard serotypes, O, A, C and Asia-1 have been isolated. In positive samples, serotype O was predominant (69.7%) followed by Asia-1 (18.0%), serotype A (8.7%) and serotype C (3.6%) (Table 1). Serotype O was identified for the first time in 1965, serotype A in 1967, serotype Asia-1 in 1984 and serotype C in 1990.

Serotype O is responsible for regular outbreak of FMD in Nepal (Table 1). Serotypes A and Asia-1 were isolated regularly but less frequently than that of O. Subtype A₂₂ was recorded in Nepal prior to its detection in India in the early seventies (Ferris et al., 1992). Serotype C was recorded between 1990 and 1996. Out of 75 districts of Nepal, FMD virus had been isolated and serotyped from 61 districts (Fig. 1). FMD virus typing has not been done from Taplejung, Panchthar, Saptari, Solukhumbu, Khotang, Manang, Myagdi, Dolpa, Rukum, Jajarkot, Kalikot, Dailekh, Baitadi and Bajura districts of Nepal. The distribution of

serotypes of FMD virus in Nepal during the period of 1983 to 1998 is presented in Figure 1. The results of FMD virus serotyping in the FMD laboratory of Nepal and the World Reference Laboratory of United Kingdom over the past years demonstrate that serotypes O and Asia-1 are major dominant serotypes of FMD virus found in this country. Serotype O was found to be the major serotype responsible for epidemic outbreak of FMD in the year 1997/98.

Table 1. Serotypes of FMD Virus identified in Nepal, 1965 – 1999

Year of collection	Number of samples	Serotypes					No Virus Detected
		O	A	Asia-1	C	Mixed*	
1965	2	2	0	0	0	0	0
1966	3	3	0	0	0	0	0
1967	5	0	4	0	0	0	1
1968	3	1	1	0	0	0	1
1975	6	3	0	0	0	0	3
1978	1	1	0	0	0	0	0
1983	7	5	1	0	0	0	1
1984	40	12	8	7	0	0	13
1985	68	37	0	7	0	0	24
1986	31	10	3	0	0	0	18
1987	54	19	0	6	0	0	29
1988	113	25	5	6	0	0	77
1989	53	13	3	8	0	0	29
1990	80	41	0	2	1	1	36
1991	10	5	0	0	0	0	5
1992	5	2	0	0	1	0	2
1993	74	8	0	0	9	0	57
1994	64	9	0	8	1	0	46
1995	51	3	0	7	0	0	41
1996	100	30	4	0	1	0	65
1997	100	9	0	13	0	0	78
1998	18	11	2	0	0	0	5
1999	12	7	0	0	0	0	5
Total	900	256	31	64	13	1	536

*O+C

NVD (%) = 59.8

Source: Institute of Animal Health, World Reference Laboratory, Pirbright, England

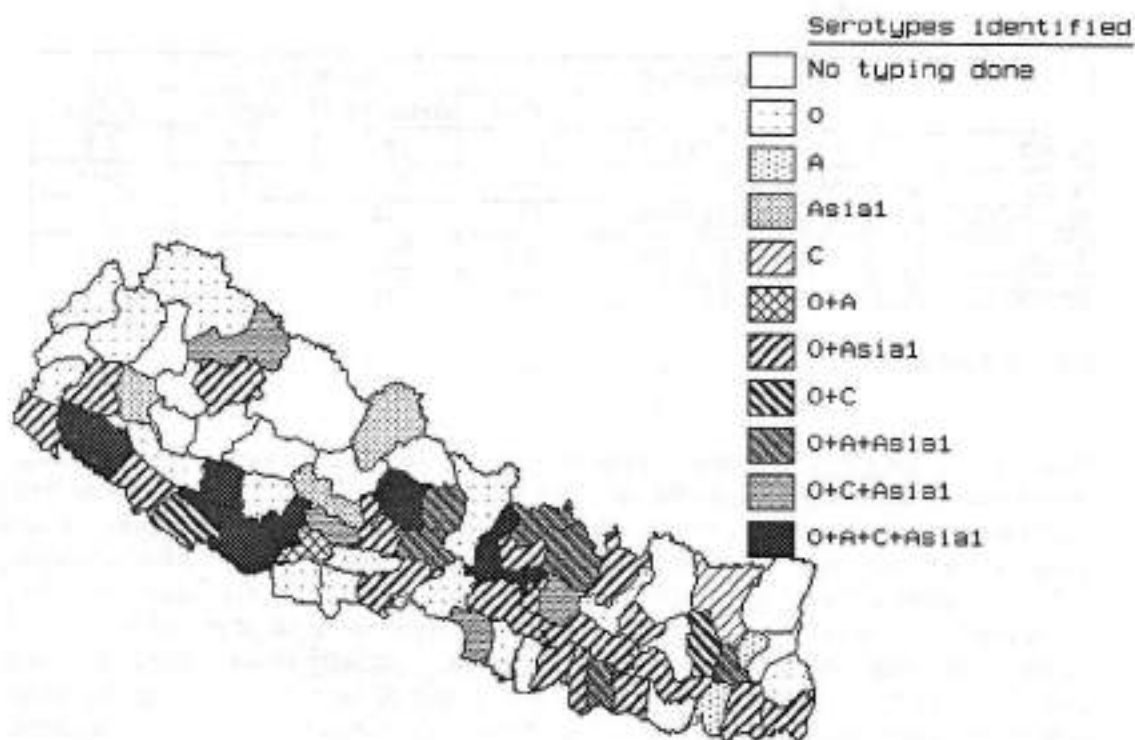


Figure 1. Distribution of serotypes of FMD virus identified from field isolates, Nepal, 1983-1998

The prevalence of different serotypes of FMD virus in neighbouring countries is presented in Table 2. The prevalence of different serotypes of FMD virus is similar in the countries of Indian sub-continent. Serotype C is rare in Nepal, Bangladesh and Pakistan. Very little is known about prevalent serotypes of FMD virus in Tibet. It has been reported that O and A are predominant serotypes responsible for epidemic outbreaks of FMD in the People's Republic of China and Bhutan.

CONTROL STRATEGIES

The socio-economic situation in the country is not conducive to adopt a slaughter policy in the control of FMD. As outbreak of FMD occurs everywhere irrespective of altitude and climatic variation which results from extensive movement of animals throughout the country without proper veterinary sanitary inspection and vaccination of susceptible livestock animals. Regular vaccination as a prophylactic measure alone could be the option for which a potent vaccine with a long duration of protective immunity is required.

Table 2. Distribution of Serotypes of FMD virus in Nepal and Neighbouring Countries

	Serotypes identified	Serotypes			
		O(%)	Asia-1(%)	A(%)	C(%)
Nepal	O, A, Asia-1 and C(1990)	69.2	18.6	8.4	3.8
India	O, A, Asia-1 and C	54.4	21.4	14.1	10.1
Bangladesh	O, A, Asia-1 and C(1994)	57	34	9	0
Pakistan	O, A, Asia-1 and C(1990)	38.5	29.3	14.4	NA
Bhutan	O, A, Asia-1 and C	NA	NA	NA	NA

NA – Not available

Existence of antigenic variation within a serotype has been recognized as a major impediment in launching a successful campaign against FMD. This is more relevant in those countries where the disease is endemic. This results in a natural selection process of the field viruses during the course of the epidemic which occurs round the year (Mukhopadhyay, 1986). Therefore, constant updating of the vaccine strain in relation to the field virus activity is necessary for which monitoring of the subtype activity is essential. It is evident from the published literature that propagation of virus through a partially immune population may give rise to gradual antigenic drift which in due course of time may endanger the whole population through the emergence and establishment of a subtype in the field, thus putting the entire immuno-prophylaxis campaign in jeopardy.

At present, polyvalent vaccine which includes serotype O, A, C and Asia-1 is imported in small quantities from India and recommended to immunise improved breeds of cattle and buffaloes. It is not known that whether imported vaccine gives enough protection against prevalent local FMD virus strain, but there are instances of its failure. There could be many reasons for it. As compared to many other vaccines, FMD vaccines are relatively poor in many aspects. For example, FMD vaccines do not effectively protect the establishment of the virus in the respiratory tract. As a result, a proportion of vaccinated animals which encounter the field virus are likely to become carriers, and thus pose a potential risk to any susceptible animals with which they may come into contact later (Donaldson and Kihm, 1996). If the vaccine virus is dominant to the field virus there will be no problem, but if the field virus is dominant to the vaccine virus there could be a problem. Once a non-immune animal has introduced disease to a farm, there is a strong possibility that the immunity of some vaccinated cattle in close contact will be swamped. In such condition, consideration on herd immunity seems to be more important for successful FMD control. It is highly recommended to have vaccination coverage of 95% for a successful implementation of FMD control programme in endemic zone. Perhaps, this is the reason why 'Stamping out' policy is preferred for FMD control and eradication than mass vaccination.

Prior to 1980 there was no separate programme and infrastructure for FMD control in Nepal. After the establishment of a separate FMD Epidemiological Laboratory, planning, execution, monitoring of the control programme along with diagnosis and serotyping are being done

within the country. Today, National FMD Control Section is a specialized government agency for the epidemiological surveillance and control of FMD in Nepal that is equipped with basic laboratory facilities for diagnosis and serotyping of the FMD virus. Initially, FMD laboratory was established with an objective of vaccine production but for economic reasons, it has not as yet started production of vaccine.

Existence of various serotypes of FMD virus with great antigenic variation, high cost of vaccination, relatively short duration of immunity, shortage of vaccine and vaccine failure makes the FMD control programme very difficult. On the other hand, majority of Nepalese farmers does not realise seriousness of the disease problem because mortality due to the disease is very low. The productivity of local breed of animal is very low and therefore farmers remain reluctant to use protective measure and are interested only at the time of outbreak of the disease.

It is very difficult to control the disease in a country like Nepal, where limited resources, lack of trained manpower, poor infrastructure, limited service network and easy access to quality vaccine is lacking. Even under such circumstances, ring vaccination at the time of outbreak and regular vaccination of exotic and crossbred cattle and buffaloes is encouraged in all districts.

Still more epidemiological work has to be accomplished and awareness of the importance of the disease in farmers has to be created. It is high time to develop a broad-based field epidemiological programme and a strong extension programme for educating the public regarding the importance of this disease in the productive animal population. It is proposed to establish a FMD vaccine bank at the central level so that farmer could get it in time. As private sector is reluctant to keep the stock of FMD vaccine for various reasons and it has become the prime responsibility of government to ensure adequate supply of vaccine for prevention and control of disease. Community based FMD control programme will be launched in the near future in some potential dairy belts and busy animal trading routes with the involvement of farmers group and cooperatives.

FUTURE STRATEGY FOR ERADICATION

FMD has become a disease of regional importance, as there is frequent movement of a large numbers of animals between the countries. Nepal has fairly established livestock trade with India, China and Bangladesh with to and fro movement in both northern and southern borders. Besides the trade, similar animal movement is taking place to adjust pasturing of animals which has created difficulties in controlling movement through the legislation alone. Therefore, a common methodology and strategy at the regional level seems much more important in attempting eradication of animal diseases of the region. The past experience of rinderpest control and the present eradication programme clearly illustrate an example of such cooperation. It is important that India has already initiated state wise FMD vaccination campaign and we will be in a better position to control FMD within a short time due to importation of non-infected animals in large numbers.

The present policy of vaccination of only high yielding animals in dairy pocket area or prophylactic vaccination practices in an outbreak area will never result in the control of

FMD. There is an urgent need to develop technically feasible, economically viable and socially acceptable national programme for FMD eradication in our country.

Different disease control strategies may be applied within the time frame of the programme. The countries of Western Europe and parts of South America followed zonal concept and eradicated FMD. Similarly, it may be more acceptable for an endemic country like Nepal to control and subsequently eradicate the disease zone by zone in a phased manner by the application of large scale immunization of all the susceptible livestock, rather than initially attempting a nation wide strategy. Because of the very large population of susceptible domestic animals in our country, it is not possible to run a nation wide FMD vaccination campaign due to financial and technical constraints. In order to plan mass vaccination programme with minimum risk, zonal concept is taken into consideration. Based on difficult geographical situation and large susceptible animal population, a practical and realistic approach will have to be undertaken which may include;

- Creation of mass vaccination zones based upon animal population size, disease prevalence and animal movement activities.
- Stratification of each region into high, medium and low endemic zones to minimize the cost of operation.
- Use of monovalent vaccines for ring vaccination so that the cost is affordable for farmers.
- Control of animal movement especially in mountain region.

The livestock population in Nepal is unevenly distributed. The population densities of cattle, buffaloes and goats are high as compared to sheep and pigs. A significant number of pigs are in the eastern region that is very important from the epidemiological point of view.

The proposed national FMD vaccination programme is presented in Table 2.

Mass vaccination campaign will be initiated in the Far-Western region where livestock population density is low. It will be easier to control animal movement due to the existence of geographical barriers. The experience gained in this region will help to strengthen vaccination, logistic support and management aspects in other regions. It will take five years to cover the whole country.

The vaccination will have to be made compulsory and free of cost for all susceptible animals (cattle, buffaloes, sheep, goats and pigs) in the project area covering at least 80-90% of the total population. Vaccination campaign will be launched twice a year so that newborn, young or imported animals could be vaccinated in order to maintain herd immunity at the highest possible level. The vaccination time is proposed according to the FMD epidemiology, free time available for farmer's active participation and suitable seasonal conditions. It will be wise to declare FMD week for mass vaccination campaign so that it will be possible to mobilize government, non-government technicians and village animal health workers (VAHW) in the project area.

Table 2. Work-plan and time schedule for mass FMD vaccination campaign in Nepal

Development Region	Time-frame				
	Year 1	Year 2	Year 3	Year 4	Year 5
Far-Western					
Mid-Western					
Western					
Central					
Eastern					
Area Coverage(%)	13.3	42	62	80.7	100
Vaccination Coverage(%)	80 or above	80 or above	80 or above	80 or above	80 or above
Frequency of Vaccination	2	2	2	2	2
Time of vaccination	April - May(Baisakh) and October - November(Ashwin)				
Public awareness campaign	February - April and August - October				

The selection of vaccine and its quality is crucial for the effective control and eradication of FMD. Since Serotype C is rare in Nepal and India and does not pose a serious threat to the livestock population, it is recommended to use Trivalent (O, A and Asia-1) vaccine for mass immunization.

Experience in the control of FMD in South America has clearly brought out the useful application of a concentrated virus vaccine with oil adjuvant. It will be wise to import Trivalent vaccine for mass immunization and monovalent (O – 50%, Asia-1 – 25% and A – 25%) vaccines for ring and strategic vaccination.

Disease surveillance and dissemination of information:

The rapid collection and distribution of information on all aspects of FMD outbreak and its control are essential not only for purposes of epidemiological surveillance, but also to ensure that interested parties (veterinarians, farmers, donor and international agencies) are kept well informed. National FMD Control Section will have to develop close working relationship with Epidemiology unit to produce quality data and epidemiological information. An intensive and efficient system of epidemiological investigation including high quality case history data and field samples for analysis is an essential component of National Control Programme. The information can be used to design or modify vaccination schedules and monitor the effect of vaccination programme on disease control. In fact, epidemiological investigations can help in setting priorities for tracing origin of FMD outbreak in a particular area.

Sero-monitoring of vaccinated population need to be initiated to understand vaccine break down and to evolve a sustainable vaccine strategy.

Diagnostic service

Training and facilities for the identification of field isolates by Sandwich ELISA techniques will be made available in existing Veterinary Diagnostic Laboratories in each region in a phased manner during the project period. Introduction of FMD diagnostic services in each region will help to do serotyping in time so that monovalent FMD vaccine could be used to reduce disease incidence in an outbreak area. Moreover, it will be helpful to receive quality samples from the field. At present, 60% of field samples received in the FMD laboratory are of poor quality.

All the reagents required for FMD diagnosis will be standardized and made available by the National FMD Control Section. It will be necessary to introduce basic facilities for subtyping, antigenic characterization of FMD virus field isolates and sero-epidemiological studies.

REFERENCES

- Donaldson, A.I. and Kihm, U. (1996). Research and technological developments required for more rapid control and eradication of foot and mouth disease. *Rev. Sci. Tech. Off. Int. Epiz.*, 15(3): 863-873.
- Ferris, N.P., Donaldson, A.I., Shrestha, R.M. and Kitching, R.P. (1992). A Review of FMD in Nepal. *Rev. Sci. Tech. Off. Int. Epiz.*, 11(3): 685-698.
- Mukhopadhyay, A.K. (1986). Foot and Mouth Disease - Present Position and Prospect for its Control. *Animal Productivity*, IVRI, Izatnagar. 287-296.

OCCURRENCE OF POULTRY DISEASES IN CHITWAN VALLEY OF NEPAL

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ABSTRACT

This study was undertaken during 1996 to 1998 to find out the occurrence of poultry diseases in Chitwan district of Nepal. Disease-wise, season-wise and age-wise cases of broiler and layer birds examined at different diagnostic centres of Chitwan valley namely Veterinary Teaching Hospital, Rampur, Agroveter Laboratory, Narayangarh, National Livestock and Poultry Disease Diagnostic Clinic, Narayangarh and District Livestock Services Office, Bharatpur were analysed. Out of 1956 cases studied, viral diseases were encountered highest (31%) followed by bacterial (25%) and protozoan diseases (17%) in both broiler and layers. Among the viral diseases Gumboro disease was the most common (50%) followed by Newcastle disease (40%), infectious bronchitis (4%) and Marek's disease (3%). Among the bacterial diseases, chronic respiratory disease was the most prevalent (58%) followed by omphalitis (17%), colibacillosis (11%), enteritis and necrotic enteritis (6%) and bacillary white diarrhoea (5%). Coccidiosis was the most common (77%) protozoan and parasitic disease followed by ascariasis and tapeworm infection. Brooder pneumonia was found most prevalent (21%) among the fungal diseases followed by mycotoxicosis. Visceral gout was also prevalent (18%) in various parts of Chitwan. Viral and protozoan diseases were most common in the summer season whereas bacterial and fungal diseases were common in the autumn. In broilers, 15 to 30 days age groups were mostly affected whereas in layers 1 to 3 months old birds were most susceptible to various types of diseases.

INTRODUCTION

Poultry industry in Chitwan is growing day by day from small-scale farmers rearing 500 commercial birds to big farmers having 40,000 birds. It has developed as one of the commercial enterprises in private sector of Nepal. There were about 1.2 million poultry in Chitwan with 0.8 million broiler and 0.4 million layer in 1995 (Dhakal, 1995). The population has increased to 1.5 million with 0.9 million broiler and 0.6 million layers currently. The broiler breeds available in Chitwan district are Hubbard, Vencobb, Arborecre, Anak 3000, Avian 43, Hubchicks Ross 208 and Shaver Starbro. Similarly, the layer breeds in Chitwan are Hubbard Golden Comet, Hyline brown, Lohman brown, Babcock, Babolna Tetra SL and Keystone brown.

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Chicken contributed more than 8% of the agriculture GDP and nearly 3% of the national GDP. Poultry industry in Nepal is developing as one of the commercial enterprises in the private sector since 1972. A dramatic growth rate in chicken population was observed from late 1988/89 to 1992/93. Evolution of high yielding breeds, advancement in management, nutrition and disease control measures have contributed for the development of poultry industry to develop as a major sector of agribusiness.

Together with the commercialisation of the industry, more and more diseases are becoming prevalent. Accordingly the disease pattern is also changing. Coccidiosis was the most common poultry disease in Chitwan in the early eighties (Dhakal, 1984). By early nineties, infectious bursal disease, Newcastle disease, infectious bronchitis and mycotoxicosis were the major problems (Bhattarai et al., 1991). Newcastle disease with a high morbidity and mortality was recorded in 1996 (Bohara et al., 1996). This study therefore aimed to analyse the current trends of poultry diseases in Chitwan.

MATERIALS AND METHODS

In this study 1956 poultry cases examined at different diagnostic centres of Chitwan district during 1996 to 1998 were analysed. The post mortem diagnosis records of Veterinary Teaching Hospital, IAAS, Rampur, Nepal Agroveter Laboratory, Narayangarh, National Livestock and Poultry Disease Diagnostic Clinic, Narayangarh and District Livestock Services Office, Bharatpur between 1996 and 1998 were analysed. All together, 1956 cases were available for study. Disease-wise, season-wise and age-wise occurrence of different poultry diseases were studied. Laboratory diagnosis report obtained from Indian Veterinary Research Institute (IVRI), India, Nepal Agricultural Research Council (Veterinary Laboratory), Tripureswor and Veterinary Teaching Hospital Laboratory, Rampur was also included.

RESULTS

Study Population

Total number of poultry cases examined at different diagnostic centres of Chitwan during 1996 to 1998 are presented in Table 1. Of 1956 cases included for study, 35% were from National Livestock and Poultry Diseases Diagnostic Clinic, 28% were from Nepal Agroveter Laboratory, 23% were from Veterinary Teaching Hospital Rampur and 13% were from District Livestock Services Office, Bharatpur.

Table 1. Poultry cases examined at different diagnostic centres of Chitwan during 1996 to 1998

S.N.	Diagnostic Centres	Number	Percent
1	Veterinary Teaching Hospital, Rampur	457	23.4
2	Nepal Agroveter Laboratory, Narayangarh	546	27.9
3	National Livestock and Poultry Diseases Diagnostic Clinic, Narayangarh	692	35.4
4	District Livestock Services Office, Bharatpur	261	13.3
	Total	1956	100.0

Table 4. Viral diseases of poultry in Chitwan

S.N.	Disease	Number	Percent
1	Gumboro (Infectious bursal disease)	306	50.0
2	Newcastle disease	247	40.4
3	Egg drop syndrome-76	6	1.0
4	Infectious bronchitis	24	3.9
5	Fowl pox	8	1.3
6	Marek's disease	21	3.4
	Total	612	100.0

Table 5. Bacterial diseases of poultry in Chitwan

S.N.	Disease	Number	Percent
1	Omphalitis	84	17.4
2	Chronic respiratory disease (CRD)	280	58.1
3	Colibacillosis	53	11.0
4	Enteritis and necrotic enteritis	31	6.4
5	Fowl cholera	9	1.9
6	Bacillary white diarrhoea	25	5.2
	Total	482	100.0

Table 6. Protozoan and parasitic diseases of poultry in Chitwan

S.N.	Disease	Number	Percent
1	Coccidiosis	250	76.9
2	Ascariasis	63	19.4
3	Tapeworm	12	3.7
	Total	325	100.0

Fungal, metabolic and nutritional deficiency diseases are presented in Table 7. Brooder pneumonia was the most common fungal disease followed by mycotoxicosis. Metabolic diseases like visceral gout, ascites and fatty liver haemorrhagic syndrome were also recorded in Chitwan. Vitamin and mineral deficiency cases represented 28% of cases of this category.

Table 7. Fungal, metabolic and nutritional deficiency diseases of poultry in Chitwan

S.N.	Disease	Number	Percent
1	Vitamin and mineral deficiency	81	28.4
2	Brooder pneumonia	62	21.8
3	Visceral gout	50	17.5
4	Mycotoxicosis	39	13.7
5	Ascites	29	10.2
6	Fatty liver haemorrhagic syndrome	24	8.4
	Total	285	100.0

Seasonal occurrence of poultry diseases

Viral diseases were found most prevalent in summer season (38%) followed by spring (34%), autumn (20%) and winter (8%) (Table 8). Bacterial diseases were most common in the autumn (42%) followed by summer (24%) and spring (22%). Protozoan and parasitic diseases were recorded highest in summer (37%) followed by spring (29%) and autumn (22%). Fungal, metabolic and nutritional diseases were most common in autumn (39%) followed by spring (27%) and summer (23%). Mixed Infections were found mostly in summer season. The prevalence of poultry diseases in winter was comparatively lower than other seasons.

Table 8. Seasonal occurrence of poultry diseases (%) in Chitwan

S.N.	Diseases	Winter	Spring	Summer	Autumn
1	Viral	7.5	34.5	37.5	20.5
2	Bacterial	12.2	22.4	23.6	41.7
3	Protozoan and Parasitic	11.6	29.1	37.2	22.1
4	Fungal, metabolic and nutritional	10.7	27.3	23.1	38.8
5	Mixed infection	8.1	37.3	44.6	10.0

Winter: December - February Spring: March - May
 Summer: June - August Autumn: September - November

Age-wise occurrence of poultry diseases

Table 9 presents age-wise occurrence of broiler and layer diseases in Chitwan. Broilers of 15 to 30 day age groups were most susceptible to the diseases. The next susceptible age group of broilers was of 30 to 45 days.

Table 9. Age-wise occurrence of diseases in broilers and layers

Breed type and age group	No. of cases	Percent	Common diseases
Broilers (age in days)			
0-15	188	19.04	omphalitis, brooder pneumonia
15-30	374	37.89	Gumboro, coccidiosis, Newcastle, colibacillosis
30-45	295	29.88	Gumboro, CRD, Coccidiosis, Newcastle
Above 45	130	13.17	Newcastle, CRD, Coccidiosis
Layers (age in months)			
0-1	234	24.14	Omphalitis, brooder pneumonia
1-3	263	27.14	Gumboro, coccidiosis, Newcastle, colibacillosis
3-5	161	16.61	Infectious bronchitis, Marek's, CRD, deficiency disease
5-7	142	14.65	CRD, Newcastle, Marek's, Mixed infection
Above 7	169	17.44	Infectious bronchitis, EDS-76, Mixed infection

In layers, more than half of the recorded diseases were among the birds of age group 0 to 3 months.

Common diseases

Common poultry diseases in Chitwan are summarized in Table 10. Gumboro disease was the most common (18%) followed by chronic respiratory disease (17%), coccidiosis (15%) and Newcastle disease (15%). Other diseases recorded were omphalitis, deficiency disease, ascariasis, brooder pneumonia, colibacillosis, visceral gout, mycotoxicosis, enteritis and necrotic enteritis, ascites, bacillary white diarrhoea, infectious bronchitis, fatty liver haemorrhagic syndrome and Marek's disease.

Table 10. Common poultry diseases in Chitwan.

S.N.	Disease	Number	Percent
1	Gumboro disease	306	18.3
2	Chronic respiratory disease	280	16.8
3	Coccidiosis	250	15.0
4	Newcastle	247	14.8
5	Omphalitis	84	5.0
6	Vitamin and mineral deficiency	81	4.9
7	Ascariasis	63	3.8
8	Brooder pneumonia	62	3.7
9	Colibacillosis	53	3.2
10	Visceral gout	50	3.0
11	Mycotoxicosis	39	2.3
12	Enteritis and necrotic enteritis	31	1.9
13	Ascites	29	1.7
14	Bacillary white diarrhoea	25	1.5
15	Infectious bronchitis	24	1.4
16	Fatty liver haemorrhagic Syndrome	24	1.4
17	Marek's disease	21	1.3
	Total	1669	100

DISCUSSION

This study was conducted during 1996 to 1998 to investigate the occurrence of poultry diseases in Chitwan in different seasons and in different age groups of poultry. Of 1956 cases included in the study, only 13% of the poultry birds were examined at district veterinary hospital, Bharatpur. This finding indicated that poultry farmers of Chitwan are attracted to the private clinics and laboratories rather than government hospitals. Alternatively, the private clinics might have the advantages of easy access or opening hours more convenient to the farmers.

Viral diseases were the most common group of poultry diseases identified in the present study. Gumboro, Newcastle, infectious bronchitis and Marek's disease were predominant among viral diseases. During the study period, severe outbreak of Newcastle disease (Bohara et al., 1996), infectious bronchitis in layers (Dhakal, 1997) and Marek's disease was recorded in Chitwan. These outbreaks might have contributed for the higher number of cases of viral diseases recorded in the present study. Egg drop syndrome-76, Infectious bronchitis and Marek's disease were the emerging poultry diseases of economic importance.

Chronic respiratory disease (CRD) was the major bacterial disease recorded. CRD affects birds of all ages and transmits either by direct contact or through egg. CRD is often complicated by viral infections such as infectious bronchitis and Newcastle disease, and bacterial infection like colibacillosis. Omphalitis and colibacillosis caused by *E. coli* is mainly due to bad hygienic hatchery management and poor sanitation. This indicates the need to improve the hygiene both at the hatchery and the farm level. Coccidiosis, a significant problem of poultry industry throughout the tropics, is also a major problem in Nepal. Good litter management practices need further extension to reduce the losses due to this disease.

Brooder pneumonia and mycotoxicosis were the common fungal diseases of chickens recorded in Chitwan. Fungal diseases are mostly transmitted through contaminated feed and litters. Mouldy feeds are to be avoided to prevent spread of infection. Mycotoxins affecting poultry industry are aflatoxin, ochratoxin, T2toxin, citrinin, vomitoxin, zeralenone etc. Visceral gout was prevalent in various parts of Chitwan. Ascites and fatty liver haemorrhagic syndrome are other metabolic diseases observed in broiler and layer birds.

Nutritional deficiency diseases such as curled toe paralysis, star gazing posture, encephalo malacia, lameness, rickets etc have been recorded.

Viral diseases like Gumboro disease, Newcastle disease and infectious bronchitis were most common in summer seasons. Other viral diseases have been reported all the year round. Bacterial diseases were most common in the autumn seasons followed by summer season. In this study, CRD was common in winter season. This is in contrast to Govindarajan (1996) who reported CRD to be common in summer season. Protozoan and parasitic diseases were common in the summer season followed by spring season. Coccidiosis was observed mostly in spring followed by summer season. Other nutritional and metabolic diseases occurred all the year round.

Both in layers and broilers, diseases recorded during the early age group was omphalitis and brooder pneumonia. Gumboro, coccidiosis, Newcastle and colibacillosis were the common diseases in the next age group of poultry. In layer, chronic respiratory disease (CRD) was seen more in the birds aged 3 to 5 months which is similar to that reported by Shakila et al. (1996).

In the present study most susceptible age group of poultry affected by Gumboro disease was 1 to 3 months, by Newcastle disease 5 to 7 months, by Marek's disease 3 to 5 months and by infectious bronchitis more than 5 months. Major causes of mortality in the layers aged 0 to 1

month was found to be unabsorbed yolk. Aflatoxicosis and gout were observed in the birds aged 5 months old.

CONCLUSIONS

- Gumboro disease, chronic respiratory disease, coccidiosis and Newcastle diseases were the most common diseases encountered in Chitwan.
- Egg drop syndrome-76, infectious bronchitis and Marek's disease were the emerging poultry diseases of economic importance.
- Visceral gout, ascites and fatty liver haemorrhagic syndrome were common metabolic diseases.
- Occurrence of poultry diseases was found higher in summer and autumn seasons in comparison to spring and winter.
- In broiler, 15 to 30 days age groups were most susceptible to diseases.
- In layers, 0 to 3 months old birds were most susceptible to diseases.
- Diseases of poor hatchery management and sanitation, such as omphalitis and colibacillosis were common in young birds. Strict biosecurity measures from hatchery to marketing and timely vaccinations are required for the control of poultry diseases in Chitwan.

ACKNOWLEDGMENT

Authors are thankful to the clinicians of National Livestock and Poultry Disease Diagnostic Clinic, Nepal Agroveter Laboratory, Veterinary Teaching Hospital, Rampur and District Livestock Service Officer, Bharatpur.

REFERENCES

- Basnet, H.B.; Bhandari, D.R. and Shrestha, D.R. (1996). *Preliminary study on poultry husbandry and health at Rampur and its vicinity*. souvenir Vet. Con. 96, pp.35.
- Bhattarai, P.C.; Singh, S.B.; Shrestha, and Kandel, K.P. (1991). *Economic of poultry raising in Chitwan, Nepal*. IAAS Research Report pp. 638-644.
- Bohora, K.B.; Dhakal, I.P.; Bhattarai, and Adhikary, D.R. (1996). *Outbreak of Newcastle disease in commercial chickens in Nepal*. bull. Vet. Sci. & A.H. Vol 24:100-104.
- Cullen, G.A; Gordon, R.F.; Harry, E.G. and Jordon, F.T.W. (1979). *Bacterial diseases*. In *Poultry Disease* (Edited by Gordon, F.R.) 1st edition, ELBS. London pp 10-35.
- Dhakal, I.P. (1984). *Research note on the common disease of livestock and poultry in Chitwan*. J. Inst. Agric. Anim. Sci. 5:115-119.
- Dhakal, I.P. (1992). *Occurrence and counter measures of Gumboro diseases, an experience in Chitwan*. Poultry Munch, Apr./May 1992 pp 31-35.
- Dhakal, I.P. (1995). *Poultry farming in Chitwan, an overlook*. Poultry bulletin, No. 1 pp 1-6.
- Dhakal, I.P. (1996). *ND ravages poultry in Nepal*. Poultry Punch 12(10) pp 116.
- Dhakal, I.P. (1997). *Occurrence of Infectious bronchitis in commercial layers of Chitwan*. The blue cross vol 1 pp 8-9.
- Govendarajan, J and Seshdri, S.J. (1996). *Disease experience of 1995*. Poultry Punch vol. 12 (6) pp 67-76.

- Mahoto, S.N.; Thakuri, K.C.; Thapa, P.B. and Yadav, J.N. (1989/90). *Prevalence of Salmonella infection among chickens of Pakhribas Agricultural Center*. Bull. Vet. Sci. & A.H. Nepal, 17 & 18:1-8.
- Rasali, D.P.; Bhattarai, T.C. and Yadav, C.D. (1992). *Preliminary report on an outbreak of Infectious bursal disease in commercial chickens in Chitwan district of Nepal*. Poultry Munch, Apr/May 1992, pp. 25-28.
- Shikila, S.; Raniprameela, D.; Kalpana, G.; Ravi Kumar, A.; Sudhakaran, R. (1996). *Mortality pattern of layer birds in Chittoor District of Andhra Pradesh*. Poultry Punch vol 12(7) pp 89 to 93.

CLINICO-HAEMATOLOGICAL CHANGES IN EXPERIMENTAL AFLATOXICOSIS IN BUFFALO CALVES

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ABSTRACT

Clinical and haematological studies such as bleeding time, clotting time, total erythrocytic count, total and differential leucocytic count, haemoglobin percent, packed cell volume, mean corpuscular volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration percent and erythrocytic sedimentation rate were studied in buffalo calves given aflatoxin at the dose of 10 µg/kg body weight daily. Blood samples for analysis were collected at 0 day and at 10 days of interval for 30 days. Increased bleeding time, clotting time, total leucocytic count, neutrophil percent, packed cell volume and erythrocytic sedimentation rate were observed where as total erythrocytic count and haemoglobin percent decreased significantly after feeding the toxin. The changes were found to be dose dependent. Clinical symptoms observed and different haematological changes were discussed.

INTRODUCTION

Aflatoxicosis is the poisoning by ingestion of aflatoxin through food contaminated by toxigenic strains of *Aspergillus flavus* and *Aspergillus parasiticus*. *Aspergilli* are ubiquitous and the potential for contamination of food-stuff and animal feed is widespread. The occurrence and magnitude of aflatoxin contamination varies with geographical and seasonal factors. In most part of the year, the season of Bihar is more conducive for growth of *aspergilli* due to high humidity and temperature. Most of the human food (Sinha et al., 1997) and animal feed (Sinha and Singh, 1989) have been found contaminated with aflatoxins. When the contaminated feed is consumed by animals aflatoxicosis occurs. Though aflatoxins cause a wide variety of clinical signs depending upon the nature and concentration of toxin present, duration of exposure and health status (Prelusky et al., 1994), yet the diagnosis of aflatoxicosis poses a challenging problem to Veterinarians. Thus, the present investigation was under taken to find out the clinico-Haematological changes in Experimentally produced aflatoxicosis in buffalo calves, which may aid in diagnosis of aflatoxicosis.

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MATERIALS AND METHODS

Apparently healthy Murrah male buffalo calves of one year old and weighing about 35-40 kg were the subject of study in the present investigation. These were screened for the presence of any protozoal cysts and helminth ova, and for any clinical disease. Only such animals were used which were found to be negative. Aflatoxins extracted in the laboratory, characterised by TLC and quantitated spectrophotometrically (Sinha et al., 1997) were used. Aflatoxin B₁ was given to three buffalo calves at 10 µg/kg of body weight daily till the end of the experiment or animal died. Blood samples were collected at 0 day and at 10 days of interval until death of calves. Blood was collected from jugular vein in sodium and potassium oxalate anticoagulant at 0 day (pre-feeding) and at 10, 20 and 30 day post feeding. The haematological parameters including bleeding time (BT), clotting time (CT), haemoglobin percent (Hb%), packed cell volume (PCV), total erythrocytic count (TEC), total leucocytic count (TLC) and differential count (DC) were determined, and mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) was calculated as described by Schalm et al. (1975). The clinical symptoms were also noted during the period of observations.

RESULTS AND DISCUSSION

Clinically all the three buffalo calves started showing generalised depression and anxious look after 10 days of post intoxication. Later, variable signs of depression, somnolence, anorexia, reduced thirst, nervous sign and emaciation were observed. Out of the three experimental calves, one died on 22nd days of post intoxication, whereas two died 30 days of post intoxication. Similar findings were observed by Gopal et al. (1968) and Lynch et al. (1972). Early death in one calf may be due to the rapid liver damage as suggested by Edds (1973). Results of the different haematological tests have been depicted in Table-1.

It is evident from Table-1 that total erythrocytic count decreased significantly ($P < 0.01$) i.e. from 6.56 ± 0.12 (0 day) to 2.98 ± 0.29 (30 days post intoxication) which was evident from day 20 post intoxication. In contrast, the total leucocytic count increased significantly from day 20 onwards ($P < 0.001$) i.e. from 6.85 ± 0.16 (0 day) to 12.85 ± 0.25 (30 days post intoxication). Percentage of neutrophils also increased significantly ($P < 0.01$). This increase of TLC and neutrophils may be due to the inflammatory processes set in the different visceral organs of animals. Consumption of more level of aflatoxin resulted in lymphopenia with compensatory neutrophilia. This significant reduction in lymphocyte population might be due to the damage of lymphoid follicles as observed by Sahoo et al. (1996). They opined that this is due to the immunosuppression by aflatoxin. Similar finding was also reported by Tung et al. (1975).

The value of MCV, MCH and MCHC indicate the health status of animal in respect to erythropoiesis and certain metabolism. The MCV provides the average cell size in μ^3 , MCH express the average weight of Hb in red blood cells while MCHC gives the average percentage of MCV, which the Hb occupies. The above parameters are highly variable in different species (Schalm et al., 1975).

Table 1. Haematological profile of experimental aflatoxicosis.

S. N.	Name of the tests	Number in days			
		0 (n=3)	10 (n=3)	20 (n=3)	30 (n=2)
1	Bleeding time (Minutes)	3.84 ± 0.25	4.02 ± 0.15*	4.78 ± 0.25**	5.4 ± 0.16***
2	Clotting time (Minutes)	6.57 ± 0.13	6.82 ± 0.25 ^{NS}	7.32 ± 0.24 ^{NS}	11.25 ± 0.30***
3	Total Erythrocyte count (TEC × 10 ⁶)/cmm	6.56 ± 0.12	7.05 ± 0.08 ^{NS}	5.85 ± 0.11**	2.98 ± 0.29***
4	Total Leucocyte count (TLC × 10 ³)/cmm	6.85 ± 0.16	6.98 ± 0.25 ^{NS}	10.80 ± 0.21 ***	12.85 ± 0.25 ***
5	Differential count :				
	a. Neutrophils (%)	34.33 ± 2.80	49.67 ± 2.76*	63.34 ± 3.84**	68.0 ± 2.89**
	b. Eosinophils (%)	3.33 ± 0.58	3.33 ± 0.58 ^{NS}	3.33 ± 0.58 ^{NS}	2.67 ± 0.82 ^{NS}
	c. Lymphocytes (%)	61.67 ± 3.68	46.33 ± 2.88 ^{NS}	32.33 ± 2.22 ^{NS}	28.33 ± 2.40 ^{NS}
	d. Monocytes (%)	0.67 ± 0.2	0.67 ± 0.2 ^{NS}	1.0 ± 0.00 ^{NS}	1.0 ± 0.00 ^{NS}
	e. Basophils (%)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
	Total	100.00	100.00	100.00	100.00
6	Haemoglobin (Hb) gm%	10.93 ± 0.06	9.8 ± 0.14**	7.75 ± 0.22***	6.43 ± 0.64**
7	Packed cell volume (PCV%)	34.6 ± 0.48	35.54 ± 0.22 ^{NS}	37.48 ± 1.24 ^{NS}	39.20 ± 0.44**
8	Mean corpuscular volume(MCV μ ³)	52.74 ± 0.98	50.41 ± 0.82 ^{NS}	64.06 ± 0.69***	131.54 ± 1.22***
9	Mean corpuscular haemoglobin (MCH μ/gm)	16.66 ± 0.68	13.90 ± 0.50*	13.24 ± 1.26 ^{NS}	21.57 ± 0.83*
10	Mean corpuscular haemoglobin concentration (MCHC %)	31.58 ± 0.92	27.97 ± 0.66*	20.67 ± 1.99**	16.40 ± 0.98***
11	Erythrocyte sedimentation rate (ESR/mm/hr)	53.6 ± 0.48	52.2 ± 0.50 ^{NS}	60.27 ± 0.25***	81.15 ± 3.38***

N.B.: NS = Non-significant.

* = P<0.05 ** = P<0.01 *** = P<0.001

The MCV increased as the dose of aflatoxin B₁ (AF B₁) increased, this indicates the effect of AF B₁ on erythropoiesis which is evident as the MCV increased significantly (P<0.001) i.e. 131.54 ± 1.22 μ³ after 30 days of post feeding as compared to 52.74 ± 0.98 μ³ at 0 day intoxication. The increased size of RBC of rat, pigeon and human beings after administration of aflatoxin has been reported by Verma and Raval (1989). They also observed fall in TEC and suggested that this fall may be due to morphological alteration and increased destruction of RBC during their passage through reticulo-endothelial tissue as is known to occur in sickle cell anaemia. In the present investigation also TEC decreased significantly resulting into compensatory increase of MCV (Table 1). The maintenance of MCH higher after 30 days of AF B₁ intoxication over normal value reveals that incorporation of Hb in RBC is not affected rather a compensatory increase in Hb incorporation in larger cell size is discernible.

However, prolonged treatment of aflatoxin (i.e. after 30 days post intoxication) has severely affected the process of erythropoiesis resulting into drastic decrease in number of erythrocytes. Due to reduced number of erythrocytes in circulation the total Hb concentration decreased. Though the MCH in prolonged AF B₁ treated buffalo calves remained nearly similar throughout the study but the MCHC value decreased significantly due to increased MCV and lack of compensatory mechanism between the incorporation of Hb and increase in cell size directly indicate moderate degree of hypochromia.

The mean value of ESR, bleeding time and clotting time in the present study elevated significantly which are in concordance with the findings of Murthy et al. (1984); who reported low value of ESR. As suggested by Schalm et al. (1975), low level of serum protein may result in increase of ESR and blood clotting time. This may be the cause of increased value of ESR and blood clotting time as the serum protein level was decreased significantly (Sinha et al. 1998).

The various clinico-haematological findings serve as an adjunct for diagnosis of aflatoxicosis in buffalo, as the disease is of great economic and public health importance.

REFERENCES

- Edds, G.T. (1973). Acute aflatoxicosis, a review, *J. Am. Vet. Med. Assoc.* 162: 304-309.
- Gopal, T., Zaki, S., Marayanswamy, M. And Premilata, S. (1968). Aflatoxicosis in dairy cattle. *Indian vet. J.* 45: 707-712.
- Lynch, G.P. (1972). Mycotoxicosis in feed stuffs and their effect on dairy cattle. *J. Dairy Sci.* 55: 1243-1255.
- Murthy, A.S., Rama Rao, P. and Mahender, M. (1984). A study of biochemical and haematological changes in experimental aflatoxicosis in cattle and buffalo calves. *Indian Vet. J.* 61: 911-917.
- Prelusky, D.B., Rotter, B.A. and Rotter, R.G. (1994). Toxicology of Mycotoxin. In: Miller, J.D. and Trenholm, H.L. (Eds). *Mycotoxin in Grain*. Eagan press, MA, USA, pp. 359.
- Rajan, A., Naik, N.K. and Sirdas, C.G. (1973). Comparative study of experimental aflatoxicosis in crossbred jersey and buffalo calves, *Kerala J. Vet. Sci.* 4: 109-113.
- Sahoo, P.K., Chattopadhyay, S.K. and Sikdar, A. (1996). Immunosuppression effects of induced aflatoxicosis in rabbits. *J. Appl. Anim. Res.* 9: 17-26.
- Schalm, G.W., Jain, N.C. and Corrol, E.J. (1975). *Veterinary Haematology*, 3rd Eds, Lea and Febiger Philadelphia.

- Sinha, B.K. and Singh, R.N. (1989). Studies on aflatoxigenic fungi from human and animal feed, extraction and characterization of aflatoxin and its pathogenicity. *Toxin. Rev.* (USA), **8**: 321-327.
- Sinha, B.K., Ranjan, K.S. and Pandey, T.N. (1997). Aflatoxin level in commonly used pulses in Bihar. *Indian Nutri. and Diet.* **34** : 225.
- Sinha, B.K., Ranjan, K.S., Pandey, T.N., Gupta, R.P. and Prasad K.R. (1998). Some biochemical profile in experimental aflatoxicosis in buffalo calves. *Buffalo J.* (In Press).
- Tung, H.T., Cook, F.W., Wyatt, R.D. and Hamilton, P.B. (1975). The anaemia caused by aflatoxin. *Poult. Sci.*, **54**: 1962-1970.
- Verma, R. J. and Raval, P.J. (1989). Morphological alteration in red blood cells after aflatoxin treatment. *International symposium on Agricultural and Biological aspects of aflatoxin related health hazards held at Delhi 22-25 March, 1989* p.21.

DEMAND AND SUPPLY OF RAW MATERIALS FOR LIVESTOCK AND POULTRY FEED

M.N. Lohani¹ and J.L. Amatya²

ABSTRACT

The feed industries of Nepal produced a total of 205,158 MT of balanced feed for livestock and poultry in 1998. This amount was sufficient to meet the demand of the country for these products. The industry however has a dependency of 46 percent on import for the supply of its raw materials. This dependency ranged from 25 to 100 percent for various raw materials. The dependency on import was higher for protein feed ingredients (90 percent) than for energy feed ingredients (28 percent). The effects arising from this dependency has been analysed. An appropriate policy from the Government has a great potential for substantially reducing this dependency.

INTRODUCTION

Commercial Poultry Industry of Nepal experienced a tremendous growth in 1990s. The annual growth in the production of chicken eggs and broiler meat has been estimated at 10.5 and 18.3 percents, respectively in this decade (Lohani et.al., 1996). This industry was comparatively more matured in the eastern, central and western regions of Nepal. The mid-west and far-west regions were lagging far behind in poultry production.

In 1996, the commercial poultry sector registered a contribution of NPRs 6.5 billion to the National GDP and created 33,000 rural employment (Lohani et.al., 1996). This industry produced 75,000 MT poultry manure - a very high quality fertiliser, as by-product, which significantly substituted the import of chemical fertiliser from the third country.

The animal feed industries of Nepal were mainly responsible for the expansion and development of local poultry industry. As per the data available from Nepal Feed Industries Association, there were 149 feed industries located in 25 districts of Nepal in 1999. Of them, major ones were,

- Pancharatna Feed Pvt. Ltd., Chitwan
- Himalaya Feeds. Pvt. Ltd., Chitwan
- Everest Feed Industries, Chitwan
- Nawadurga Poultry Feed Industries, Chitwan
- Sanjiwani Aahar Udhyog, Chitwan
- Nepal Feed Products, Kathmandu
- Ratna Feed Pvt. Ltd., Kathmandu

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- Valley Feed Pvt. Ltd., Kathmandu
- Quality Feed Pvt. Ltd., Kathmandu
- Adhunik Poultry Feed Industries, Kathmandu
- Adhunik Feed Industries Pvt. Ltd., Kathmandu
- Star Feed Products Pvt. Ltd., Kathmandu
- Kantipur Feed Industries, Bhaktapur
- Udaya Feed Industries, Bhaktapur
- Kisan Feed Industries, Lalitpur

The production and supply of the above 15 feed industries was about half of the country's demand for balanced livestock and poultry feed. The other half of the feed was produced and supplied by the remaining 134 smaller feed industries. They provided not only the feed, but also other inputs and services such as day-old chicks, vaccines, veterinary services, marketing of eggs and broiler birds, etc. to farmers, either directly or through their dealers.

LIVESTOCK AND POULTRY FEED PRODUCTION

Nepal was not only self sufficient in balanced animal and poultry feeds, but was also exporting it across the border into India. In 1998, a total of 205,158 metric ton of animal and poultry feed was produced in Nepal (Table 1). Of this quantity, 94.6% was poultry feed, 4.4% cattle feed and 1% others (pig feed, quail, duck, rabbit, horse and fish feed). This figure included the mixed commercial feed only. Besides this, wheat bran, rice polish, corn flour etc. were also extensively used in animal feeding, especially in the subsistence production systems.

Table 1. Livestock and Poultry Feed Production in Nepal in 1998

S. No.	Type of Feed	Quantity MT	Percent
1	Poultry Feed (Commercial Broiler ration, Commercial Layer ration, and Breeder Chicken ration)	194,153	94.6
2	Cattle Feed	9,125	4.4
3	Other Feeds (Pig, Quail, Duck, Rabbit, Horse and Fish feed)	1,880	1.0
	Total, MT	205,158	100

DEMAND FOR FEED INGREDIENTS

The demand for different ingredients for livestock and poultry feed in 1998 has been presented in Table 2. The industry consumed over 138 thousand metric tons of energy feed ingredients which is 67.5 percent of total. Similarly, the demand for protein feeds was over 56 thousand metric tons, which is 27.5 percent of the total ingredient requirement. The macro-mineral source, micro-minerals, vitamins and other feed additives together constituted about 5 percent of the total feed ingredients.

Of the major ingredients, Yellow maize topped the list with over 41 percent of the total, followed by soybean oil meal and rice bran/polish each with 10 percent. These feeds were mainly yellow maize and soybean oil meal based ration, with low use of grain by-products. The low use of the by-products was due to their poor quality and high market price. Wheat bran and Til cake were used by the feed industry at the rate of about 6 percent while it was 5 percent with sunflower cake. The demand for mustard cake and fishmeal was about 3 percent each.

Table 2. Demand for Livestock and Poultry Feed Ingredients in Nepal in 1998.

S. No.	Feed Ingredients	Quantity, MT	Percent
	<i>Energy Source ingredients</i>	138368	67.5
1	Yellow Maize	85,469	41.7
2	Rice Bran/Rice Polish	20,510	10.0
3	Deoiled Rice Bran/ Rice Polish	8,496	4.1
4	Wheat	6,135	3.0
5	Wheat Bran	12,744	6.2
6	Molases	5,014	2.5
	<i>Protein Source Ingredients</i>	56,425	27.5
7	Soybean Oil Meal	21,995	10.7
8	Til Cake	12,196	5.9
9	Sunflower Cake	10,255	5.0
10	Mustard Cake	6,099	3.0
11	Fish Meal	5,880	2.9
	<i>Minerals/Vitamins/Feed Additives</i>	10,365	5.0
12	Bone Meal	2,051	1.0
13	Oyster Shell	2,142	1.0
14	Limestone	3,993	1.9
15	Feed Supplement (Vitamins/Minerals/feed additives)	2,179	1.1
	TOTAL, MT.	205,158	100

SUPPLY SITUATION OF FEED INGREDIENTS

The data on different feed ingredients and source of supply in 1998 are presented in Table 3. The data show that Nepalese feed industry had a dependency of 46 percent on imported raw materials, the local supply accounting for only 54 percent of the total demand. About 95 percent of the feed ingredients were agriculture products/ by-products. Even though Nepal is an agricultural country, it was surprising to see that the local supply could meet only 54 percent of the total demand.

The data also show that the local feed industry was about 72 percent self sufficient in energy feed ingredients. The local supply of yellow maize could meet 60 percent of its demand. The

remaining 40 percent demand was met through import from India. While the country was self sufficient in rice bran/polish, deoiled rice bran/polish wheat and wheat bran as animal feed ingredients, the local supply of molasses could meet only 20 percent of its demand.

The situation was very different with protein feed ingredient. The country's feed industry depended on import for 90 percent of its protein ingredients. This dependency was 100 percent for soybean oil meal, sunflower cake and fishmeal and 90 percent for Til cake. It was only mustard cake supply from local source that met 75 percent demand of the industry. There were several traders that were involved in cross-border trading of feed ingredients, and therefore no difficulty was faced in their supply. It nevertheless showed a very high vulnerability of the Nepalese feed industry.

Table 3. Supply Quantity and Source of Feed Ingredients in Nepal in 1998.

S. No.	Feed Ingredients	Quantity, MT		Percent	
		Local Source		Imported	
		MT	%	MT	%
	<i>Energy Source Ingredients</i>	100,169	72	38,199	28
1	Yellow Maize	51,280	60	34,187	40
2	Rice Bran/Rice Polish	20,510	100	0	0
3	Deoiled Rice Bran/ Rice Polish	8,496	100	0	0
4	Wheat	6,135	100	0	0
5	Wheat Bran	12,744	100	0	0
6	Molasses	1,003	20	4,011	80
	<i>Protein Source Ingredients</i>	5,794	10	50,631	90
7	Soybean Oil Meal	0	0	21,995	100
8	Til Cake	1,220	10	10,976	90
9	Sunflower Cake	0	0	10,255	100
10	Mustard Cake	4,574	75	1,525	25
11	Fish Meal	0	0	5,880	100
	<i>Minerals/Vitamins/Feed Additives</i>	5,019	48	5,346	52
12	Bone Meal	1,026	50	1,025	50
13	Oyster Shell	0	0	2,142	100
14	Limestone	3,993	100	0	0
15	Vitamins/Minerals/feed additives	0	0	2,179	100
	TOTAL	110,982	54	94,176	46

The local supply met 50% of the demand for bone meal and 100 percent for limestone. The local industry's dependency on import for oyster shell and vitamins/micro-minerals/feed additives was 100 percent. The industry imported vitamins, amino acids and other feed additives from India as well as from the third country.

CONCLUSION AND RECOMMENDATION

Even though Nepal was self sufficient in meeting the demand for balanced livestock and poultry feed, it has to depend 46 percent on imported raw feed ingredients. Except part of vitamins, amino acids and some feed additives, the country imports these raw materials from India. The total dependence for all the imported raw materials in only one country made this industry highly vulnerable and risky. Its negative effect was being experienced in the form of high fluctuation in quality and price of these raw materials. As a result, the quality of the feed products manufactured by using these materials was quite variable and the price fluctuation was significantly high. This negative effect was finally transferred to the livestock and poultry farmers of Nepal.

It is recommended that the Ministry of Agriculture should give priority to increase the production of crops that are used in livestock and poultry feed, particularly the yellow maize and oil seeds. Likewise, the Ministry of Industry should promote the oil seed solvent industry in the country by discouraging the industries that import edible oil in bulk and market it in smaller packets.

It is getting late for His Majesty's Government of Nepal to create an independent autonomous institution to address the problems faced by the poultry industry including the feed industry and to reduce such vulnerabilities.

EFFECT OF INDIAN CHICKS DUMPING ON NEPALESE POULTRY INDUSTRY

M.N. Lohani¹ and J.L. Amatya²

ABSTRACT

The hatchery industries of Nepal produced and distributed 400,000 day-old broiler chicks per week, sufficient to meet its demand in the country in 1997/98. But five large Indian hatcheries dumped 132,000 day-old broiler chicks per week in the Nepalese market during that period. As aftermath shock of this dumping, the hatchery industries and the commercial broiler farmers incurred a loss of Rs. 104 million and Rs. 1,269.7 million, respectively, in one year. This dumping also caused severe negative impact on Nepalese feed industry, chicken marketing industry, feed ingredient supply sector, poultry health sector, financial institutions as well as the country's economy. Such an unhealthy practice should be discouraged by all means from both the government and private sector. The National Veterinary Quarantine Service has an important role to play in preventing the transmission of poultry diseases across the border into Nepal.

INTRODUCTION

The Nepalese Poultry Industry took a big leap forward in the last 15 years. From a situation of early eighties when there were but a few ill-equipped, small poultry breeding and hatchery operations, we have attained the present day status where there are over 50 such enterprises in the country. Most of these units are well equipped with the state-of-art poultry breeding and hatchery technology. All the hatcheries maintain world famous highbred commercial breeding strains to produce good quality day-old chicks of white broiler and brown egg layer types.

The following are some of the major hatchery industries in Nepal:

- Pancharatna Hatchery, Chitwan
- Avinash Poultry Breeding Farm, Chitwan
- Siddhi Vinayak Hatchery, Chitwan
- Kalyan Poultry Breeding and Research Farm, Chitwan
- Ashapuri Hatchery Bhaktapur
- New Himalayan Hatchery, Thimi
- Subha Hatchery, Thimi
- Gaurishanker Hatchery, Banepa
- Siddhi Ganesh Hatchery, Bhaktapur
- Himalaya Hatchery, Kathmandu

¹ Pancharatna Feeds Pvt. Ltd. and Pancharatna Hatchery Pvt. Ltd., Narayangarh, Chitwan

² Srijana Poultry Pvt. Ltd., Thankot, Kathmandu

- Quality Hatchery, Balaju
- Srijana Poultry, Thankot
- Ratna Hatchery, Kathmandu
- Pravat Poultry, Bhaktapur
- Great Himalayan Hatchery, Chhauni
- Pashupati Hatchery, Biratnagar
- Panchakanya Hatchery, Pokhara
- Pratistha Hatchery, Butwal

The major strains of breeder flocks maintained by these hatcheries were as follows:

1. Brown Egg type strains: Lohmann, Hyline, Hisex, Keystone, Golden Comet, Isa Brown and Babcock.
2. White Broiler strains: Vencobb, Hubbard, Hubchicks, Arbor Acres, Ross, Avian and Anak-2000.

All the above strains of brown egg-type layer and white broilers are world class hybrids. Some of these breeder parent chicks were imported from India while others were imported from third countries.

DEMAND AND SUPPLY OF DAY-OLD CHICKS

The total chick production from all the hatcheries of Nepal combined, in 1997/98 was, on an average, 400 thousand chicks per week (20.8 million chicks per year). While this number was enough to meet the country's demand for day-old chicks, the local hatcheries' capacity remained significantly under utilized.

Since the day old chicks were produced and supplied by about 50 local hatcheries in the market, there was a very good competition for quality and price of chicks among them.

DUMPING OF DAY-OLD BROILER CHICKS FROM INDIA

Even though the local supply was sufficient to meet the country's demand for day-old chicks, a couple of big Indian hatcheries were flooding the Nepalese market with a large number of broiler chicks (Table 1). The weekly dumping of chicks from India during 1997/98 was 132 thousand chicks per week, which is 33 percent of the local supply. These hatcheries sold chicks in a very low price, often less than half the production cost. This trend was more pronounced during summer and monsoon season when the demand for chicks falls down in India.

The local entrepreneurs believed that the Indian hatcheries benefited from this action in two ways. Number one, they got at least some return on the chicks that they dumped in the Nepalese market. Number two, the smaller Nepalese hatcheries went bankrupt from this action leaving the local market under Indian control. Additionally, the oversupply of chicks had a serious long-term negative impact on Nepalese poultry industry, thereby increasing the dependency of Nepalese consumers on imported Indian poultry products.

Table 1. Dumping of broiler chicks in Nepalese Market from India (1997/98)

S No.	Name of Indian Hatcheries	Weekly	1997/98 Total
1	Arambagh Hatchery, Siligurhi, West Bengal	36,000	1,872,000
2	Himalayan Hatchery, Siligurhi, West Bengal	24,000	1,248,000
3	Phoenix Poultry, Jabalpur, Madhya Pradesh	24,000	1,248,000
4	Durga Hatchery, Gorakhpur, Utter Pradesh	36,000	1,872,000
5	Akash Hatchery, Utter Pradesh	12,000	624,000
	TOTAL Chicks (No.)	132,000	6,864,000

DIRECT LOSSES FROM CHICK DUMPING ON HATCHERY INDUSTRY

The direct effect of Indian chick dumping in the Nepalese poultry industry has been worked out in Table 2 and 3. Due to the over supply and the low price of dumped chicks, the local hatcheries were forced to sell chicks in prices lower than its production cost. It is estimated that during the one-year period the Nepalese hatcheries had a combined loss of Rs. 104 million (Table 2).

Table 2. Effect of dumping on Nepalese Hatcheries in 1997/98

Particulars	Value
Cost of production and marketing Broiler chicks	Rs. 17.0 / chick
Normal selling price of broiler chicks	Rs. 21.0 / chick
Average price of chicks during that period	Rs. 12.0 / chick
Net loss on every chick produced and sold	Rs. 5.0 / chick
Total chicks produced (number)	20.8 million
Total loss to local hatcheries	Rs. 104,000,000.0

There was a general belief among commercial broiler farmers that over supply of broiler chicks would bring extra benefit to them. While this philosophy may stand true in certain consumer good items, it was not so with the broiler. Over supply of chicks increased the production and supply of broiler in the market, thereby decreasing its selling price, quite often way below its production cost. The saving on cost of chicks was not enough to cover the losses due to over supply of broiler in 1997/98. The net loss to farmers for every kg live wt. broiler sold was Rs. 17.00 during that period (Table 3). There were other problems too that the farmers had to face. They could not pay the bank loan in time and therefore lost their good-will and credibility with financial institutions. They also had a big headache to find the live broiler traders. They often had to sell the birds to unreliable temporary slaughter facilities in credit.

A farmer with 1000 commercial broiler operation incurred a total loss of Rs 183,600 in 1997/98 as a result of Indian chick dumping. The total loss to the commercial broiler farmers as a result of this unhealthy practice was Rs. 1, 269.7 million (Table 3).

The total direct financial loss to hatchery industry (Table 2) and the commercial broiler industry (Table 3) combined came out to be Rs. 1,373,777,600 in one year. This is a huge loss to the country's economy.

Table 4. Effect of dumping on Nepalese Commercial Broiler Industry in 1997/98

Particulars	Value
Cost of Production of broiler (10 wk. old, 3kg Live Wt.)*	Rs. 65 / kg
Selling price of broiler during that period	Rs. 45 / kg
Total loss to farmers on broiler sale	Rs. 20 / kg
Saving to farmers on cost of chicks / kg live weight produced**	Rs. 3 / kg
Net loss to farmers on broiler sale	Rs 17 / kg
Total broilers chicks sold during that period (20,800,000 locally produced plus 6,864,000 Indian chicks)	27,664,000
Total loss to commercial broiler industry***	Rs. 1,269,777,600.00
Direct loss to a farmer with 1000 birds***	Rs. 45,900
In 4 cycles (flocks) of one year, total loss to the farmer	Rs. 183,600

*Cost of production of broiler at 7-wk. age was Rs 60/kg live wt, but the farmers had to wait until 10-wk. age due to over supply

**Chicks during that period were Rs 9 cheaper than the normal price (Table 1)

***No. of chicks x 90% livability x 3 kg live weight x Rs. 17 loss / kg

INDIRECT LOSSES FROM THE CHICKS DUMPING

The effect of this dumping was not limited to hatchery and commercial broilers only. There were other related institutions/industries that also suffered significantly. The effect of chick dumping on some of the other related institutions/industries were as follows:

A. Effect of chick dumping on local feed industries:

1. They were forced to sell feeds to farmers on credit
2. There was an increase of bad debt
3. Their working capital requirement increased significantly.

B. Effect of chick dumping on Chicken Marketing Industries:

1. There was an increased loss on live wt. of birds at slaughter house
2. They were forced to sell broiler meat on credit
3. There was an increase of bad debt
4. Their working capital requirement increased significantly.

C. Effect of chick dumping on Feed Ingredient Suppliers:

1. They were forced to sell feed ingredients to feed industry on credit

2. There was an increase of bad debt
 3. Their working capital requirement increased significantly.
- D. Effect of chick dumping on Financial Institution: There was no payment or delayed payment of loan and interest from clients:
1. Hatchery Industries
 2. Feed Industries
 3. Commercial Broiler Farmers
 4. Feed Ingredient Suppliers
- E. Effect of this dumping on Poultry Health:
1. There was import of new poultry diseases (Reo virus, Fowl Cholera, Marek's disease, Litchie disease, Hydropericarditis, etc.) into the country.
 2. There was high incidence of several poultry diseases
 3. There was a severe decrease in the productivity of broilers
- F. Effect of this dumping to the Country
1. There was huge loss to national economy/GDP
 2. It shattered the local poultry industry
 3. There was an increase in poverty and urban migration, social unrest.
 4. It increased dependency of local consumers on imported poultry products.

CONCLUSION AND RECOMMENDATION

The dumping of broiler chicks from India to Nepalese market hit very hard to the country's poultry industry in 1997/98. All the concerned parties should learn lesson from that crisis. The policy makers must immediately stop looking at this issue with "It is your problem" attitude. It is not the problem of one group only; it is rather a national problem. It is therefore recommended that:

1. The Government should constitute an independent autonomous body to look after the issues related to Nepalese poultry industry by involving both the government and private sector representatives.
2. Every effort should be exercised by the Government as well as the private sector to stop such an unhealthy practice from across the border.
3. The National Veterinary Quarantine Service must act swiftly to prevent the cross-border transmission of poultry diseases into Nepal.

Failure to tackle such problems effectively will have a serious, long lasting negative impact on the poultry industry of Nepal.

RELATED BIBLIOGRAPHY

Lohani, M. N., Bhattarai, T. C., and Amatya, J. L. (1996). Commercial Poultry Production in Nepal: Critical Overview. Paper presented in the 5th National Veterinary Conference September 11-13, 1996, BICC, New Baneshwor, Kathmandu.

Nepal Feed Industries Association, 1999. Personal Communication.

Nepal Hatchery Industries Association, Adhoc Committee, 1999. Personal Communication.

Pancharatna Feeds Pvt. Ltd., 1999. Personal Communication.

Pancharatna Hatchery Pvt. Ltd., 1999. Personal Communication.

Srijana Poultry Pvt. Ltd., 1999. Personal Communication.

COMPARATIVE PATHOGENECITY OF *FASCIOLA GIGANTICA* AND *FASCIOLA HEPATICA* IN SHEEP

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ABSTRACT

Four sheep experimentally infected with *Fasciola gigantica*, four with *F. hepatica* and four uninfected controls were monitored through clinical, parasitological, haematological, biochemical and pathological observations for 21 weeks. Two sheep, harbouring 80 and 181 *F. gigantica* died due to subacute fasciolosis at 14 and 15 weeks after infection respectively, while clinical chronic fasciolosis developed in one sheep which had a burden of 83 *F. gigantica*. On the other hand, *F. hepatica* infected sheep, even those with burdens of 203 flukes survived until the end of the experiment. Although, the numbers of the sheep infected with each *Fasciola* spp. were not large enough to draw a firm conclusion, there was an indication that liveweight gain in infected sheep was more severely depressed by *F. gigantica* than *F. hepatica*. From the results of this pilot experiment there was a clear evidence that *F. gigantica* is more pathogenic than *F. hepatica* in sheep.

INTRODUCTION

Among the several species of *Fasciola*, *F. hepatica* and *F. gigantica* are considered to be of major importance because fasciolosis caused by these two species is an almost worldwide problem. Fasciolosis due to *F. hepatica* occurs mostly in the temperate climate of Europe, North America, North Asia and Australia while the disease caused by *F. gigantica* is more prevalent in the tropical and sub-tropical environments of Africa and South and South-east Asia. There is a large amount of literature published on the pathogenesis of *F. hepatica* infection. However, relatively little comparable information has been published on *F. gigantica* infection and thus much of the work on *F. hepatica* are generally extrapolated to *F. gigantica*. Therefore, this pilot study was undertaken to compare the pathogenicity of *F. hepatica* and *F. gigantica* in sheep.

MATERIALS AND METHODS

Infective material

The *F. hepatica* metacercariae were purchased from the Compton Paddock Laboratories,

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Newbury, Berks, England while the *F. gigantica* metacercariae were obtained from naturally infected *L. natalensis* in Karatina, Kenya.

Experimental animals

Twelve sheep, 2 male and 4 female Blackface, and 4 male and 2 female Suffolk cross, all approximately 18 months old and raised under fluke-free conditions, were obtained from the Firth Mains Farm of Moredun Research Institute, Edinburgh. The sheep were dosed orally with oxfendazole (Systamex, Wellcome Foundation Ltd., London) at a rate of 5 mg/kg body weight to eliminate nematode worms, and thereafter housed in concrete pens provided with wood shavings as bedding. The sheep were fed a daily ration of 300 g cubes (dry matter 88%; protein 18%, oil 2.5%, fibre 10% and ash 8% of dry matter) per head. Chopped hay and drinking water were available *ad libitum*.

Experimental design

The sheep were divided into three groups of four animals so that each group was represented by an equal number of animals of the same breed and sex. Also, the mean liveweight for each group was more or less equal. The animals of each group were further sub-divided into two pairs. The two groups to be infected were selected at random and then designated as A and B. One pair from group A was infected with 150 and another with 350 metacercariae of *F. hepatica*. Similarly, the first pair from group B was infected with 150 and the second pair with 350 metacercariae of *F. gigantica*. Both pairs of sheep in group C constituted the uninfected control. Experimental details of the study are presented in Table 1.

Monitoring of the experiment

The animals were weighed at weekly intervals. A weighing crush (Weigh-bridge, Leslie P. Morris Ltd, Salop) was used to weigh the sheep.

Faecal samples were collected directly from the rectum and examined the same day for helminth eggs. The blood samples for haematological studies were taken from the jugular vein into vacutainers containing disodium or dipotassium ethylenediamine tetra-acetic acid (EDTA) as an anticoagulant and examined the same day. The blood samples for biochemistry and serology were obtained from the jugular vein into plain vacutainers (Becton, Dickinson & Co Ltd, U K) and allowed to clot in the incubator at 37°C for 1 hour, then kept at 4-6°C for 1 hour for the clot to retract. The serum was separated by centrifugation at 2,500 g for 20 minutes, dispensed into 1 ml aliquots and stored in labelled Eppendorf tubes frozen at -20°C until required. All sampling was carried out at weekly intervals between 0830 and 0930 hours.

Table 1. Experimental details of the study on pathogenicity of *F. hepatica* and *F. gigantica* infection in sheep

Group	Sub-group	Tag No.	Breed	Sex	Body weight (kg)	Metacercariae given (number)	Infective dose (metacercariae/kg)	
							Individual	Mean
A <i>F. hepatica</i> infected	A ₁ (lower dose)	24	Suffolk cross	F	44.5	150	3.4	3.2
		28	Suffolk cross	M	54.0	150	2.8	
	A ₂ (higher dose)	26	Black-face	F	29.0	350	12.6	11.3
		30	Black-face	M	35.5	350	9.9	
B <i>F. gigantica</i> infected	B ₁ (lower dose)	23	Suffolk cross	F	45.0	150	3.3	3.2
		27	Suffolk cross	M	48.5	150	3.1	
	B ₂ (higher dose)	25	Black-face	F	25.5	350	13.7	11.4
		29	Black-face	M	39.0	350	9.0	
C Uninfected	C ₁ (controls)	21	Suffolk cross	F	48.0	0	0.0	0.0
		22	Suffolk cross	M	53.0	0	0.0	
	C ₂ (controls)	31	Black-face	F	43.0	0	0.0	0.0
		32	Black-face	M	26.0	0	0.0	

RESULTS

Clinical data

All 8 infected sheep were clinically normal until 11 WPI. Thereafter, sheep 23, 27 (given 150 *F. gigantica* metacercariae) and 25 (given 350 *F. gigantica* metacercariae) had loss of appetite, abdominal distension and showed signs of anaemia. The condition of these sheep deteriorated progressively. Sheep 23 died 14 WPI whereas sheep 25 was slaughtered 15 WPI while in extremis. A day before death, both sheep were restless, unable to stand and had dyspnoea and subnormal body temperature. Except for sheep 26 (given 350 *F. hepatica*

metacercariae) which had loss of appetite around 10 WPI, none of the remaining 4 infected or any uninfected sheep showed clinical signs during the experimental period.

Liveweight data

Depression in liveweight of all the sheep seen at 5 WPI was due to shearing them the day before weighing. The infected sheep lost weight between 8 and 11 WPI and grew at a slower rate thereafter. However, the infected sheep gained weight over the experimental period, although less than the uninfected controls, except for sheep 27 (given 150 *F. gigantica* metacercariae) which lost 2.5 kg.

Parasitological data

The prepatent period for the *F. gigantica* infections ranged from 90 to 101 days while that for *F. hepatica* ranged from 68 to 84 days. Soon after patency, a peak in the faecal egg counts (EPG) was observed in *F. hepatica* infected sheep at 11 WPI. Thereafter, the EPG values rose gradually until the end of the experiment. In general, the faecal egg counts were higher in *F. hepatica* infected sheep than in the *F. gigantica* infected ones.

The recovery of flukes from *F. gigantica* infected sheep ranged from 9.7-55.3 %, the lowest percentage being recovered from sheep 29 which had been given a higher infective dose of 350 metacercariae. The recovery of flukes from *F. hepatica* infected sheep ranged from 51.3-70.6%. In sheep 23 and 25, more than 60% of the flukes were recovered from the liver parenchyma. Some ectopic flukes from sheep 25; 4 from the peritoneal cavity, 3 from the intestinal wall and 1 from the lungs were also recovered. In sheep 27, which was slaughtered at 20½ WPI, about 90% of the flukes were found in the bile ducts while remaining 10% were still in the liver parenchyma. All the flukes recovered from sheep 29 were adults and in the bile ducts. All the *F. hepatica* were adults when recovered from the livers of infected sheep at 20½ WPI, except for sheep 26 in which a few flukes were still migrating in the liver parenchyma. No flukes were recovered from the livers of any uninfected controls.

Pathological data

Lesions were widespread in sheep 23 and 25. The carcasses of these sheep were pale; the peritoneal cavity contained large quantities of blood-stained fluid and several blood clots; the small and large intestines were filled with blood and blood stained ingesta. The livers were enlarged with prominent haemorrhagic tracts and with areas of fibrinous peritonitis on the surface. The gall bladders were enlarged and filled with clotted blood. The liver of sheep 23 was paler in colour than that of sheep 25 and there was a large blood clot between the liver capsule and the parenchyma. The lungs of sheep 25 showed large areas of congestion, these areas contained typical haemorrhagic tracts due to a migrating fluke which was found in the subpleura and subpleural oedema was prominent.

The remaining infected sheep had typical lesions of chronic fasciolosis with enlarged livers and bile ducts distended with flukes.

Haematological data

All the infected sheep showed a drop in RBC counts after 8 WPI. In sheep 25, the decline became evident as early as 4 WPI. Packed cell volumes and haemoglobin concentrations of these sheep also appeared to follow similar pattern of decline. These declines were more pronounced in *F. gigantica* infected sheep, except for sheep 29 which harboured the lowest number of flukes.

The MCV values indicated a macrocytic anaemia in all the infected sheep. MCH levels in the infected sheep also appeared to rise. However, values for the MCHC showed no changes as a result of either *F. gigantica* or *F. hepatica* infections.

There was a leucocytosis from the 2nd week after infection in most of the sheep, the most pronounced being in sheep 25 with a peak at 5 WPI. In sheep 29, the total WBC counts were at their highest levels 12 weeks after infection.

The eosinophil counts rose by the 2nd week after infection and reached maximum levels shortly after that, except in the case of sheep 26, 28 and 29 where the highest peaks were not reached until 9, 10 and 12 WPI respectively. Following the first peak there was a gradual but irregular fall in values, except in sheep 29 where the eosinophil count fell very rapidly during the following 3 weeks.

Biochemical data

The serum GLDH values for the uninfected controls (Nos 21, 22, 31 and 32) remained at a uniformly low level, but those of all the infected sheep showed a rise which commenced by 2nd WPI. The maximum level was reached 3-5 WPI, except in the case of sheep 24, 28 and 29 where the highest peaks were not reached until 10, 9 and 12 WPI respectively. Sheep 27 and 28 showed a smaller rise than the other infected sheep, and this was followed by an earlier fall to reach the level of the uninfected controls by 15 and 12 WPI respectively. Sheep 29 however showed a sharp rise from 10 WPI reaching a highest level of 375.3 U/l at 12 WPI, but the level fell rapidly during the following 2 weeks.

Although a slight transient rise in serum GGT levels in the infected sheep were recorded between 3 and 5 WPI, this was not pronounced until 8-10 WPI in the sheep infected with *F. hepatica* or 11-12 WPI in the sheep infected with *F. gigantica*, except in the case of sheep 25 where the rise was delayed until 14 WPI. The maximum levels of serum GGT in the *F. hepatica* infected sheep were reached at 10 WPI while those in the *F. gigantica* infected sheep were reached 12-13 WPI. Thereafter, the levels declined gradually and reached to those of uninfected controls by 15, 17 and 18 WPI in sheep 28, 27 and 26 respectively. However, other sheep showed higher levels until the end of the experiment.

DISCUSSION

The conclusions that may be drawn from this experiment are severely limited by the individual variation and the small numbers of experimental animals involved. There was a clear evidence, however, that *F. gigantica* is more pathogenic than *F. hepatica* in sheep. Two

sheep, harbouring 80 (1.7 flukes/kg of initial liveweight) and 181 (7.0 flukes/kg) *F. gigantica* died due to subacute fasciolosis at 14 and 15 WPI respectively, while clinical chronic fasciolosis developed in one sheep which had burden of 83 (1.7 flukes/kg) *F. gigantica*. On the other hand, *F. hepatica* infected sheep, even those with burdens of 203 (5.7 flukes/kg) and 247 (8.5 flukes/kg) flukes survived until the end of the experiment, and only one sheep which had a burden of 247 (8.5 flukes/kg) flukes showed transient clinical signs characterised by loss of appetite around 10 WPI.

These findings supported the views of Sogoyan (1956) and Ogunrinade (1984) that the number of parasites required to present the same type of fasciolosis syndrome in the same host species is lower for *F. gigantica* than *F. hepatica*. Sogoyan (1956) compared the pathogenicity of *F. gigantica* and *F. hepatica* in sheep and found that 10 sheep harbouring 33 to 265 adult *F. gigantica* died between 77 and 88 days after infection, whereas infection of 122 to 870 adult *F. hepatica* did not result in mortality in sheep for up to 156 days. In the study conducted by Ogunrinade (1984), 100 to 300 *F. gigantica* caused fatalities from the acute fasciolosis syndrome 11 to 12 weeks after infection, while Pullan et al. (1970) could only produce acute fasciolosis in sheep with burdens in excess of 1,400 *F. hepatica*. Likewise, Ross et al. (1967) found that 1,000 to 2,500 *F. hepatica* were needed to produce acute fasciolosis in sheep. Ogunrinade (1984) suggested that the longer period of migration of *F. gigantica* in the liver together with its larger size probably accounts for the greater pathogenicity of this species than of *F. hepatica* in small ruminants.

At 12 WPI, there was no gain in the mean liveweight of *F. gigantica* infected sheep which had mean burdens of 94.5 flukes (2.3 flukes/kg) while *F. hepatica* infected sheep with mean burdens of 155.5 flukes (3.8 fluke/kg) gained 1.125 kg during the same period. At the end of the experiment (20½ WPI), two *F. gigantica* infected surviving sheep which had a mean burden of 58.5 flukes (1.3 fluke/kg) gained only 1.500 kg of mean liveweight in comparison to 3.750 and 7.275 kg gained by *F. hepatica* infected and uninfected controls respectively. Thus, the results of the present study indicate that liveweight gain in infected sheep is more severely depressed by *F. gigantica* than *F. hepatica*. It should be emphasised, however, that the numbers of the sheep infected with each *Fasciola* spp. were not large enough to draw a firm conclusion on this point.

ACKNOWLEDGMENTS

The present work formed part of the first author's PhD Thesis which was submitted to the University of Edinburgh in 1993. The work was carried out at the Centre for Tropical Veterinary Medicine (CTVM), Edinburgh, Scotland with the financial assistance of Overseas Development Administration (ODA), UK. We are thankful to the ODA for providing funds for the study, Dr. M.M.H. Sewell, Director, CTVM for providing facilities and Mr. H.R. Urquhart and Mrs. E. Moore, Helminthology Section, CTVM, for their technical assistance.

REFERENCES

- Ogunrinade, A.F. (1984). Infectivity and pathogenicity of *Fasciola gigantica* in West African dwarf sheep and goats. *Tropical Animal Health and Production*, **16**: 161-166.
- Pullan, N.B., Sewell, M.M.H. and Hammond, J.A. (1970). Studies on the pathogenicity of massive infections of *Fasciola hepatica* L. in lambs. *British Veterinary Journal*, **126**: 543-547.
- Ross, J.G., Dow, C. and Todd, J.R. (1967). A study of *Fasciola hepatica* infections in sheep. *Veterinary Record*, **80**: 543-546.
- Sogoyan, I.S. (1956). Comparison of pathological changes caused in sheep by *Fasciola hepatica* and *F. gigantica*. *Trudy Armyanskogo Nauchno-issledovatel'skogo Veterinarnogo Instituta*, **1**: 113-117. [In Russian]

THE ROLE AND IMPORTANCE OF EPIDEMIOLOGY IN NATIONAL VETERINARY SERVICES

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ABSTRACT

With increasing pressures on veterinary services both internally and externally, it is more important than ever that reliable data be readily available on the disease situation within Nepal and that decisions on the allocation of resources be based on a realistic analysis of this.

The first step is to collect data, the second is to transform this into useful usable information and the third is to base decisions on the basis of this information. An epidemiology unit has a vital role to play in all of these steps by

- 1. co-ordinating and encouraging accurate data collection at field level and its transmission to a central level*
- 2. Recording and analysing the data*
- 3. Communicating the results of this analysis to district, regional, national and international bodies*
- 4. Interpreting the results for national authorities and advising on disease control methods to be applied in the light of such information.*

Data can be either passively or actively collected and each has its role within the system. This paper will detail the role and functions of an epidemiology unit in the light of current requirements of both national and international authorities.

WHAT IS EPIDEMIOLOGY?

Epidemiology is the study of disease in populations. It is an ancient art first practised by the Greeks (Hippocrates) so far as we know. Epidemiologists are specialists in their field but as importantly build and maintain links with other institutions. Epidemiologists aim to look at The pattern of disease in a population.

Importance of epidemiology

Epidemiology tries to look at the complete picture and to relate disease to the overall population and its productivity. A disease which kills 10 animals is important to an individual farmer, but is not necessarily important for the country unless it affects many other farmers in the same or different districts.

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How is epidemiology done?

However complicated epidemiology may be at times, it is always basically about the five questions and developing solutions to disease problems based on the answers to these.

The five key questions are:

- Which animals (age, sex, breed, usage)?
- How many animals?
- How badly affected i.e. how many sick, how many dead
- Where are they?
- When did the problem start and how has it developed over time?

Additionally, epidemiology attempts to answer the following questions:

- What is the impact of a disease on production?
- What is the impact of a disease on human health?
- What are the most important diseases in these terms?
- Which diseases can be controlled effectively given the resources available?

WHAT SORT OF EPIDEMIOLOGY UNIT DOES THE STATE VETERINARY SERVICE OF NEPAL NEED?

Epidemiology is a key area for government veterinary services and the epidemiology section has several roles. The Epidemiology Unit should act as a support to decision makers, offering information and advice so that these senior veterinarians have the facts and time required to consider all options and chose the most effective and appropriate. Its activities should include

- 1) Routine collection and analysis of disease data
 - a) Passive surveillance
 - b) Active surveillance
- 2) Advice on importance and impact of diseases on animal health and production
- 3) Co-ordinate follow up and investigation of telephone / fax disease outbreak reports
- 4) Co-ordination of response to acute disease outbreaks
- 5) Advice on the allocation of resources to maximum effect
- 6) Advice on formulation of disease regulations
- 7) Formulation and supervision of the implementation of disease control measures

- 8) Design and co-ordination of special programs e.g. rinderpest sample survey
- 9) Advice on risk of disease importation and response to WTO related queries

Within the present structure of the DLS and AHD, all of this should be under the direct control of the Chief of the Animal Health Division and must include close collaboration with the Quarantine and Laboratory Sections of the AHD.

The level of responsibility and lines of communication make it clear that the head of the CEU should be of equal status to the other section chiefs within the AHD and report directly to the chief of the AHD.

The CEU should be established at the vet complex in Tripureswor. However, when there are suspected or actual outbreaks, it may not always be practical for the response to come directly from the CEU. The CEU should therefore be represented at a lower level of HMGN structure. It is not possible or necessary to have an epidemiologist in each district. Therefore, it is proposed that there should be an epidemiologist in each region based in a Regional Epidemiology Unit (REU).

The regional epidemiologists would be responsible for quality checking monthly reports received from districts and chasing up any reports not received. In the future, they could be responsible for the running of the VAHS in each region as well. It is hoped that eventually for both sets of data the regional office could enter the data themselves and then send it to the CEU as an e-mail attachment which could then be imported straight into the appropriate database. However, this is too ambitious initially. The regional office would also be responsible for initial response to reports of disease outbreaks, informing the CEU of the same, and co-ordinating any control measures at regional level.

For active surveillance and many other tasks, the CEU should co-ordinate closely with the central laboratory services and in the regions with the regional lab services. For this purpose it might seem best to locate the regional units in the regional labs. Whilst this has a measure of convenience, a stronger case can be made for locating them in the Regional Directorates so that they have rapid access to the RD in case of disease outbreak reports, enabling rapid mobilisation of human and physical resources.

OUTPUTS FROM EPIDEMIOLOGY SECTION

An essential function of any epidemiology unit is to communicate information both upwards to higher level officers and downwards to those responsible for collecting the raw data which the CEU uses. This downward communication is important for several reasons. Most importantly, it gives a reason for collecting the data if those doing so benefit directly and tangibly from it. Secondly, by communicating the failures of some districts as well as the successes of others it can act as an active spur to better compliance. It is proposed that all routine and some special reports should be sent to each district as well as to DLS and regional authorities.

Over the last year, quarterly and annual reports have been produced as described below. These formats have gradually become fixed and their production can now be made semi-automatic.

- 1) National Annual report
These are already being produced and the format is concise and clear although lacking in details. There are also clear problems of data quality and data checking. These reports should be sent to each district and regional office as well as used at the central level
- 2) National Quarterly Reports
These are produced showing disease by ecozone within region i.e. splitting the country into 15 areas (3 ecozones for each of 5 regions). This is sufficiently detailed and makes epidemiological sense.
- 3) Quarterly Regional Reports
For each region, a quarterly report showing disease occurrence by district is being produced.
- 4) District level reports: Annual
It is proposed that each district should receive a report on its own disease situation every year
- 5) OIE monthly, quarterly and annual reports
The quarterly report is for List A and List B diseases (see appendix for full list and codes) and the annual report covers these and also List C diseases (see appendix for full list and codes). There are standard formats and these should be built into the national disease monitoring database. The Epidemiology Unit is currently producing these reports when requests are received from the OIE.
- 6) Special reports
At the request of the AHD. The routine reports will indicate which are the commonest diseases and give some idea of their overall impact in terms of mortality. Where a particular disease is of great importance, a special report on this disease could be compiled.
- 7) Reports on Active Surveillance activities
During the lifetime of the SVSLDC project, surveillance of village animal health is to be carried out. This should expand to include 15 sites (3 per region, one in each broad agro-ecological zone).

Stripping sample is defined as the fraction of the quarter milk collected when the milking is completed.

The purpose of this study was to examine the feasibility of the use of foremilk and stripping samples for the diagnosis of subclinical mastitis in a buffalo population, and determine the relationship between lactose content, cultural examination, and viable and/or differential cell counts of foremilk and stripping samples in the diagnosis of subclinical mastitis in a buffalo population.

MATERIALS AND METHODS

Study population

Fifty apparently healthy lactating buffalo stationed at the Buffalo Research Centre, Hisar - India were included in this study. To our knowledge these buffaloes represent the average water buffalo population in most of the Indian subcontinent. Approximately 10 ml each of foremilk and stripping samples were collected aseptically from each quarter following the USA, National Mastitis Council (NMC) procedures. Information related to animal identification, previous date of calving, and date of collection of milk samples was recorded for each individual animal. A total of 200 quarters were included in this study population.

Test procedures and criteria

Somatic cell count (SCC) was performed as described by Schalm et. al. (1971) by using Newmans Lampert stain instead of Broadhurst-Paley stain. Culturing for bacterial species was by following procedures recommended by NMC (1987). Aseptically collected milk samples were streaked on nutrient agar, MacConkey lactose agar, and 5% bovine blood agar plates. The plates were incubated at 37°C for 24-48 hrs. The cultures were purified and identified on the basis of Gram's reaction, morphology and colony characteristics. *Staphylococcus* species were identified on the basis of rabbit plasma coagulase test. *Streptococcus* species were identified biochemically on the basis of fermentation of raffinose and inulin, hydrolysis of esculin and sodium hippurate, reduction of 0.1% methylene blue milk and CAMP reaction (McDonald and McDonald, 1975).

A subclinical mastitic quarter was defined as a quarter having SSC > 500,000/ml and was culture positive. However, latent infection in the quarter samples was indicated by normal SSC and a positive bacteriological test.

Viable cell count (VCC) was performed on each fraction of quarter milk samples. The Jain and Jasper procedure (1967) was followed for its determination. The samples were prepared following the above reference. Trypan blue stain was used in enumeration of viable cells in milk. Unstained and stained cells were counted in four corners primary squares of the hemocytometer and the percentage of both unstained and stained cells were calculated. Viable cells appeared as colorless, refractile, and round objects containing nuclei. The dead cells were stained blue.

Differential cell count (DCC) was performed by using the technique described by Schalm et. al. (1971). Preparation of milk film was employed following the above reference. Giemsa's stain (Merck) was used in counting the DCC in both foremilk and stripping milk samples.

Lactose estimation was calculated using the method described by Feitosa Teles et. al. (1978). A standard curve was prepared by plotting the graph against the optical density and mg lactose per ml. Then gram of lactose per 100 ml of the milk samples was estimated from the standard curve.

California mastitis test (CMT) was performed as a diagnostic test on each fraction of quarter milk samples. The Pandit and Mehta (1969) procedure was followed in the performance of this test. The reagent was prepared locally following the above reference. The reaction was classified as negative, trace, 1, 2 and 3 as strong positive.

Statistical procedures

The student test and one-way analysis of variances were calculated to detect statistical differences among the means of two groups or more than two groups respectively. Person bivariate correlation was calculated to determine the quantitative linear relationship between lactose content and the blood cell types.

RESULT AND DISCUSSION

Of the 50 apparently healthy buffaloes examined, 74 percent were culturally positive, 32 percent had SCC > 500,000/ml, 22 percent had subclinical mastitis and 52 percent were having latent infection (Table 1). Infection rate in our study was higher than that reported by Mishra (1976). On a quarter basis 26 percent, 32 percent and 15 percent of the quarters were culturally positive in foremilk, in stripping samples and both fractions respectively. However, only 12, 13 and 9 percent of the quarters had SCC > 500,000/ml with foremilk, stripping samples and both the fractions respectively. Our findings from foremilk samples are in agreement with results reported by Kalorrey et. al. (1983).

Table 1. Prevalence of subclinical/latent mastitis as detected by CMT, SCC and cultural examination.

Parameters	Animals tested (n=50)	Quarters tested (n=200)		Foremilk and stripping
		Foremilk	Stripping	
CMT positive	12 (24)	19 (9)	17 (8)	11 (5)
SCC positive	16 (32)	25 (12)	27 (13)	19 (9)
Cultural positive	37 (74)	52 (26)	64 (32)	31 (15)
SCC positive and cultural positive	11 (22)	15 (7)	13 (6)	10 (5)
SCC negative and cultural positive	26 (52)	37 (18)	51 (25)	20 (10)

CMT = California Mastitis Test, SCC = Somatic Cell Count

Figures within parentheses indicate the approximate percentages out of the total samples.

On the basis of the International Dairy Federation Criteria 7.50 and 6.50 percent of the quarters with foremilk and stripping samples respectively were diagnosed as subclinical mastitis (Table 1). However, latent infection was observed in 18.50 and 25.50 percent of the quarters examined with foremilk and stripping samples respectively. Only 5 and 10 percent of the quarter milk samples were positive for both subclinical and latent mastitis infection with foremilk and stripping samples respectively. Schalm and Lasmanis (1968) observed that mammary quarters in cows with advanced mastitis were positive in all the fractions. Milder mastitis was detected to a much greater extent with stripping samples than with foremilk samples. On the basis of a single test, a maximum number of buffaloes was found positive by cultural examination (74 percent) followed by SCC (32 percent) and CMT (24 percent).

The mean SCC on foremilk and stripping milk samples of normal and subclinical mastitic quarters are presented in Table 2. In normal milk, SCC was higher in stripping ($1.12 \times 10^6/\text{ml}$) than in foremilk ($0.096 \times 10^6/\text{ml}$) with a significant difference ($P < 0.01$). In subclinical mastitic milk, SCC was found higher in stripping samples ($3.25 \times 10^6/\text{ml}$) than in foremilk ($2.30 \times 10^6/\text{ml}$). However, the differences were not statistically significant ($P > 0.05$). There was a statistically significant difference in the SCC of normal and mastitic fractionate milk ($P < 0.01$). Similar observations in cows were reported by Paape and Tucker (1966) and Schalm and Lasmanis (1968). Variation in the cell numbers in different fractions of milk could possibly be due to the fact that polymorphonuclear (PMN) leucocytes and sloughed epithelial cells are retained in the alveoli as pressure from accumulation of milk increases. When milk is removed, the pressure falls and cells are released into the stripping samples.

Table 2. Mean $\pm$ S.D. of somatic cell count (SCC) and viable cell count (VCC) on foremilk and stripping of normal and subclinical mastitic quarter.

Milk fraction	Normal milk			Mastitic milk*		
	Quarters	SCC ($10^6/\text{ml}$)	VCC ($10^6/\text{ml}$)	Quarters	SCC ($10^6/\text{ml}$)	VCC ($10^6/\text{ml}$)
Foremilk	138	0.096 ± 0.06^a	0.14 ± 0.04^a	15	2030 ± 1.95^b	0.45 ± 0.21^b
Stripping	122	1012 ± 0.07^c	0.08 ± 0.03^a	13	3025 ± 2.05^b	0.43 ± 17^b

S.D. = Standard deviation.

* Mastitic milk is defined as a milk sample containing somatic cell count 500,000/ml and a bacterial culture positive.

Values with different superscripts within each class (VCC or SCC) were significantly different ($P < 0.01$) in t-test.

Staphylococcus species were found to be the most prevalent organisms accounting for 41.5 percent in the foremilk and 61.38 percent in stripping samples (Table 3) of the total 120 isolates. *Streptococcus* species were the second most prevalent species with 24.52 and 41.5 percent respectively. Similar results were reported by Kalorey et. al. (1983) and Yas et. al. (1983) in only foremilk samples. *Staphylococcus epidermidis* was the most prevalent whereas *staphylococcus aureus* was detected in a lesser degree from both foremilk and stripping samples. Our findings from the foremilk samples agreed with reported results by Rahman et. al. (1983). However, Kalorey et. al. (1983) reported a high infection rate of

Staph. aureus (45.60 percent) as compared to *Staph. epidermidis* (31.70 percent). Among the *Streptococcus* species isolated from both the foremilk and stripping samples, *Streptococcus bovis* was most prevalent, *Strep. dysgalactiae* was second, and unclassified streptococci accounted for the least number of isolates. Kalra (1967) found a higher incidence of *Strep. dysgalactiae* in the infected quarters of buffalo.

Table 3. Relative frequency of different types of microorganisms in 120 isolates from 116 apparently healthy quarters.

Microorganisms	No. of isolates		Foremilk and stripping
	Foremilk	Stripping	
<i>Staph. epidermidis</i>	9 (17)	7 (10)	4 (3)
<i>Staph. aureus</i>	13 (24)	27 (51)	4 (3)
<i>Micrococci</i>	10 (19)	6 (11)	2 (2)
<i>Strep. dysgalactiae</i>	4 (7)	3 (6)	3 (2)
<i>Strep. bovis</i>	9 (16)	17 (32)	2 (2)
Unclassified <i>Streptococci</i>	-	2 (4)	-
Others: <i>E. coli</i> , diphtheroids etc.	8 (15)	5 (9)	2 (2)
Total	53	67	17

Figures within parentheses indicate the approximate percentages out of the total isolates for each column.

The numbers of VCC in both foremilk and stripping samples were not significantly different (Table 2). However, VCC was found significantly higher ($P < 0.01$) in mastitic milk than the normal milk in both the fractions. Similar findings in cows were reported by Jain and Jasper (1967). The higher percentage of viable cells in milk from an infected quarter may reflect an inflammatory response to counteract *in-vivo* bacterial multiplication. There was a positive correlation between total cell count and bacterial counts in both fractions of the milk. An $r = 0.85$ was calculated between foremilk and total cell count and $r = 0.40$ was calculated between stripping samples and total cell count.

In normal milk, epithelial cells both in foremilk (48 percent) and stripping samples (47 percent) were predominant. Lymphocytes, neutrophils and monocytes were also seen (Table 4). Epithelial cells are present in normal milk as a result of the normal breakdown and repair but it increases in late lactation as a consequence of injury due to mastitis. Elevated epithelial cell count in cow's milk cannot be considered to be pathognomic for subclinical mastitis as suggested by Tolle et. al. (1968). In our study, neutrophils were found in approximately 20 percent of both fractions of normal milk. However, Reiter (1978) reported that milk from healthy udders of cows contained less than 8 percent neutrophils.

Among the subclinical mastitic milk, neutrophils were predominant both in foremilk samples (67 percent) and stripping samples (76 percent) followed by lymphocytes, epithelial cells and monocytes. Neutrophil count was significantly ($P < 0.01$) higher in mastitic than in normal milk of both fractions. This is in agreement with the earlier observations of Badran (1987) in

cow's milk. The number of neutrophils observed was relatively low in foremilk than in stripping samples. The relatively low number of neutrophils in the foremilk may probably be due to the pressures within the filled udder that prevents epithelial damage resulting from mechanical stress such as intracellular biosynthesis of fat as suggested by Feldman (1961). Schalm et al. (1971) pointed out that neutrophils were first to appear in the cow's milk in significant numbers at the beginning of the mastitic process even though there may not be a significant elevation in total cell count. They also suggested that epithelial cells might be shed in large numbers in mastitis but their relative proportions still remains low due to an overwhelming number of neutrophils infiltrating from the blood.

Table 4. Mean $\pm$ S.D. of differential cell counts and lactose content on fraction normal and subclinical mastitic milk.

Parameters	Normal milk		Mastitic milk	
	Foremilk	Stripping	Foremilk	Stripping
Sample size	138	122	15	13
Neutrophils	20.98 $\pm$ 17.08 ^b	19.59 $\pm$ 15.07 ^b	67.33 $\pm$ 23.44 ^a	76.38 $\pm$ 9.30 ^a
Lymphocytes	29.28 $\pm$ 11.12 ^a	32.59 $\pm$ 16.28 ^a	20.40 $\pm$ 11.20 ^b	16.69 $\pm$ 5.57 ^b
Epithelial cells	48.42 $\pm$ 15.82 ^a	46.85 $\pm$ 17.84 ^a	10.80 $\pm$ 13.68 ^b	5053 $\pm$ 5.58 ^b
Monocytes	1.62 $\pm$ 0.75 ^a	1.72 $\pm$ 0.77 ^a	2.10 $\pm$ 1.37 ^a	1.50 $\pm$ 0.64 ^a
Lactose content (g/100 ml)	4.25 $\pm$ 0.39 ^a	4.18 $\pm$ 0.39 ^a	3.62 $\pm$ 0.51 ^b	3.31 $\pm$ 0.48 ^b

All the values in a row with a different superscript were significantly ($P < 0.01$) different using the one way analysis of variance.

The DCC on normal foremilk and stripping samples were not significantly different ($P > 0.05$). However, the differences were found significant ($P < 0.01$) between normal and mastitic milk in both fractions.

Lactose content of normal foremilk (4.25 gm/100 ml.) was higher than that of stripping samples (4.18 gm/100 ml.) (Table 4). Most likely it may be due to the high fat content of stripping samples (Schalm et al. 1971). In mastitic cow's milk lactose content was found significantly decreased, possibly because of synthesis reduction (Schultz et al. 1978). Samples which were culturally positive were found to have a lower lactose content than the culturally negative samples. In our study, lactose content was significantly lower in CMT positive samples. As the CMT reaction increased the lactose content got lower. This finding supports the earlier observation of Sabbag et al. (1981) in cow's milk.

A negative correlation ($r = -0.37$) was observed between lactose content and the total cell count. Singh and Singh (1980) reported that lactose content was inversely related to the cell count in buffalo milk. Neutrophil and VCC were negatively correlated with lactose content, $r = -0.40$ and -0.39 , respectively. However, epithelial cell count was positively correlated with lactose content of $r = 0.43$. These findings are contrary to the findings of Hassan et al. (1984). They reported that both neutrophil and epithelial cell counts were negatively correlated with the lactose content.

In conclusion, it is imperative to make an early diagnosis of subclinical cases with reliable and practical tests to achieve a successful mastitis control program. In the diagnosis of subclinical mastitis, the type and time of sampling play an important role in the reliability and validity of a diagnostic test. The choice between foremilk or stripping samples for an early diagnosis of mastitis in buffaloes is an important issue specifically in developing countries where sources are limited. Our study has indicated that stripping samples detected more subclinical cases than foremilk samples. In addition, the study suggested that VCC, DCC, and lactose content were more reliable in detecting subclinical cases than the use of SCC.

REFERENCES

- Badran A.E. (1987). Milk conductivity and its use for detection of mastitis. *Indian Vet. J.*, **64**: 733-737.
- Feitosa Teles F.F.; Cheryl, K.V. and Stull J.W. (1987). A method for rapid diagnosis of lactose. *J.Dairy Sci.*, **61**: 506-508.
- Feldman, J.D. (1961). Fine structure of the cows udder during gestation and lactation. *Lab. Invest.*, **10**:238-255.
- Giesecke, W.H. and Van Den Heever L.W. (1974). The diagnosis of bovine mastitis with particular reference to subclinical mastitis. A critical review of relevant literature. *Onderstepoort J. Vet. Res.*, **41**:169-212.
- Hassan, G.A.; Badran, A.E. and Sharaby M.A. (1984). Susceptibility of cows and buffaloes to mastitis infection. I. Milk cells and mastitis detection tests. *Indian Vet. J.*, **61**: 480-485.
- Jain, N.C. and Jasper, D.E. (1967). Viable cells in bovine milk. *Br. Vet. J.*, **123**: 57-62.
- Kalorey, D.R.; Purchit, J.H. and Dholkia, P.M. (1983). Studies on the incidence of subclinical mastitis, its etiology and invitro sensitivity of isolates. *Indian J. Anim. Sci.*, **53**: 961-963, 1983.
- Kalra, D.S. (1967). Observations on the incidence of mastitis in relation to predisposing factors. *PAU J. Res.* **4**: 249-256.
- National Mastitis Council (1987). *Current concepts of bovine mastitis*. 3rd edition Arlington, V.A. 22201.
- McDonald, J.S. and McDonald, J.J. (1975). Differentiation of streptococci recovered from bovine intramammary infections. *18th Annual Proceedings, American Association of Veterinary Laboratory Diagnosticians*, pp. 251-256.
- Mishra, D.S. (1976). Bacterial buffalo mastitis. *Pantnagar J. Res.*, **1**: 67-69.

- Paape, M.J. and Tucker HA (1966). Somatic cell variation in fraction collected milk. *J. Dairy Sci.*, **49**: 265-267.
- Pandit A.V. and Mehta ML (1969). Sodium lauryl sulphate as substitute for CMT reagent (California Mastitis test reagent) for diagnosis of subclinical mastitis in buffaloes. *Indian Vet. J.*, **46**: 111-119.
- Rahman, H.; Sambyal, D.S. and Baxi, K.K. (1983). Incidence and etiology of subclinical mastitis in cows and buffaloes in Punjab. *J. Res. Punjab Agric. Univ.*, **20**: 208-212.
- Reiter, B. (1978). Review of the progress of dairy science antimicrobial system in milk. *J. Dairy Res.*, **45**: 131-147.
- Sabbag, N.G.; Esponda, J.C.; Freyre, M.R.; Sbodio, O.A. and Weidman, P. (1981). Physicochemical parameters of technological interest in normal and mastitis milk. *Revista del Institute de Tecnologia de Alimentos*, **3**: 99-106 (Abstr. Dairy Sci. **44**: 5682).
- Schalm, O.W.; Carroll, E.J and Jain, N.C. (1971). *Bovine mastitis*, 1st. ed. Lea and Febger, Philadelphia, pp. 76-77.
- Schalm, O.W. and Lasmanis, J. (1968). The Leucocytes origin and function in mastitis. *J. Am. Vet. Med. Assoc.*, **153**: 1688.
- Schultz, L.H.; Brown, R.W.; Jasper, D.E.; Mellenburger, R.W.; Natzke, R.P.; Philpot, W.N.; Smith, J.W. and Thompson D.D. (1978). Current concepts of bovine mastitis. *National mastitis council. Inc.* Washington, D.C., pp. 8.
- Singh, S.P. and Singh, R.S. (1980). Somatic cell count an index of quality for milk and milk products. *Indian Dairyman*, **32**: 547-548.
- Sinha, M.N. and Prasad, B.M.L. (1969). Observation on etiological agents and diagnosis of bovine mastitis. *Indian J. Anim. Hlth.* **8**: 171-174.
- Tolle, A.; Zeidler, H.; Heeschen, W.; Kraack, J.; Reichmuth, J.; Kieler (1969). *Milchw. Forschber*, **20**: 343-394 (Cited by Giesecke and Van Den Heever, 1974).
- Yas, A.A.; Kalra, D.S. and Khalaf, A.M. (1983). Studies on mastitis in buffaloes in Iraq 1. prevalence rate and etiology. *Tropical Vet. Anim. Sci. Res.*, **1**: 23-28 (Abstr. Dairy Sci. (**45**): 7991).

REPORT ON AN OUTBREAK OF PPR IN GOATS AT ARS, BANDIPUR

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ABSTRACT

During August 1998, an outbreak of a disease in goats was reported at Agricultural Research Station (ARS), Bandipur of Tanahu district. A detailed investigation of the outbreak was carried out immediately and pathological samples were collected for the laboratory investigation. Laboratory investigation was carried out at Animal Health Research Division and Central Veterinary Laboratory, Tripureshwor, Kathmandu. For further laboratory confirmation the pathological samples were sent to the Institute of Animal Health (IAH), Pirbright, UK. The overall morbidity and mortality rates were 46% and 33% respectively. The results of the clinical findings and post-mortem examination were suggestive of Peste Des Petits Ruminants (PPR). Laboratory findings also confirmed the disease to be PPR. Serum samples from affected goats were tested positive for PPR antibodies by monoclonal antibody based competitive ELISA for PPR both at the Central Veterinary Hospital, Kathmandu and at IAH, Pirbright. PPR virus specific polymerase chain reaction test conducted at IAH, Pirbright on the pleural fluid and lung samples were found positive for PPR virus.

INTRODUCTION

Peste des petits ruminants (PPR) is a highly contagious, systemic disease of goats and sheep very similar to rinderpest and caused by a closely related morbillivirus. After an incubation period of 5 to 6 days, clinical signs develop which include fever, anorexia, necrotic enteritis, gingivitis, diarrhoea and respiratory signs including pneumonia. The course of the disease may be peracute, acute or chronic. However, the virus does not persist in recovered animals. The first outbreak of PPR in Nepal was reported from Dhanusha, Bara, Parsa, Rautahat and Sarlahi districts of central region of Nepal (Jha et al., 1995). The morbidity and mortality rates recorded in the first outbreak was 82.6% and 33.8% respectively. The present report deals with an outbreak of PPR in goats at Agricultural Research Station (ARS), Bandipur of Tanahun district of Nepal.

MATERIALS AND METHODS

During August 1998 an outbreak of a disease in goats was reported from Agricultural Research Station (ARS), Bandipur of Tanahu district. Investigation was done immediately after receiving the reports of the outbreak. During the investigation clinical examination of the affected goats was carried out thoroughly. Furthermore, twelve dead goats were post-

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mortem. Epidemiological data such as total population of goats at the affected farm, number of goats affected, recovered and dead at the farm was recorded. Blood, serum samples and nasopharyngeal swabs from the sick goats were collected. Similarly, lymph nodes, lungs, spleen and other affected organs were collected from the autopsied goats and brought to the laboratory on ice. The collected pathological samples were subjected for haematological and bacteriological examinations at Animal Health Research Division, Tripureswor and for serological laboratory investigation at Central Veterinary Laboratory, Tripureswor, Kathmandu. For further laboratory confirmation the pathological samples were sent to the IAH, Pirbright, UK.

RESULTS

Epidemiological findings

The outbreak occurred among goats maintained at upper farm of ARS, Bandipur. The population at risk was 219. The goat population at risk consisted of Khari, crossbreds of Kari with Jamnapari (Khapari) and Sinhal breeds of goats. The number of affected goats were 101 which included 67 adults and 44 kids (below 3 months of age). The total goats dead were 72, of which 42 were kids and 30 were adults. The overall morbidity and mortality rates were therefore 46% and 33%, respectively.

Clinical findings

The affected goats had pyrexia (104° - 106° F), though a few severely affected had subnormal temperature (98° - 100° F). Most of the affected goats had dyspnoea, increased heart rate (120-148) and respiration (30-40/minute) rate. The major clinical signs were mucopurulent oculonasal discharge with crust formation, conjunctivitis and some females had erosions around vulva. The affected goats had pneumonic sound on auscultation.

General debility, depression, anaemia, diarrhoea with soiled back, dehydration, recumbency for 3-4 days followed by death were observed. Most of the sick goats had crusty lesions in the mouth cavity and some goats had gingivitis. A few goats also had erosions on soft palate.

Post-mortem findings

Erosions and stomatitis were observed in mouth cavity. White cheesy material deposits were found on the surface and base of the tongue. Abomasum had inflammation together with congestion and haemorrhages along the mucosal folds. General congestion and hyperemia of small intestine was observed. Mesenteric blood vessels were congested. Small intestine did not reveal any parasites macroscopically, however 3 dead goats revealed tapeworm parasitic cysts. All dead goats revealed almost normal liver with distended gall bladder.

All cases had pneumonia and accumulation of frothy fluid in trachea. In severe cases, lungs were highly congested. Cut surface of the lungs revealed frothy fluid. Bronchial lymph nodes were swollen. In one dead goat there was serous fluid in the thoracic cavity. Pneumonic lesions were seen both in anterior and posterior lobes in kids. In adult goats, pneumonic lesions were seen in anterior lobe and at the brim of the posterior lobe with emphysema on

dorsal part. Some of the lungs were fully consolidated with dark red colour and others were partially consolidated with mottled haemorrhage. No any significant changes were observed in heart, kidney and joints.

Laboratory findings

Bacteriological examination

The lungs and nasal swabs were subjected for bacterial isolation on the media such as blood agar, Mac-Conkey agar, heart infusion agar and DHL agar. The lungs and tracheal fluid samples collected in mycoplasma broth were further subcultured and subjected to laboratory investigation for *Mycoplasma*, *Pasteurella spp.*, *Staphylococcus spp.* and *Mycoplasma spp.* were the major isolates identified by bacteriological examination of the culture.

Faecal Examination

The results of this examination revealed that 78% of 14 faecal samples were positive for *Strongyle spp.*, 28% positive for *Trichuris spp.*, and 21% were positive for *Eimeria spp.* of parasites.

Serological examination

Twenty one serum samples collected from the affected goats were subjected to competitive ELISA for PPR and rinderpest antibody detection. Of them, 18 were positive for PPR antibodies. Of these 21 goats, 15 had been vaccinated with tissue culture rinderpest vaccine in the past. Only 11 of 15 vaccinated goats had antibody against rinderpest.

Haematological examination

A total of 12 blood samples and blood smears collected from the affected goats were subjected for haematological studies. In general, sick goats had leucopenia, neutrophilia, lymphopenia, anaemia and decreased PCV.

DISCUSSION

The epidemiological pattern of the disease, clinical signs and symptoms, post-mortem lesions and laboratory investigation results revealed that the primary cause of the disease outbreak was due to PPR infection. The haematological pictures were indicative of viral infection. Similarly, the presence of antibody to PPR in the sick goats sera revealed that there was exposure of goats to the PPR virus. Although the majority of the goats was already vaccinated against rinderpest and rinderpest antibodies were expected in these animals, only 11 of 15 vaccinated goats had antibodies against rinderpest. The analysis of optical density obtained from the competitive ELISA tests indicated that these animals had low antibody titre against rinderpest but a high antibody titre against PPR. Serum samples from non vaccinated goats had no detectable antibodies against rinderpest but 5 of 6 sera had high antibody titre against PPR. Serum from four rinderpest vaccinated animals were negative for rinderpest antibodies but positive for PPR antibodies. Why all vaccinated animals did not

seroconvert is not known. Poor efficacy of the vaccine or mistakes in the vaccination techniques might have contributed for this apparent failure of vaccination. Though antibodies against rinderpest is generally considered to protect animals against PPR infection (Anderson, personal communication), goats with moderate levels of rinderpest antibodies were not fully protected against PPR infection. The laboratory results obtained from IAH, UK revealed that sick goats were PPR sero-positive and harboured PPR viruses as suggested by positive PCR test specific for PPR virus. These findings further supported the disease to be PPR. The bacterial examination results indicated that there was also involvement of bacteria of *Pasturella* and *Mycoplasma* species. These pathogens might be secondary because they play a great role to aggravate the condition in a disease like PPR.

After the confirmation of the outbreak, sick animals were treated with antibiotics, vitamins and electrolytes. Unexposed healthy animals of the farm were ring vaccinated with specific cell culture PPR vaccine. By these measures, the outbreak was controlled.

CONCLUSION AND RECOMMENDATION FOR CONTROL

The disease outbreak in goats at ARS, Bandipur was due to PPR. The following recommendations were made on the basis of experience gained through this investigation

- Supportive therapy such as antibiotics, vitamins, electrolytes etc. should be given to all the sick goats immediately. This will certainly help in reduction of the mortality.
- All the non-vaccinated goats should be vaccinated preferably with PPR vaccine two weeks before introducing them in the new farm.
- Strict quarantine measures like isolation of sick goats, proper disposal of fomites, training the technical staff for disease prevention and disinfection of the premises should be adopted.

ACKNOWLEDGMENT

We are grateful to Dr. J. Anderson, Chief of the World Reference Laboratory for Rinderpest IAH, Pirbright, UK for his help in laboratory diagnosis. Our sincere thanks to Dr. S.C. Ghimire, Laboratory Supervisor, SVSLDC Project, Nepal for his help in conducting ELISA for rinderpest and PPR. Thanks are also due to Mr. Chet Raj Upreti, Chief of ARS, Bandipur for his help during our visit to ARS, Bandipur.

REFERENCE

- Jha, V.C.; Bohara, K.B.; Singh, U.M.; Singh, S.K. and Thakur, R. (1995). Peste des petits ruminants of goats in Nepal. *Veterinary Review, Nepal*, vol. 10(2): .

HEIFER PROJECT INTERNATIONAL - IN ASIA AND AROUND THE WORLD

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ABSTRACT

Heifer Project International, Inc. (HPI) is a 55-year-old voluntary, non-governmental organisation (NGO). It was established as a way for common citizens to provide food-producing animals to those suffering in different parts of the world as a result of wars, natural disasters, or ever-present poverty. It has grown from an organisation that simply shipped live animals and frozen semen to dozens of countries, including Nepal, to one that is involved today in holistic development in over 40 countries. HPI has eight country offices in Asia - Nepal, China, Cambodia, India, Indonesia, Vietnam, Thailand, and the Philippines. Worldwide, there are over 25 country offices/programs. Around the globe, HPI uses animals as diverse as alpacas, guinea pigs, rabbits, water buffalo and camels. In Nepal, HPI is currently running projects with water buffalo, goats, pigs and rabbits. But our focus is not on the animals; our focus is on critical issues such as helping to build local capacity to carry out income and food-generating programs that continue after HPI funding has ceased. This paper will provide a brief overview of HPI around the world, and experience within the kingdom of Nepal.

BACKGROUND

Heifer Project International, Inc. (HPI) was formed in 1944 when a farmer and minister from the Midwestern United States returned from serving in Spain during their civil war. His name was Dan West, and he worked serving food and milk to children and adults who lost their homes during the unrest. He eventually thought that just handing out milk was not enough to help these people get on their feet again. He felt they needed a hand up, not a handout. So, after returning to Indiana in the USA, he convinced some of his friends and neighbours to donate dairy heifers, so the people could not only have milk, but have live animals from which to get more milk, offspring, income and manure. The idea seemed to explode like it was shot from a cannon! Soon, HPI was shipping cattle, pigs, goats, sheep, honeybees and the like to countries in Asia, such as China in 1947, South Korea in the 1950s, and Nepal in the 1950s and 1960s. Shipments were made to post-war Holland, Greece, France, Italy, and even Russia. Later, programs were started in Africa, and Central and South American countries as well. Finally, at present, we are working in over 40 countries, but have provided some type of assistance to over 115 countries since 1944.

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The size of the staff and operations grew along with the number of shipments and financial support to projects around the world. Our international headquarters is located in Little Rock, Arkansas, and houses over 70 full time staff. We rely on thousands of volunteers who generously give their time and money to support our programs for those living in poverty around the world. Most of our volunteers live in the US, but we have many in China, the Philippines and are even getting started here in Nepal.

Although when reading our name, or thinking of our organisation, the word "heifer" comes directly to mind, we are truly, first and foremost, about people. We are about people having a chance to use their potential. About people getting the dignity they deserve, the food they should have a right to. We are about compassion for an ever-changing world that needs to remember those who cannot help themselves, or who have not yet been shown the way to improve their lot in life.

HPI STRATEGY

Heifer Project International never works alone. In each and every country, with Nepal being an excellent example of what can work and why, we have strong partnerships with governmental and private institutions and agencies. We work hard on building these relationships - building bridges - so that we can learn from each other, build on each others' contributions, and focus on getting the job done. By working with others, we can extend our scope to include education, skills training, banking, safe drinking water, irrigation, roads, bridges, and so much more. In Nepal, we are just beginning to form partnerships with organizations both large and small, local and international, generous with time, energy, funds, or other resources.

RATIONALE

One can sit alone in a room and ponder all the troubles of our ailing world. Just counting the wars that are currently being fought around the world today is enough reason to get discouraged. Not to mention disease, poverty, hunger, illiteracy, discrimination, and the like. Many of these problems go unmentioned, their stories not making the headlines of the papers or the top of the TV news. It takes special people to look at such a large challenge as hunger and poverty. It takes people who have "service" in their mind, and in their actions. People with real vision for a better planet, real leaders.

We believe that the veterinary profession is providing and will continue to provide leadership in creating ideas for sustainable development. Nowhere else are there professionals who have the joint training in nutrition, metabolic systems, and comparative anatomy and physiology. Veterinarians are uniquely trained to think in "systems"; uniquely trained to look at the epidemiology of zoonotic diseases.

Heifer Project International has long relied on qualified veterinarians, and so-called "paravets" in some places where graduate veterinarians don't exist, to use their knowledge and skills in improving the quality of life for people around the world. Not just animals, but people too.

Currently, Heifer Project International has projects in over 40 countries throughout the world. In Asia, HPI is working in China, Vietnam, Philippines, Thailand, Indonesia, Cambodia, Laos, India, Sri Lanka, Bangladesh, Afghanistan and Nepal.

HEIFER PROJECT INTERNATIONAL IN NEPAL

Heifer Project International began its assistance program in Nepal during 1950s. It supplied Jersey and Brown Swiss cattle, Hampshire pigs, and Dorset Horn, Romney and Rambouillet and Corriedale sheep in 1957-58 to the Government Livestock Farm at Singh Durbar. Likewise, it supplied Hariyana cattle to Tarahara Farm in 1966-67. There was no continuity of these HPI assistance until 1990s. At present, HPI has a country office in Nepal located at the following address:

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PASSING ON THE GIFT

Giving life to another, giving hope for a better future. The Heifer is a symbol of life. A young female cow, a giver of life, a giver of milk, a symbol of motherhood, care, and love. In the same way, Heifer Project hopes to bring people to a new life. But HPI, as stated earlier, is not about animals. It is not about heifers, water buffaloes, goats, or rabbits. HPI is about helping those in need to learn to help themselves. It is about realising our own physical and spiritual wealth, and sharing it with those less fortunate. It is about helping people uplift themselves from poverty, by providing education and encouragement, because each man, woman, or child is the responsibility of his brother, and we are all brothers.

Each time we help our fellow man, we perform a selfless act. We help him regardless of race, regardless of religion, regardless of caste. A person who has food must share with one who is starving. A person who has knowledge must teach those who do not. This is what our higher power asks of us – to pass on the gift – to acknowledge and accept the gifts given to us and share them with others.

HPI encourages learning. Through HPI projects, people learn about themselves and their abilities. They learn to develop skills and share them with others. When people join together to work as a group, they accomplish so much more than individually. From a small family to a large community, in giving, each person gains, and the group thrives on the power of its own energy. They become unified and strong, an unstoppable force, filled with the courage to change and improve their lives.

HPI/NEPAL EXPERIENCE

In Gitanagar, Chitwan, a women's federation has been formed by over three hundred women who want to improve their communities. It began six years ago, when a small group of

women joined to find a community project that would help them earn money, gain respect from the men, and occupy their time by learning a useful skill. Their initial vision was to buy goats, then earn income by selling its offspring at the local market in Narayangarh. They approached a local bank for loan based on a group guarantee, but the bank denied them. Luckily, HPI gave the group 25 goats, each with twin kids in 1993. When the goats gave birth, they passed on the first offspring to other needy women, including the skills learned. The women began to gain confidence. They pooled their savings into a fund and began to finance small micro-credit projects. Women who never believed that they could have power over their lives now earn income, improve their homes, and rally against drinking and gambling in their communities. Their example is an inspiration and encouragement to other women, showing that with hard work and dedication, each person can make a difference.

In Okhaldhunga, a scattering of small women's groups has unified to form GMJS (Gramin Mahila Jagaran Samuha). This umbrella organization manages smaller groups, helping educate women in basic literacy, income earning, record keeping, and savings. These women run a goat and pig project with assistance from HPI.

The women of Talku Dude Chaur VDC of Kathmandu became aware of HPI and its mission to assist poor and needy people by providing animals that could bring income. Since two community members were successfully raising Angora rabbits in local climate, the group decided to expand rabbit farming and sell its wool. They requested HPI for more rabbits and vegetable seeds to grow vegetable for rabbit feeding. Families were trained to build a suitable pen, grow feed, and care for their rabbits. In July 1998, 40 families received a pair of rabbits each. Twice already by January 1999, the community has sold Angora wool, earning nearly NR 25,000 for the group. Using rabbit manure, crop production has improved, and they are also earning income from sales of surplus vegetables. At this time, the group savings fund loans money to members. In the future, they hope to build a local school, and a Village Animal Health Care center, staffed by a trained member of their group.

Heifer Project Intl. Nepal program has similar projects in Nuwakot, Palpa, and Mahottari districts also. Several other proposals are in various stages of approval process.

ANALYSIS OF THE PRESENT DAIRY FARMING SYSTEM IN CHITWAN AND KAVRE

D.D. Joshi¹ and M. Sharma¹

ABSTRACT

Chitwan and Kavre are among districts of Nepal where commercialisation of milk production has gained momentum. In this study, 57 dairy farmers, 40 from Chitwan and 17 from Kavre, supplying milk to milk collection centres of Dairy Development Corporation through milk producers' cooperative societies were interviewed on various aspects of dairy farming. The information collected included breeding, feeding and management practices, marketing aspects of live animals and their products, the major diseases perceived by the farmers. These results are analysed and described.

INTRODUCTION

Nepal is primarily an agricultural country. The share of livestock contribution in GDP is 18% and livestock and agriculture production is 29%. There are 6.3 million cattle and 3.1 million buffalo in the country and producing 871000 MT milk annually. There is a high livestock population density in the country. There have been efforts in the past to increase the milk production through cross breeding programme. Several districts have now come up with the semi-commercial dairy production system. This study was therefore undertaken to study the dairy farming system in semi-commercial dairy pockets of Nepal. Chitwan and Kavre districts of the Central Development Region where semi-commercial dairy farming is flourishing well were selected for this study.

METHODOLOGY

A team from National Zoonoses and Food Hygiene Research Centre (NZFHRC) visited milk collection centres of Dairy Development Corporation (DDC) in Chitwan and Kavre districts. A total of 107 dairy farmers supplying milk through Milk producer's cooperative societies were identified. From them, 57 farmers were selected randomly for detailed interview. Of them 40 families were from Chitwan and 17 were from Kavre. A questionnaire was used for farmers interview which included details on livestock breeds and breeding practices, feeds and feeding practices, disease pattern its treatment, prevention and control, and mass awareness programme.

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RESULTS AND DISCUSSION

Farm family population structure

The literacy rate among the interviewed farmers was 90% and 70% in Chitwan and Kavre, respectively. It has been observed that majority of the farmers have crossbred milking animals along with their traditional local, indigenous breeds of animals. On an average 7 were kept per farming household in Chitwan and Kavre (CBS, 1997). The total livestock population in Chitwan and Kavre is presented in Table 1.

Table 1. Livestock population and herd composition in Chitwan and Kavre

Species	Chitwan		Kavre	
	Total population	No. per farming family	Total population	No. per farming family
Cattle	105742	1.98	101965	1.89
Buffalo	83724	1.57	66700	1.24
Sheep	4100	0.08	4200	0.08
Goat	80860	1.51	189980	3.52
Pig	6630	0.12	8930	0.17
Poultry	915928	17.14	363400	6.74
Duck	21200	0.4	3950	0.07

Source: CBS (1997)

BREEDING PRACTICES

Information on the present animal breeding practices were collected (Table 2). It has observed that in Chitwan 75% farmers breed their buffaloes by improved bull whereas in Kavre 88% of the farmers breed their buffaloes by local bull. In case of cattle, 80% in Chitwan and 73% in Kavre farmers breed with local hilly zebu bull. None of the farmers interviewed used artificial insemination for breeding.

Table 2. Animal breeding practices in 57 families interviewed in Chitwan and Kavre

Breeding practice	Chitwan (n=40)				Kavre (n=17)			
	Cattle	%	Buffalo	%	Cattle	%	Buffalo	%
Natural with local bull	32	80	10	25	8	73	15	88
Natural with improve bull	8	20	30	75	3	27	2	12
Artificial insemination	0	0	0	0	0	0	0	0
Total	40	100	40	100	11	100	17	100

Most of the farmers buy / purchase their improved /crossbred/ improved milking animals cattle and buffalo from the adjoining animal hatbazars of Indian border.

FEEDING PRACTICES

About 95% of the interviewed farmers in Chitwan and 100% in Kavre keep their animals in stall feeding. In Chitwan 100% farmers feed their milking animals every day a commercially prepared animal feed whereas in Kavre only 76% feed commercial feed. However 100% farmers in both the districts feed their milking animals locally prepared concentrated feed called "Kundo or khole" and cut green grasses (collected from the agricultural field, forest and cultivated at farm). Almost 90% farmers in Chitwan and only 53% farmers in Kavre feed paddy straw to their animals without any pre-treatment. Though most of the farmers have heard of and seen the molasses urea block, only a few of them use them for supplementary feeding. Most of the farmers used crop residues and grains grown on their farm for feeding livestock (Table 3).

Table 3. Feeding Practices in 57 families interviewed in Chitwan and Kavre

Feeding practice	Chitwan (40)				Kavre (17)			
	Yes	%	No	%	Yes	%	No	%
Grazing	6	15	34	85	0	0	0	0
Stall feeding	38	95	2	5	17	100	0	0
Mixed (stall feeding and grazing)	31	78	9	22	17	100	0	0
Concentrate feed	40	100	0	0	13	76	4	24
Molasses urea block	2	5	38	95	0	0	17	100
Paddy straw	36	90	4	10	9	53	8	47
Urea treated paddy straw	3	8	37	92	6	35	11	65
Chopped and soaked straw	30	75	10	25	12	71	5	29
Green grass	40	100	0	0	17	100	17	100
Kundo or khole	12	30	28	70	17	100	17	100

COST OF FEED

Cost of feed is tabulated in Table 4. The cost of per Kg concentrate feed in Chitwan is one rupee higher than in Kavre. However, paddy straw is more expensive in Kavre than in Chitwan.

Table 4. Cost of livestock feed in Chitwan and Kavre

Feed Types	Chitwan (n=40)	Kavre (n=17)
Concentrate feed	Rs. 10 per kg.	Rs. 9 per kg.
Paddy straw (one load) 30 kg.	Rs. 100 per load	Rs. 126 per load
Other crop residue (one load) 30 kg.	Rs. 100 per load	Rs. 93 per load
Green Grass (one load) 40 kg.	Rs. 150 per load	NA

Cost of different categories of live animals is presented in Table 5. The milking cow and buffalo on an average cost Rs. 25,000 and 30,000 in Chitwan and Rs. 15,000 and 25,000 in Kavre, respectively.

Table 5. Cost of animals in Chitwan and Kavre Districts

Type of Animal	Chitwan (n=40)		Kavre (n=17)	
	Cattle	Buffalo	Cattle	Buffalo
Milking	22,750	20,800	12,300	22,125
Pregnant	17,600	17,000	4,000	17,900
Dry	7,000	8,000	1,025	8,800
Heifer	5000	5,400	775	5,600
Calves	2,700	2,400	775	2,700

ANIMAL DISEASES

It was reported by the farmers interviewed that the most prevalent animal diseases are foot and mouth disease (FMD), haemorrhagic septicemia (HS), mastitis, rabies, liver fluke, tape worm, round worm, mange, ring worm, milk fever and black quarter (BQ). The existing district livestock services offices, livestock centres and sub-centres are responsible to control the outbreak of the diseases and make prevention and control programme by regular vaccination and drenching. In some milk producer's cooperative societies, there is a provision of first aid treatment of the animals.

MILK PRODUCTION AND MILK MARKETING

Milk production by local, crossbred and improved animals in the farms interviewed were collected (Table 6). There is a difference in production of milk from local, crossbred and improved animal both in Chitwan and Kavre.

Table 6. Daily milk production (l) by different categories of animals

Type of Animals	Chitwan (40)	Kavre (17)
Local Cow	3	2
Crossbred cow (with Jersey)	8	5
Improved cow (Jersey)	15	0
Local buffalo	4	5
Crossbred buffalo (with Murrah)	10	8
Improved buffalo (Murrah)	12	15

Almost 100% farmers of the interviewed farmers sold their milk to the milk collection centres of DDC through milk producer's cooperative societies in both Chitwan and Kavre districts. Some farmers those who are not member of milk producer's cooperative societies also sell milk to private dairies and local markets at a slightly higher price. In Chitwan farmers sell their milk to DDC both in the morning and evening, whereas in Kavre only

morning milk is sold to the dairy and evening milk is consumed at home as the DDC does not collect the evening milk (DDC, 1998).

ACKNOWLEDGMENT

We are most grateful to US/Epa Washington DC USA particularly Mr. Mark Orlic and Mr. Tom With for their Kind continuous support and Ms. Kelly Cripe, ATI USA for her technical as well as financial support. We would like to thank all of the team members particularly Mr. Narayan Karki from National Zoonoses & Food Hygiene Research Centre (NZFHRC), Tahachal, Kathmandu, Nepal for their hard work to complete this study and prepare report. We are also thankful to Dr. Dev. Raj Adhikari, Chief, District Livestock Services Office, Bharatpur, Chitwan, Dr. B.N. Chaulagai, Chief, District Livestock Services Office, Dhulikhel, Kavre and Mr. Narayan B. Shrestha, Chief, Financial Division, Dairy Development Corporation for their technical support. We must thank to all the chairmen, secretaries, milk producing farmers of the different MPCS and MCC of the project area for their help.

REFERENCE

- DDC (1998). Annual Progress Report of Dairy Development Corporation (DDC) for the FY. 1997/98 Kathmandu Nepal.
- CBS (1997). Agricultural Statistics of Nepal. Centre Bureau of Statistics, Kathmandu

NON SURGICAL MANAGEMENT OF UROLITHIASIS IN MALE BUFFALO CALVES – CASE REPORTS

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ABSTRACT

Ten male buffalo calves aged 3-6 months raised by various farmers in Chitwan district, and two male calves aged 3-4 months raised on station at Lumle Agricultural Research Centre, Pokhara, had amuria, anorexia, straining and in majority of cases distension of lower abdomen. Clinical symptoms were suggestive of urethral obstruction, probably complicated with the rupture of urinary bladder. Urolithiasis, with whole of penile urethra being packed with amorphous white powder, was confirmed in four cases, in which urethrotomy was performed. Because of the difficulty in performing ischial urethrotomy in small calves in field conditions, and because of the poor success (50%) of surgical treatment, non-surgical treatment using herbal products was later adopted. Non surgical treatment was successful in all these cases. This paper describes the successful non-surgical treatment of urolithiasis in buffalo calves and management of a complicated urolithiasis with possible rupture of the bladder.

CASE HISTORY

Sick animals included for the study were ten male buffalo calves from Chitwan district during 1989-90, and two male buffalo calves from the Livestock Farm of Lumle Agricultural Research Centre (LARC), Pokhara during 1998. All the sick buffalo calves were of 3-6 months age group.

All calves had common symptoms of unthriftiness, restlessness, discomfort, straining and colic. All of them were also anuric. The duration of illness ranged from one to seven days. Treatment these calves received before presenting to us ranged from none to indigenous herbal preparation to parenteral antibiotic therapy with or without diuretics and analgesics. Calves with more than 4 days of illness had distended lower abdomen. Clinically, calves had either normal (presented within 3 days of illness) to slightly sub-normal (long-standing cases) temperature. Prepuccial orifice was dry in all calves and a distinct urethral pulse was visible just below the anus in some calves. Abdominal paracentesis revealed the presence of straw coloured fluid with ammoniac odour (probably urine). Some of the calves which were sick for longer duration, seemed to be active and recovered from sickness on the 4th or 5th day of illness, but became severely depressed thereafter.

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Owners of most of the calves were semi-commercial farmers and provided some home made concentrate (e.g. wheat bran with or without mustard cake) to the buffalo as well as their calves. Similarly, calves at LARC farm were also supplemented with commercial concentrate mixture.

CLINICAL DIAGNOSIS

Based on the history and clinical findings the cases were diagnosed as urethral obstruction, most likely due to urolithiasis in the penile urethra. Long-standing cases with distension of lower abdomen and abdominal cavity full of urine were probably due to rupture of the urinary bladder.

TREATMENT AND DISCUSSION

Surgical management

First two buffalo calves

First two cases were initially treated with intra-muscular injection of triflupromazine 20 mg I/M (Siquil®, Sarabhai chemicals) to rule out the probable cases of urinary obstruction due to urethral spasm. However, both calves did not urinate within one hour of treatment. It was therefore, decided to operate to locate and remove calculi by surgery. It was assumed that as in the adult cattle (bullocks), calculi in young buffalo calves would be located in the sigmoid flexure of the penis, and hence similar surgical approach was adopted.

While preparing the calves for surgery, urine accumulated in the peritoneal cavity was removed by abdominal paracentesis and calves received parenteral fluid therapy.

Calves were restrained in lateral recumbency. "Base of scrotum approach" was adopted for surgery (Lundvall, 1974). The site was prepared for aseptic surgery, and was infiltrated with local anaesthetic (Lignocaine) starting from the base of scrotum to the level of tuber ischii. The skin incision was made over the median raphe from just above the base of scrotum and extending upwards about three to four inches. Subcutaneous fascia was separated by blunt dissection and the penis was pulled out of the incision by straightening the sigmoid flexure. Part of the penis that was accessible was digitally palpated giving special attention to sigmoid flexure region to locate any abnormalities especially the hard calculi. Uniform hard mass all over the urethra was detected, but no distinct calculus could be detected.

When the urethra was slit open with the aim to pass a catheter to the urinary bladder, the whole of penile urethra was found to be packed up with white amorphous powder. It was therefore decided to open the urethra near the tuber ischii and pass a catheter to establish urine flow. "Ischial urethrotomy" was thus performed (Lundvall, 1974). A catheter (human paediatric) was then passed to the urinary bladder, but with a great difficulty.

These calves received routine post-operative care of antibiotics, analgesics and fluid therapy. However, of the two calves operated, one did not recover after treatment and died 12 hours after operation. The second calf recovered after operation with establishment of a

regular flow of urine through the catheter, which was fixed at the site of incision. Unfortunately, this calf died on day 19 post-operation, reportedly due to tympany. Nothing is known whether the death was related to post-operative infections / complications or was totally independent.

Second two buffalo calves

Based on the experience with the first two calves, ischial urethrotomy was performed in other two calves as described above. Urine flow was established by passing a catheter to the bladder which was fixed at the site of incision. These calves received normal post-operative care. Again one calf died 24 hour post operation. The second calf completely recovered and was active until two months after surgery, after which it was not followed.

The success rate of surgery was therefore only 50%, but could be 25% if the death in the second calf was also related to post operative complications.

Non-surgical management of urolithiasis

Because of the difficulty associated with the surgery especially in catheterising young animals and a poor success rate, it was decided to adopt non-surgical management approach. A total of eight calves were treated as outlined below.

Calves were treated with a herbal (Ayurvedic) product marketed for human use and claimed to be urolithiolytic and diuretic (Tab. Cystone, Himalaya Drug Company, India). The dose rate administered was five tablets three times a day for 5 days followed by 5 tablets two times a day for 5 days and then 5 tablets daily for 15 days orally. Depending on the severity of the condition, abdominal paracentesis was performed (except two calves) and calves received intravenous 5% Dextrose saline, 500 – 1000 ml. Paracentesis was done using three 16G needles, and all fluid that could be drained out from the peritoneal cavity was removed. Fluid therapy and paracentesis was repeated on the second day. However, two calves presented within 24 hrs of illness did not receive fluid therapy, and abdominal paracentesis was not performed in them.

All calves started urination 18-36 hours post treatment. Normal flow of urine, however, was established only after 2-3 days. All eight calves treated recovered completely. No follow up was done in these calves after the treatment period to investigate whether the disease recurred later on. However, recurrence of the disease is considered unlikely as two calves of LARC farm remained disease free for six months after the treatment.

Long standing cases were likely to have rupture of bladder. However, no attempt was made to repair it through surgery. How was the ruptured bladder repaired is unknown. Fibrosis of the bladder wall probably helped to self repair the bladder. It is said that, in bullocks at least, rupture of the bladder normally occurs on the dorsal wall near the urethral opening and majority of them recover naturally through fibrosis which occurs quickly in ruminants (Prof. D.M. Tadkod, personal communication). It is possible that ruptured bladder in buffaloes also heal similarly.

Urolithiasis obstructing penile urethra is commonly observed in castrated male ruminants. In bullocks, calculi are normally lodged in sigmoid flexor of the penis (Lundvall, 1974). However, there is scant literature on its occurrence in young animals, particularly in buffalo calves. Heavy concentrate ration is known to be the most common predisposing factor for urolithiasis in ruminants (Blood and Radostits, 1989), a common management practice observed in all calves of this study. Diet management would probably be an important aspect for prevention of obstructive urolithiasis in buffalo calves.

There are certain limitations of the study presented in this communication. Important ones to mention are lack of confirmatory diagnosis of cases treated non-surgically other than from clinical examination and experience, lack of knowledge on the composition of the calculi, lack of proper experimental design to prove the claim and lack of knowledge on the action of the drug to breakdown the urethral calculi. The findings presented here are therefore, meant only to share our experience with professional colleagues so that the field veterinarians are aware of the disease situation and will be encouraged to investigate similar cases in more detail. Furthermore, if considered important, the disease could be investigated systematically in future.

REFERENCES

- Blood, D.C. and Radostits, O.M. (1989). *Veterinary Medicine*, 7th Edition, Bailliere Tindal Ltd, London, pp. 402-409.
- Lundvall, R.L. (1974). The urinary system. In: Oehme, F.W. and Prier, J.E. (Eds.), *Textbook of large animal surgery*, Williams and Wilkins, Baltimore, pp 450-460.

A COMPARATIVE STUDY OF DIFFERENT TEST METHODS FOR CONFIRMATORY DIAGNOSIS OF RABIES

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ABSTRACT

Biological and histo-pathological tests used for confirmatory diagnosis of rabies in the State Medical, Food and Veterinary Laboratory of Kassel were compared with fluorescent antibody test on the basis of existing laboratory records. The test results were analysed to define sensitivity, specificity and other characteristics of the diagnostic tests. Fluorescent antibody test appeared to be the most reliable diagnostic method together with biological test (Virus isolation in cell culture) for confirmatory diagnosis of rabies.

Introduction

Majority of rabies cases in animals and humans are still diagnosed on the basis of clinical signs and symptoms in many areas of the world. However, it is difficult to make any conclusion when the suspected animal has 'dumb' form of rabies or the history of dog bite is unknown. The situation is complicated when a rabid or suspected dog seen in the urban or rural areas is killed by community members. Therefore, it is necessary to confirm rabies cases using various laboratory diagnostic methods.

The hippocampus is commonly used for the diagnosis of rabies. Brain specimens to be submitted for laboratory examination include cerebrum, cerebellum, hippocampus, medulla and thalamus preserved in 50 per cent glycerol-saline solution for virological examination and in 10 per cent buffered formalin for histo-pathological examination. Occasionally, virus antigen or infectivity may be demonstrated in salivary glands and not brain, and it is recommended that samples of sub-maxillary salivary gland should also be submitted in glycerol-saline and formalin preservatives. Histo-pathological examination of specimens has the merit that it sometimes allows an alternative diagnosis to be established in cases of nervous disease which are not due to rabies.

Negri bodies can be demonstrated in impression smears prepared from fresh brain tissue and stained by the method of Sellers. Although it is a less sensitive method, experienced personnel can obtain a correct diagnosis in 80 to 90 percent of the cases, especially in dogs dead of furious rabies (Acha and Szyfres, 1987).

The standard method of making a diagnosis is to demonstrate rabies virus antigen in impression smears of fresh brain by immunofluorescence. Fluorescent antibody test (FA

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test) may demonstrate antigen in instances in which specimens are too decomposed to yield virus isolates. The FA test for detection of rabies antigen has been perfected to such an extent that it is the quickest, most reliable method available, both for diagnostic and research purposes (Dean and Abelse, 1973). The effectiveness of the test depends on the competence of the technicians and the quality of the reagents, especially the conjugate. Most laboratory use immunofluorescence as the sole diagnostic test and resort to mouse inoculation a confirmatory test only in instances where a negative result is recorded in an animal which has potentially been involved in the exposure of a man to infection.

The demonstration of virus antigen by immunofluorescence in corneal impression smears, or in nerve fibres surrounding hair follicles in frozen sections of nuchal skin biopsies has been applied to *intra vitam* diagnosis of the disease in human patients, but the results obtained has been inconsistent (WHO, 1992). Examination of skin biopsies and corneal smears may also be used for post-mortem diagnosis in countries where opening of the skull of the dead person is not accepted by relatives on cultural or religious grounds.

Virus antigen may be demonstrated by enzyme-linked immunoassay, and kits for performing the test in microplates are available commercially (Perrin and Sureau, 1987). The test can be performed in the course of a working day even under field conditions.

The isolation of virus by intracerebral inoculation of brain suspension into weaned mice was the standard diagnostic method prior to the widespread adoption of the immunofluorescence test. The method is sensitive, yielding isolates from virtually all infected specimens received at the laboratory in a well-preserved state, but it may take up to 2 weeks or longer to obtain a result. The mouse inoculation test (MI test), in spite of its simplicity, depends greatly on the accuracy of its performance for dependable results (Koprowski, 1996). Virus may also be isolated in a variety of line cell culture in place of mice. Since rabies virus is poorly cytopathic, its isolation has to be demonstrated by performing immunofluorescence tests on the cultures. Virus isolation in cell culture has been shown to be as efficient as the FA test and the MI test for demonstrating small amounts of rabies virus (Rudd and Trimarchi, 1987).

Antibody to rabies virus may be demonstrated by a variety of methods including complement fixation, indirect immunofluorescence and neutralisation tests. The tests can be used to establish a diagnosis of rabies in live human patients, who develop an antibody response five days or more after the onset of the illness.

MATERIAL AND METHODS

Examination of impressions or smears of tissue samples from Ammon's horn and brain stem of suspected domestic and wild animals were carried out for routine rabies diagnosis at the State Medical, Food and Veterinary Laboratory of Kassel. Positive specimens were detected by the FA test, the MI test or virus isolation in neuroblastoma cells.

An investigation of laboratory records of rabies diagnosis of the State Medical, Food and Veterinary Laboratory of Kassel from 1981 to 1995 was carried out. Biological and histopathological tests used for confirmatory diagnosis of rabies were compared with FA test on

the basis of existing laboratory records and the results were analysed to define sensitivity, specificity and other characteristics of the diagnostic tests.

RESULTS

The comparison of biological and histo-pathological test methods with FA test was done on the basis of laboratory record which is presented in Table 1.

Table 1. Comparison of different diagnostic tests with FA test for the diagnosis of rabies

	FA test/MI test	FA test/VICC	FA test/HIST
Both positive +/+	4	78	275
Both negative -/-	3,923	2,088	1,056
Positive/Negative	2	4	23
Negative/Positive	14	12	0
Total samples	3,943	2,182	1,354

Note: FA test = Fluorescent Antibody Test,
VICC=Virus Isolation in Cell Culture

MI test=Mouse Inoculation Test
HIST=Histological examination

The analytical part of comparative study of different diagnostic tests with FAT for rabies diagnosis is presented in Table 2.

Table 2. Characteristics of different diagnostic tests used for rabies diagnosis when FA test is used as a gold standard

Indicators	MI test	VICC	HIST
Sensitivity of test	0.6667	0.9512	0.9228
Specificity of test	0.9964	0.9943	1.00
Predictive value of a +ve test	0.2222	0.8667	1.00
Predictive value of a -ve test	0.9995	0.9981	0.9787
Apparent Prevalence	0.0046	0.0412	0.2031
True Prevalence	0.0015	0.0376	0.2201
Kappa value	0.33	0.90	0.95

We found that the performance of biological and histo-pathological test methods was greatly varied when FA test was used as a gold standard. Under the given conditions, the sensitivity of MI test was very low but specificity of all diagnostic tests was almost similar. Biological test method gave a significant number of false positive cases whereas histo-pathological test gave a significant number of false negative cases. The Kappa value indicates that there is a high degree of agreement between FA test and VICC as well as HIST.

The characteristics of different diagnostic test methods used for confirmatory diagnosis of rabies in the laboratory is presented in Table 3.

Table 3. Characteristics of different diagnostic tests used for confirmatory diagnosis of rabies

Indicators	Results				
	Probability	FA test	VICC	MI test	HIST
Sensitivity of test	P(T+/D+)	0.9369	0.9574	0.90	0.9228
Specificity of test	P(T-/D-)	1.00	1.00	1.00	1.00
Predictive value of a +ve test (Diagnosability)	P(D+/T+)	1.00	1.00	1.00	1.00
Predictive value of a -ve test	P(D-/T-)	0.9963	0.9981	0.9995	0.9787
Probability of a false +ve result related to test	P(D-/T+)	0.00	0.00	0.00	0.00
Probability of a false -ve result related to test	P(D+/T-)	0.0037	0.0019	0.0005	0.0213
Probability of a false +ve result related to state of nature	P(T+/D-)	0.00	0.00	0.00	0.00
Probability of a false -ve result related to state of nature	P(T-/D+)	0.0631	0.0426	0.10	0.07718
Apparent Prevalence	P(T+)	0.0516	0.0412	0.0046	0.2031
True Prevalence	P(D+)	0.0551	0.0431	0.0051	0.2201

Note: Rabies was considered as a confirmed case when FA test was positive or any of the biological test was positive when it was carried out in FA test negative samples.

The result demonstrates that the specificity of the test was same in all diagnostic tests, however, the sensitivity of each test was different. The apparent prevalence rate of rabies in each test was always lower than the true prevalence rate.

DISCUSSION

The comparative study of different diagnostic test methods indicates that FA test alone can not detect all rabies cases. In a comparative study of 19,107 infected brains, positive diagnosis was possible in an average of at least 98.2 per cent by FA test and at least 98.4 per cent by MI test (Wachendörfer et al., 1985). Fluorescence-positive, biological test negative specimens can occasionally be expected, especially in decomposed material since FA test detects inactivated as well as live antigen (Wachendörfer, 1966). In this respect, FA test is more sensitive than biological tests.

Biological test positive, fluorescent negative results also occur, because early stages of infection can not be diagnosed by FA test. In a study carried out in the Laboratories for Veterinary Science of the New York State Department of Health, 802 of a total of 804 were found positive by the FA test. In only 2 cases was the mouse test positive and the FA test negative (Dean et al., 1996). Therefore, the confirmatory diagnosis of rabies should be carried out with any of the available biological diagnostic test methods when FA test negative result is doubtful. There is no need to carry out histo-pathological examination as an additional test for rabies diagnosis when FA test is negative. The lower sensitivity as well as Kappa value of MI test was observed when FA test was used as a gold standard. Most

probably, such a result may be associated with selective character of sampling. It is important to note that MI test was carried out in cases when FA test result was doubtful or negative. FA test appeared to be the most reliable diagnostic method together with VICC for confirmatory diagnosis of rabies.

CONCLUSION

Fluorescent antibody test appeared to be the most reliable diagnostic method together with biological test (especially virus isolation in cell culture) for confirmatory diagnosis of rabies. There is no need to carry out histo-pathological examination as an additional test for rabies diagnosis when FA test is negative.

ACKNOWLEDGMENT

This paper is based on a research work which was carried out for the partial fulfilment of the Master's degree at the Free University of Berlin in 1996.

REFERENCE

- Acha, P.N. and Szyfres, B. (1987). Rabies. In: *Zoonoses and Communicable Diseases common to Man and animals (Second Edition)*. Pan American Health Organisation, WHO. pp. 425-449.
- Dean, D.J. and Ableseth, M.K. (1973). The fluorescent antibody test. In: *Laboratory techniques in rabies, Third edition*, WHO monograph series, No.23.
- Dean, D.J.; Ableseth, M.K. and Atanasiu, P. (1996). The fluorescent antibody test. In: *Laboratory techniques in rabies. Edited by Meslin, F. X., Kaplan, M. M. and Koprowski, H., Fourth edition*, World Health Organisation. pp. 88-95.
- Koprowski, H. (1996). The mouse inoculation test. In: *Laboratory techniques in rabies. Edited by Meslin, F.X., Kaplan, M. M. and Koprowski, H., Fourth edition*, World Health Organization. pp.80-87.
- Perrin, P. and Sureau, P. (1987). A collaborative study of the experimental kit for rapid rabies enzyme immuno-diagnosis (RREID). *Bulletin of the World Health Organization*, 65: 489-493.
- Rudd, R.J. and Trimarchi, C.V. (1987). Comparison of sensitivity of BHK21 and murine neuroblastoma cells in the isolation of a street strain rabies virus. *Journal of Clinical Microbiology*, 25: 1456-1458.
- Wachendoerfer, G. (1966). *Dtsch. Tierärztl. Wschr.* pp.73. 446,485
- Wachendoerfer, G., Frost, J.W. and Fröhlich, T. (1985). Current Diagnostic Procedures of Rabies and Related Viruses. In: *Rabies in the Tropics. Editors: Kawert, C., Merieux, C., Koprowski, H. and Voegel, K.*, Springer Verlag, Berlin. pp.40-46.
- World Health Organisation (1992). Eighth Report of the WHO Expert Committee on Rabies. WHO Technical Report Series 824, Geneva.

APPETITE CYCLE AND DIGESTION IN DEER - A REVIEW

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ABSTRACT

Although deer are primarily grazing ruminants, and also have special preference to certain parts of plant material, they can select and survive from what is available in each habitat. In temperate regions of the world, there is distinct variation in feed availability, and the biology of deer developed and evolved accordingly. In spring time, for example, voluntary food intake (VFI) is found to increase upto 70% as compared to the winter season, which indicates that there are distinct cycles of appetite. These appetite cycles are influenced by photoperiod and ambient temperature. Also, various hormones such as melatonin, hypothalamic releasing factors, gonadotrophins and gonadal hormones, prolactin and thyroid hormones show respective cycles.

INTRODUCTION

Deer are primarily grazing ruminants and show a good grazing pattern. They are also good browsers. Among the pastoral livestock, deer are found to be the best in converting the feed to meat. But it is interesting to note that, although forage is the principal diet of deer, different deer species have special preference to some particular parts of the plants. They also have ability to select and survive from what is available in each habitat, especially in the wilderness. For example, in wood and shrubland, they browse on leaves, shoots and buds within reach, and on the ground they feed from low growing herbs and grasses known collectively as forbs (Chaplin, 1977). They also consume fungi and fruits in some season. If the trees and pastures are not abundant, they rely on a relatively greater proportion of ground plants.

Plant cycles in most part of the world are essentially seasonal remaining in dormant state during the winter. The flush of new plant growth in spring and summer provides plenty of highly digestible herbage, but with the onset of winter, plant growth ceases, and the deer must depend upon the scarce standing crop of low digestibility, mobilise their fat reserves until the spring (Barry *et al.*, 1991). Therefore, it is important and interesting to understand the capabilities of these animals in surviving in the abundance and dearth - their hidden physical and physiological fitness in coping with the challenges in the nature.

FEEDING BEHAVIOUR

Most of the wild deer like fallow deer, white-tailed deer etc. are crepuscular feeders, but if the disturbance in the daylight occurs, they might exhibit nocturnal patterns. At different

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intervals of the day, browsing and grazing activities can be seen in fallow deer. With the onset of winter, feeding rhythm in white-tailed deer follows a cyclic pattern with peak activity every 4 to 8 hours at sunrise, mid-day, sunset and twice at night (Denholm, 1984). However, as the winter progresses, all the grazing activities of the morning and night are ceased and concentrated during the warmest time of the day.

Odocoileus spp., rusa and fallow deer are found to exhibit the synchronisation of feeding, cudding and resting. Farmed red deer (*Cervus elaphus*), on the other hand, tend to graze like a herd of cattle, line abreast and separated by a few yards with the dominant hinds in front and the rest behind (Blaxter *et al.*, 1974).

ABILITIES OF DIGESTION

The extent of development of the four-chambered stomach - rumen, reticulum, omasum, abomasum - is not equal in all species of deer. The more evolutionary primitive subfamilies such as the musk deer, the water deer and muntjacs lack the full complexity of development achieved among the *Cervinae* and *Odocoileinae* (Putman, 1988). In the primitive varieties, the stomach is four as in more advanced species, but the third compartment, omasum, is extremely small. Besides, rumen pillars are poorly developed and so, fibrous food materials may be retained only for a brief period. This signifies that these more primitive deer are comparatively less efficient in digesting the structural carbohydrates from plant material because active digestion of cellulose occurs within the rumino-reticular complex. Therefore, these species must select foods low in fibre but high in soluble carbohydrates, proteins and fats that are more easily digestible. In contrast, the deer species that come under the family *Cervinae* such as red deer, sambar deer etc. are capable of digesting plant materials more efficiently.

SEASONAL CYCLES

The seasonal cycle in bodyweight and fatness seen in wild deer is not entirely due to alteration of abundance and scarcity of forage (Barry *et al.*, 1991). Many seasonal cycles are endogenous, determined by the nature of the animal (Kay, 1985). The appetite cycle of deer, however, coincides with the availability of forage; so the demands for major nutrients are significantly increased during summer when good quality forage are available in abundance. This means that there are pronounced seasonal changes in metabolic rates because these rates are associated with annual changes in appetite cycle or the voluntary food intake (VFI). In white-tailed deer, for example, the fasting metabolic rate, heart rate and activity are found to be the greatest in summer and the lowest in winter. Such changes can be seen even in those deer that are sheltered and fed to appetite. Not surprisingly, Adam (1991) observed that irrespective of food supply, red deer fawns fail to maintain good growth rates during mid-winter months. The phenomenon of seasonal cycles has been studied in other species of deer as well. Laudan *et al.* (1989), for instance, observed that the significant seasonal changes in the level of VFI in adult red and Pere David's deer confirm the preliminary study of two red deer hinds by Suttie *et al.*, (1984) indicating that seasonal rhythm in VFI in deer occur in both sexes. But all do not have similar appetite cycle. For example, in immature stag, castrate stag and non-pregnant hind, seasonal changes in VFI is less marked as compared to mature stags and hinds. Regarding the selection of diets, a remarkable difference can be

found between stags and hinds. To some extent, the difference in diet between the sexes is due to the fact that (in turn owing to a degree of sexual segregation) stags and hinds may occupy separate geographical ranges where availability of different foodstuffs may not be the same; but even in areas of range overlap, the difference in species composition and quality of diet were retained (Putman, 1988)

DEPRESSED APPETITE AND ACTIVITY IN WINTER

The VFI of adult red deer stags and Soay rams falls abruptly at the onset of the rut (Kay, 1985). Even in the absence of females, they may stop eating for days together. Appetite may then recover to some extent but remains low throughout winter. Suttie *et al.* (1984) on the other hand, observed that the seasonal rhythm of the females was of a lower amplitude and no identifiable drop in intake due to the rut was evident.

Although reduced forage availability during winter enforced the deer to limit intake in the wild and this phenomenon became permanently established, the appetite cycle is thought to be primarily an ambient temperature mediated behavioural response and in red deer it is associated with the photoperiod controlled cycle of sexual activity. Male red deer are capable of responding to light in a square wave pattern as they do to an annual cycle and the imposition of three such patterns in one calendar year elicited cycles of food intake, gonadal development and regression, and antler growth (Suttie *et al.*, 1984). However, seasonal changes in VFI was not as expected because this occurred towards the end of short day periods. In natural photoperiod, peak food intake would have been expected to occur during increasing days. Of the observations conducted to study the time to reach peak food intake and the time to reach minimum food intake, only the former was significantly different from the natural photoperiod. Suttie *et al.* (1984) therefore conclude 'possibly the observed annual pattern is a combination of two cycles, namely, one directly mediated by photoperiod and the other by sex hormones, especially testosterone'. The appetite cycle may be regarded as a response to the sum of the nutrient demands arising from the other seasonal cycles, rather than a primary response to changing daylength (Barry *et al.*, 1991).

Digestible energy intake in winter are reduced to 40 to 60% of the maximum spring intake (Denholm, 1984). In red deer Milne *et al.* (1978) observed an increased VFI by 65-70% in spring fed with *Festuca* species and heather diets. Limited number of studies suggest that in spite of constant food supply throughout the year, reduced winter appetite is observed in *Odocoileus* species, red deer and reindeer (*Rangifer tarandus*). Similar trend has been found in fallow deer, sika deer (*Cervus nippon*), Pere David's deer (*Elaphurus davidianus*) and Chinese water deer (*Hydropotes inermis*) (Chaplin, 1977). When the autumn and early winter is approached, white-tailed deer increase foraging activity and feed intake as an early response to decreasing temperature, but as the environmental temperature falls further, their appetite, activity and energy requirements are abruptly reduced and they enter a physiological adaptation with reduced energy intake.

ROLE OF HORMONES

Hormonal control of seasonal changes in VFI in deer is substantially unknown at present (Barry *et al.*, 1991). But it is now recognised that the pineal hormone, melatonin, which is

secreted during the hours of darkness, is the primary mediator of ambient photoperiodic information within the animal's body (Adam, 1991). Therefore, strategic administration of exogenous melatonin elicits the same physiological responses as an artificial reduction in daylength, namely reproductive activity and reduced VFI (Domingue *et al.*, 1991). During daytime, impulses from retina activate a neural centre in the hypothalamus, the suprachiasmatic nucleus, whose diurnal activity allows it to serve as an internal clock (Kay, 1985).

Formation of melatonin is suppressed during the daytime because this neural centre in turn projects by way of the cervical sympathetic nerve to the pineal gland. Whereas abrupt changes in photoperiod cause an immediate and appropriate change in the diurnal melatonin-secretion profile, there is a characteristic delay before the overt physiological changes develop (Adam, 1991). With the lengthening of nights, there is a change in the pattern of melatonin secretion that leads to the secretion of gonadotrophins from the pituitary gland and the production of gonadotrophin releasing hormone in the hypothalamus. These ultimately promote the growth and activity of the ovaries and testes. As melatonin exerts its effects in the reproductive system via changes in the secretion of these hormones, it is convenient to speculate that its effects on VFI may also be mediated by hypothalamic and pituitary hormones (Barry *et al.*, 1991). Thus melatonin does not have direct stimulatory or inhibitory effects on growth (and by inference VFI) and reproduction, but the entire rhythm is entrained by it.

Although Denholm (1984) suggests that apart from pineal gland, hypothalamus, pituitary and gonads, thymus and thyroid glands may also be responsible in mediating cyclical physiological activity, no convincing evidence is available so far. However, prolactin has been thought to be a mediator of associating with long daylength, high food intake and rapid weight gain, as entrained by melatonin. Curlew *et al.* (1988) observed that when red deer hinds are treated with bromocriptine, the springtime rise in both prolactin and appetite are significantly delayed; this suggests the probable association of prolactin and appetite. Also, there are seasonal changes in the plasma concentration of prolactin in deer. But increased VFI as an effect of prolactin injection has not been consistently observed. Barry *et al.* (1991) have reviewed that when animals are treated with melatonin, there is a decline in plasma concentration to baseline levels of 20 mg/ml within 30 days of treatment; ultimate effects on seasonal rhythm due to such associated effects are inconclusive. In an attempt to differentiate the effects of melatonin from those of prolactin, Milne *et al.* (1987) administered 20 mg bromocriptine/day orally to red deer hinds from July to October. At the same time, other groups were treated with melatonin. It was observed that although prolactin levels were low in both the treated groups, the ones treated with melatonin had an early decrease in VFI in the autumn, and the group treated with bromocriptine showed lower VFI than that of the control or melatonin-treated groups, but the decline in VFI was three weeks later than that of the control group. It is possible that bromocriptine administration induces a reduction in VFI independent of its effect upon circulating concentrations of prolactin (Barry *et al.*, 1991).

REFERENCES

- Adam, C.L. (1991). Nutrition and the implications of modifying the Seasonality of Red Deer in 'Recent Advances in Animal Nutrition'; Butterworths: 211-221.

Barry, T.N., Suttie, J.M., Milne, J.A., R.N.B., Kay (1991); Control of Food Intake in Domesticated Deer in 'Physiological Aspects of Digestion and Metabolism in Ruminants'; Proceedings of the Seventh International Symposium in Ruminant Physiology; 385-398.

Blaxter, K.L., Kay, R.N.B., Sharman, G.A.M., Cunningham, J.M.M., and Hamiltan, W.J. (1974); Farming the Red Deer; Dept of Agriculture and Fisheries for Scotland, H.M.S.O. Edinburgh.

Chaplin, R.E. (1977); Food and Feeding in Deer, Blandford Press Ltd. : 37

Curlewis, J.D., Loudon, A.S.I., Milne, J.A., and Mc Neilly, A.S. (1988); Effects of chronic long acting bromocriptine treatment on bodyweight, VFI, coat growth and breeding season in non-pregnant red deer hinds; Journal of Endocrinology 119 : 413-420.

Denholm, L.J. (1984); The Nutrition of Farmed Deer in 'Deer Refresher Course'; The University of Sydney : 662-700.

Domingue, B.M.R., Dellow, D.W., Wilson, P.R. and Barry, T.N. (1991); Nitrogen Metabolism, Rumen Fermentation, and Water Absorption in Red Deer, Goats and Sheep in 'New Zealand Journal of Agricultural Research'; 34; Crown Copyright : 391-400.

Kay, R.N.B. (1985); Seasonal Variation of Appetite in Ruminants in 'Recent Advances in Ruminant Nutrition'; Butterworths : 199-208.

Laudon, A.S., Milne, J.A., Curlewis, J.D., Mc Neilly, A.S. (1989); A comparison of the seasonal hormone changes and patterns of growth, voluntary food intake and reproduction in juvenile and adult red deer and Pere David's deer hinds in 'Journal of Endocrinology'; 122 : 733-745.

Milne, J.A., Macrae, J.C., Spence, A.M., and Wilson, S. (1978); A comparison of the voluntary intake and digestion of a range of forages at different times of the year by the sheep and the red deer in 'British Journal of Nutrition'; 40 : 347-356.

Milne, J.A., Sibbald, A.M., Mc Cormac, H.A., and Loudon, A.S.I. (1987); The influences of nutrition and management on the growth of red deer calves from weaning to 16 months of age in 'British Society of Animal Production'; 45 : 511-522.

Putman, R. (1988); Food and Feeding in 'The Natural History of Deer'; Christopher Helm : 42-56.

Suttie, J.M., Corson, I.D., and Fennessy, P.F. (1984); Voluntary intake, testis development and antler growth patterns of male red deer under a manipulated photoperiod in 'Proceedings of the New Zealand Society of Animal Production'; 44 : 170.

CEREBROSPINAL NEMATODIASIS IN GOATS IN WESTERN TERAI OF BANKE DISTRICT- A REVIEW

K. Karki and B.N. Adhikari

ABSTRACT

*Seasonal occurrence (mainly in October-November) of a disease syndrome locally called 'Kumri' meaning weakback was observed in goats in Banke and other districts of western Terai in the last few years. Traumatic injury to the lumbar region, nutritional deficiencies and parasitism in the spinal cord were the likely causes considered. Based on the epidemiological pattern (seasonal occurrence, clinical symptoms, no temperature and no systemic infection, and no response to supplementation of vitamins and minerals) of the disease, the disease was provisionally diagnosed as cerebrospinal nematodiasis. This has been further substantiated by confirmation on the occurrence of parasites of *Setaria* spp. in caitle in this region, and treatment response of affected goats to diethylcarbamazine. However, there is a further need to demonstrate microfilaria in the spinal cord of affected goats. The current knowledge on the disease is reviewed and it is suggested that until further information on the disease is available, diethylcarbamazine could be a drug of choice for treatment of affected goats.*

FIELD SITUATION

A peculiar syndrome in goats in Banke district was first observed during October/November 1987. Goats above six months of age were mostly affected. Typical clinical signs in affected goats were paralysis of one or both hind limbs, paralysis of lumbar region, incoordination and swayback gait (Karki, 1996). When hand fed, affected animals eat normally, and there was no systemic involvement (no temperature). Later, similar condition was also reported from different Terai or valley districts of mid and far western regions including Dang, Surkhet, Bardiya and Kalali. During the outbreak the morbidity and mortality rates in affected flocks ranged between 15-20% and 2-15%, respectively.

Based on the clinical findings of posterior paralysis, the possible causes suspected for further investigation included,

1. Traumatic injury to the lumbar region.
2. Nutritional deficiencies of calcium, phosphorus, vitamin A, D etc., and
3. Parasitism in the spinal cord.

PROVISIONAL DIAGNOSIS

Based upon the clinical manifestations, the disease was provisionally diagnosed as cerebrospinal nematodiasis. Though, the microfilaria have not been demonstrated in the

spinal cord of affected goats, adult parasites of *Setaria spp.* have been identified from the peritoneal cavity of cattle which was brought for post mortem examination at Veterinary Hospital, Banke in 1998 (B.S. 2055-04-19). Microfilaria of this parasite are known to migrate irrationally in non-natural hosts like goats. Confirmation on the occurrence of parasites of *Setaria spp.* further supported the hypothesis that the disease was due to cerebrospinal nematodiasis.

TREATMENT RESPONSE

Treatment of hundreds of goats suffering from posterior paralytic syndrome with diethylcarbamazine 10 mg/kg body weight has given very encouraging results. Animals treated within three days of showing clinical signs had complete recovery within few days of treatment. In long standing cases, the recovery is partial; some of the recovered animal exhibit permanent trembling gaits (Karki, 1996). Furthermore, animals treated with ivermectin (0.4 mg/kg body weight) had also responded to the treatment (Shrestha et al., 1997). These findings are additional indirect evidence that the cause of this syndrome was cerebrospinal nematodiasis.

CEREBROSPINAL NEMATODIASIS -A SHORT DESCRIPTION

Setaria labiatopapillosa (syn. *Setaria digitata*, *Setaria cervi*) occurs in the peritoneal cavity of cattle, buffaloes and deer. The parasites in the peritoneal cavity are not generally pathogenic. However, the immature forms in non-natural hosts like sheep and goats, cause cerebrospinal nematodiasis (lumbar paralysis) which is often fatal. Male parasites measure about 40-60mm and females measure about 60-120mm. Morphologically, the peribuccal ring and dorsal and ventral prominences are distinct. Mouth opening is elongated. The tail of the female terminates in a marked button, which is divided into a number of papillae (Fig. 1). The microfilaria are sheathed and measure about 240-260 microns. Microfilaria are mechanically transmitted by culicine mosquitoes. The second stage larva are ingested by mosquitoes in which development of L3 stage takes place. These microfilaria are transmitted from mosquitoes to other animals by bite. In non natural hosts, after bite, microfilaria may enter the spinal cord or the central nervous system leading to clinical manifestations of paralytic signs. Sheep and goat act as erratic hosts where there is no further development of the third stage larva. The disease occurs in the summer and autumn, when the mosquitoes intermediate hosts are active. Limited works done so far in Nepal (Karki, 1996; Shrestha et al., 1997).

RECOMMENDATION

Until further investigation, the treatment and control of posterior paralytic syndrome in goats Diethylcarbamazine citrate (Banocide, Hetrazan) at the dose rate of 10mg /kg body weight for three days or ivermectin 0.4 mg/kg body weight S/C together with vitamin B complex injections and vector control is recommended. Diethylcarbamazine citrate could also be used as prophylactic treatment during disease outbreak season. In endemic areas, animals could be strategically treated with this drug at the dose rate of ... during the autumn months, which is the principal season for disease outbreak.

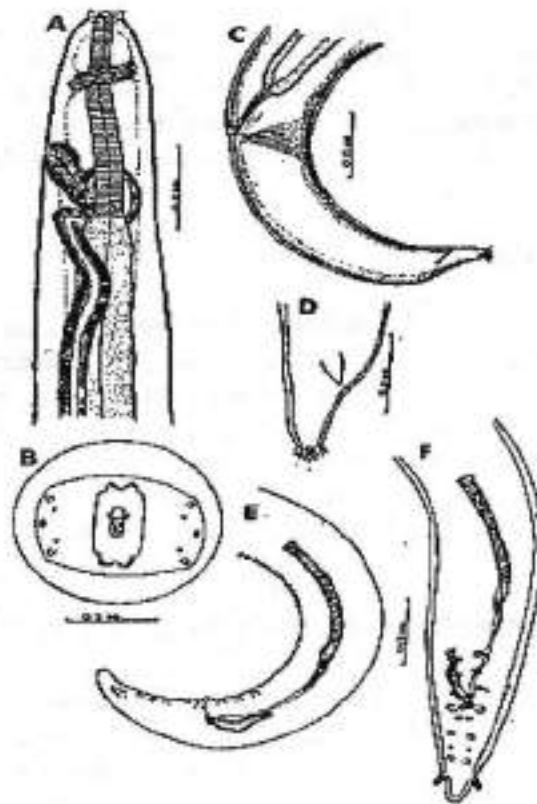


Figure 1. Morphology of *Setaria* spp. A. Female anterior end. B. Anterior end, *en face* view. C. Female posterior end. D. Female posterior end, enlarged. E. Male posterior end, lateral view. F. Male posterior end, ventral view (From Yeh, 1959)

ACKNOWLEDGMENT

We extend our sincere thanks to Dr. S.C.Ghimire, who gave critical suggestions to prepare this clinical paper. We also thank to the technical staff of District Livestock Services Office, Banke who helped in various aspects of this study.

REFERENCES

- Karki, K.B. (1996). [Paralysis in goats; cerebrospinal nematodiasis (in Nepali). Veterinary Chaumasik, Issue I, 2053, pp. 25-26.]
- Shrestha, S.P., Shah, A., Karki, K.B. and Adhikari, B.N. (1997). Posterior paralytic syndrome in goats in Banke district of Mid-Western Region of Nepal. Unpublished report.

EFFICACY OF TRICLABENDAZOLE AND OXYCLOZANIDE AGAINST FASCIOLOSIS OF LACTATING CATTLE AND BUFFALOES

K.P. Acharya¹

ABSTRACT

This study was conducted in three locations in the Eastern Terai of Nepal to evaluate the efficacy of triclabendazole and oxclozanide against fasciolosis of cattle and buffaloes. The study was conducted between January 1996 and March 1996. Of the 317 lactating cow and buffaloes examined, 21.6% (40 / 186) cows and 30% (40 / 131) buffaloes were positive for *Fasciola* infection. Treatment of infected cows with either Oxclozanide or triclabendazole was highly effective as determined by the absence of *Fasciola* eggs in their faeces 15 days after treatment. However, buffaloes did not respond as effectively as cows in either treatment groups. There was a significant species difference ($P < 0.005$) between cows and buffaloes in the effectiveness of the drugs in treating fasciolosis. An average increase of 320 ml of milk per lactating animals 40 days after the treatment was recorded, which was not statistically significant ($P = > 0.05$). However, a significant increase ($P < 0.05$) in milk yield of 970 ml per animal was observed only in group of buffaloes treated with oxclozanide.

INTRODUCTION

Liver fluke infection or fasciolosis causes hepatic insufficiency in cattle, buffaloes, sheep and goats. *Fasciola hepatica* is principally a parasite of sheep and goats and is widely distributed in temperate countries. *Fasciola gigantica* is known to be more pathogenic and occurs among cattle, buffalo and goat population in tropical and sub-tropical countries. Fasciolosis is manifested by acute, subacute or chronic symptoms.

Fasciola species mature in the bile duct of the host. The bile carries eggs from the bile duct away to the environment through faeces. Hatching of the eggs is influenced by the ambient temperature and moisture. Under optimum condition hatching of eggs takes place within 10-12 days, which may be protracted or restricted if the ambient temperature is not appropriate. Miracidium released from the eggs enter the intermediate host - snail of *Lymnaea* spp. within 24-36 hr or die. The common species of *Lymnaea* snail found in Nepal are *L. auricularia race rufescens*, *L. auricularia sensu stricto*, *L. viridis* and *L. luteola* (Mahato, 1993). Habitat such as over irrigated land, land with small streams, or slow moving water favors the breeding and survival of snails. In Nepal the *Lymnaeae* species was found up to the altitude of 2100 meters (Mahato, 1993). Cercaria released from the snail encyst in the herbage around the water. Encysted cercaria, metacercaria, are the source of infection to ruminants.

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In Nepal, fasciolosis is one of the most common diseases of ruminants. Studies done so far indicates its high prevalence (>50%) in most of the areas of Nepal (Mahato, 1993; Shrestha et al., 1992; Mahato et al., 1992; Ghimire and Karki, 1996). However, the majority of these studies were done in the hills, and there are only limited studies done Terai (Mahato, 1993). The economic losses due to fasciolosis in Nepal was estimated to be Rs. 14.2 crore (Lohani and Rasali, 1995). Mahato (1993) estimated the loss of meat production in Nepal due to fasciolosis to be worth of Rs. 72 crore resulting from reduction of 95,000 tonnes of buffalo meat and 4,220 tones of goat meat per year. In other countries, it is accepted that chronic fasciolosis reduces beef production by 10%, lamb/mutton production by 25 % and milk production by 16% (Gracey and Collins, 1994).

The best way of controlling fasciolosis in Nepal is by strategic drenching (Mahato, 1993). However, there have been only limited studies to study the effectiveness of commercially available anthelmintics in Nepalese cattle and buffaloes. This study was therefore undertaken to study the the prevalence of Fasciolosis in Eastern Terai of Nepal, compare the efficacy of triclabendazole and oxclozanide against liver fluke in lactating cattle and buffaloes, and study the effect of drenching on milk production.

MATERIALS AND METHODS

This study was carried out between late January and early March 1996 in four villages of three districts of Eastern Terai, which included Ranjani and Babiya Birta of Morang district, Topa of Saptari district, and Dhubi of Sunsari districts. All locations fell in areas that were well irrigated by canals and annual rainfall of 1780 - 2030 mm. Semi-stall feeding was adopted in all sites. Only lactating cows and buffaloes were included for this study.

Technicians supporting the study were initially oriented and trained in the methods and procedures to do the work efficiently in the field and in the laboratory. A total of 186 faecal sample from lactating cows and 131 samples from lactating buffaloes were examined for the presence of *Fasciola* eggs. The eggs were identified by the methods as described by Soulsby (1982). All the samples were examined by sedimentation method.

Animals positive for *Fasciola* eggs were divided into two groups, one group (20 buffaloes, and 20 cattle) was drenched with oxclozanide and other group with triclabendazole (20 cattle and 20 buffaloes). Oxclozanide (Clozan 3.4%) was drenched at the rate of 10 mg per kg body weight while triclabendazole (Fasinex Oblet 900 mg triclabendazole) was administered at the rate of 12 mg per kg body weight.

Faecal sample from all treated animals was examined 15 days post treatment. In this study 'efficacy' of a drug was defined as its apparent ability to stop shedding of *Fasciola* eggs from the treated animals 15 days post-treatment. Daily milk production record of individual animal before and 40 days post treatment was collected by farmers interview. Only animals available for recording by the end of study were included for analysis.

Chi squared test was used to analyse differences in treatment response between cattle and buffaloes. Paired comparisons were made using t-test on daily milk production before and

after the drenching assuming that the milk production and their differences were normally distributed.

RESULTS

Prevalence of fasciolosis

A total of 186 cattle and 131 buffaloes were examined for the presence of *Fasciola* eggs in their faeces. The Prevalence of fasciolosis was 21.5% and 30.5% for cattle and buffaloes, respectively (Table 1).

Table 1. Prevalence of fasciolosis in cattle and Buffaloes in the Eastern Terai

Species	Proportion positive for <i>Fasciola</i> eggs	Percent positive
Cattle	40 / 186	21.5
Buffalo	40 / 131	30.5
Total	80 / 317	25.2

Though more buffaloes were affected with fasciolosis, the difference was not significant. Magnitude of prevalence depends upon the activity of intermediate host, feeding practices, ecozones, rainfall and temperature.

Response to anthelmintic treatment

Of the sixty one treated animals still available at the end of the study, 31 were cows and 30 were buffaloes. This included 17 cattle and 19 buffaloes treated with oxclozanide and 14 cattle and 11 buffaloes treated with triclabendazole. Fifteen days after the treatment, all cattle were negative for *Fasciola* eggs while 11 buffaloes were still positive (Table 2). The difference in the treatment response between cattle and buffaloes was highly significant ($P < 0.005$) indicating a species variation in response to treatment. Both drugs eliminated at least all adult flukes in cattle but not in buffaloes.

Table 2. Efficacy of oxclozanide and triclabendazole in treatment of chronic fasciolosis in milking cows and buffaloes

Anthelmintic	Proportion positive for <i>Fasciola</i> eggs 15 days post-treatment	
	Cattle	Buffaloes
Oxclozanide	0 / 17	5 / 19
Triclabendazole	0 / 14	6 / 11
Total	0 / 31	11 / 30

Within a species, no significant difference ($P > 0.05$) was observed in the efficacy of either drugs (Table 2).

Impact on milk production

The difference in milk production of individual animal before drenching and 40 days after drenching was analysed. The analysis was done to see the effect of species, treatment regime and their interaction (Table 4). Significant increase in mean milk production of 970 ml was observed only in buffaloes drenched with oxyclozanide. The overall slight increase in milk production observed after treatment was mainly due to increase in that group.

Table 4. Summary of paired comparisons on milk yield before and after the treatment with oxyclozanide and triclabendazole

Comparisons	Spp.	sample size	Drug	Av. milk yield (l/day)		Average difference	% change	t-Statistic	p-value
				Before treatment	After treatment				
1	C	14	O	3.14	2.77	-0.38	-11.93	-0.91	0.19
2	B	19	O	3.45	4.42	0.97	28.24	2.54	0.01
3	C	17	T	3.00	3.18	0.18	5.88	1.46	0.08
4	B	11	T	4.05	4.32	0.27	6.74	0.35	0.35
5	C & B	33	O	3.32	3.72	0.40	12.10	1.33	0.10
6	C & B	28	T	3.41	3.63	0.21	6.28	0.77	0.23
7	C	31	O&T	3.06	2.99	-0.07	-2.37	-0.36	0.36
8	B	30	O&T	3.67	4.38	0.72	19.55	2.03	0.03
9	C & B	61	O & T	3.36	3.68	0.32	9.39	1.53	0.07

C=Cattle, B= Buffalo, O= Oxyclozanide, T= Triclabendazole, Av= Average

Though statistically insignificant, an overall increase of 320 ml (9.39%) of milk per animal per day was observed after the drenching. However, the limitation of this study means that several other confounding factors that may influence the milk yield have not been recorded and analysed to determine the true impact of milk production due to treatment alone. Random allocation of animals from a large population into treatment groups and the same season of study, however, should have minimised the effects of other confounding factors. The distribution of differences is shown in figure 1.

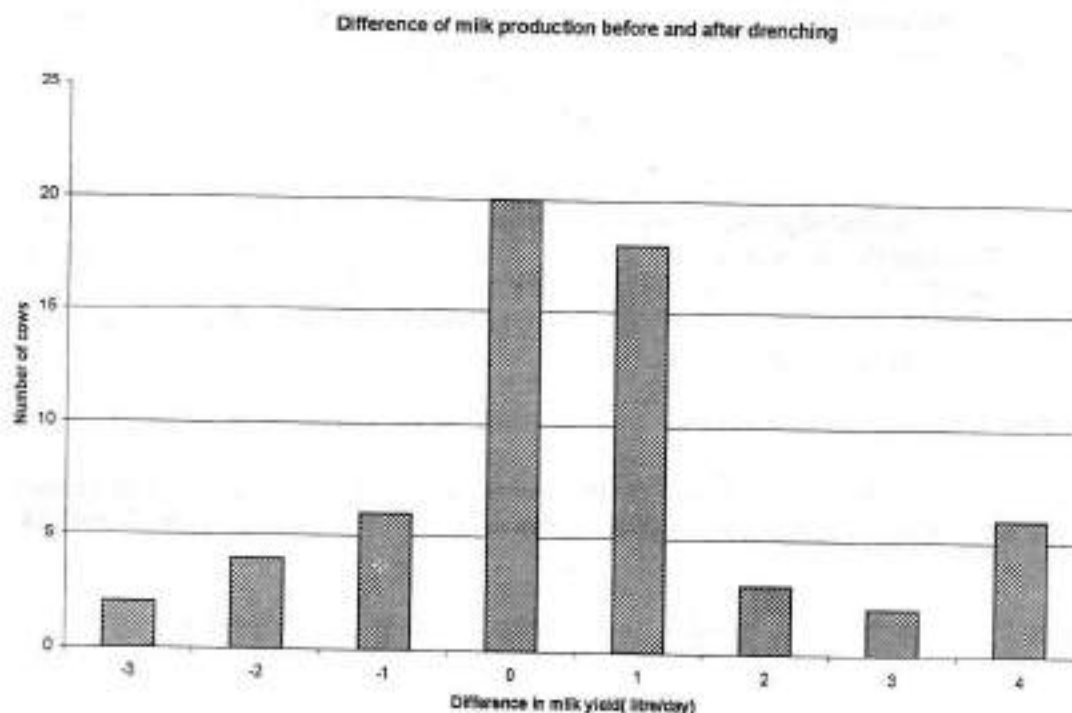


Figure 1. Frequency distribution in the difference in milk production before and after drenching

Though overall increase in milk production was not significant, drenching as a whole was found useful.

DISCUSSION

All the positive animals had symptoms chronic fasciolosis. In Nepal, it is chronic fasciolosis that mostly occurs which results from ingesting small number of metacercariae over a longer period of time.

The response to therapy with either drug had a significant species difference with the treatment. Both drugs were highly effective in cattle but not effective in more than 50 % of buffaloes. Triclabendazole has been reported highly effective against all stages of liver fluke in cattle, and treatment response trials conducted in Nepal in the past also supported this (Shrestha et al, 1992). Mahato and Rai (1992) reported that triclabendazole was not effective against the liver fluke of buffalo calves when they were treated with dose rate of 12 mg per Kg body weight (recommended dose). However, when the dose was doubled i.e. 24 mg per Kg, the drug was effective against liver flukes in buffaloes. In another study, rafoxanide was most effective (efficacy 67%) as compared to the albendazole (efficacy 48%) and triclabendazole (efficacy 52%) in treatment of fasciolosis in buffaloes (Ghimire and Karki,

(1996). This study therefore supports the view that buffaloes respond differently to anthelmintic drenching and drugs recommended for cattle cannot and should not be blankly recommended for use in this species.

ACKNOWLEDGMENT

I would like to acknowledge the support and encouragement received from Dr S N Mahato, Chief AHD during the preparation of this paper. I appreciate the suggestions made by Dr S C Ghimire. I am thankful to Miss Devi K. Limbu, Mr Kumar S. Khatri, Mr. Rishav Dev Aryal, Prem K. Pokhrel and other staffs of Regional-Veterinary Laboratory, Biratnagar for their assistance in the field and laboratory works.

REFERENCES

- Ghimire, N.P. and Karki, N.P.S. (1996). Prevalence of fasciolosis and efficacy of various anthelmintics in buffaloes of rural Kathmandu. *Bull. Vet. Sc. An. Husb., Nepal*, 24: 108-111.
- Gracey JF, Collins DS. Meat Hygiene Ninth Edition. ELBS Bailliere Tindall 421-425 (1994) 24 108-111
- Lohani, M.N. and Rasali, D.P. (1995).
- Mahato, S.N. (1993).
- Mahato S.N. and K Rai. Efficacy of triclabendazole against Fasciolosis in Buffaloes. *Veterinary Review* 1992 7(1), 17-20
- Mahato SN, K Rai, IB Karki. Prevalence of Fasciolosis in calves in Dhankutta District. *Veterinary Review* 1992 7(1), 20-21
- Shrestha EK, Thakur RP, Dhakal IP, Mahato SN: *Veterinary Review*: (1992) 7(2): 47-49, PAC, Dhankutta.
- Soulsby E.J.L. Helminths, Arthropods and Protozoa of domesticated animals. Seventh edition ELBS& Bailliere Tindall 1982: 40-52.

A RETROSPECTIVE STUDY ON KHARI DISEASE IN DARCHULA AND BAITADI DISTRICTS OF NEPAL

Dr. B.K. Nirmal, D. P.S. Thaguna, Dr. D.P. Chaudhary

ABSTRACT

Monthly epidemiological reports of Khari disease between June 1996 and December 1998 from Darchula and Baitadi districts were analysed. During that period, out of 8210 cases reported 29 animals (0.35%) died while other animals recovered. A large number of cases were reported from December to March every year and almost all the sick animals were in lactation. The cases were reported from 700 m to 2000m altitudes and recurrence of the disease in the same animal was quite common.

INTRODUCTION:

Khari or chaukhari disease is a disease syndrome of unknown etiology recorded in buffaloes in Baitadi and Darchula districts since many years. The disease has become economically important because the milk production in affected animals drops and sometimes animal dies due to its poor body condition. District reports suggested that on an average 1500-2000 animals get affected by this disease every year in each district. The disease is characterised by general dermatitis, chronic wasting, abducted limbs, extrusion of scales and chalk like powder from the hooves of the sick buffaloes, lameness, recumbency and death in some cases (Ratala et al., 1992). The disease is suspected to be caused by various predisposing factors like malnutrition, mange mites, mycotoxicoses and poor management conditions of the farm.

There have been efforts in the past for diagnosis of this syndrome jointly by the Department of Livestock Services and Nepal Agriculture Research Council. The laboratory examination revealed increased eosinophil and monocyte and decreased lymphocytes values in affected animals when compared with the normal values for buffaloes. Examination of straw and grasses fed to the animals during investigation were mouldy and infected with *Aspergilla* sp. (Singh and Shrestha, 1996). They also observed severe nit (louse) infestation in animals affected with khari and found *Psoroptes* mites in skin scrapings. Experimental infection with blood and skin scraping from sick buffaloes to healthy one resulted in the development of skin lesions similar to natural infection, but not the hoof lesions, which ultimately recovered with a single dose of ivermectin injection. Ratala and Singh (1994) conducted a field trial to evaluate the response of ivermectin in affected animals. 75% of treated animals recovered by a single dose of ivermectin at the dose rate of 200 µg/kg. Buffaloes with Khari disease had consistent hypophosphataemia ranging from 1.5-2.2 mg%. Clinical symptoms observed in affected animals such as typical posture with abducted elbows and with diffuse dermatitis and skin cracks were also similar to those observed deficiency of phosphorus (Singh and Shrestha, 1996).

MATERIALS AND METHODS

Monthly epidemiological reports submitted to the Regional Veterinary Laboratory, Dhangadhi from District Livestock Services Offices of Baitadi and Darchula during the period June 1996 to December 1998 were compiled and analysed. Reports were grouped for three seasons; monsoon (June - September), winter (October - January) and spring (February - May). In addition, observations made during clinical examination and investigation of Khari disease in the district hospitals were also included.

RESULTS AND DISCUSSION

Epidemiological findings:

Total number of cases recorded during the study period is presented in Table 1. Mortality was high during June to September while the number of cases was high during October to January. Usually the disease is chronic affecting principally the milking animals. There is a seasonal increase in the number of cases between October - January. Affected animals tend to recover slowly after April when green season starts. The disease was more frequent in animals which were stalled and kept under closed shed with poor ventilation and light. The disease has been reported from altitude ranging from 700 m to 2000 m but is uncommon in river basin areas. Proper exercise of the animal often leads to self recovery.

Table 1. Number of cases of Khari disease in Baitadi and Darchula districts between June 1996 and December 1998

S.N.	Period	Number of cases			Total No. of cases	No. of animals dead	Case fatality %
		1996/97	1997/98	1998*			
1	June - September	194	93	55	342	7	2.04%
2	October - January	2030	1005	32	3067	9	0.29%
3	February - May	347	1409	-	1756	12	0.68%

*Only up to December

Animals affected with the disease have been treated with injections of phosphorus and or ivermectin and supplementation of mineral mixtures. For the skin lesions, topical application of herbal ointments such as himax or charmil are used. The success of treatment, however, is variable.

CONCLUSIONS

The disease occurs mostly during lactation period or early pregnancy in buffaloes and during October to March / April. Affected animals recover during lush pasture season. Though, the losses due to mortality is low, recurrence of the disease in affected animals is a serious concern to the farmers. There is a need to improve the feeding, housing and management practices in the area to reduce the losses. Furthermore, detailed investigations to identify the aetiology and recommendation for reduction of disease prevalence is required.

REFERENCES

- Ratala D.R.; Manandhar P. and Pant G.R. (1992). Preliminary study of *Khari* disease of buffalo in Baitadi district. *Bull. Vet. Sci. Anim. Husband., Nepal*, 19-20: .
- Ratala D.R. and Singh U.M. (1996). Study of therapeutic evaluation of ivermectin in the treatment of *Khari* disease in buffaloes of Baitadi district. Paper presented at the Fifth National Conference of Nepal Veterinary Association. 1996.
- Singh U.M. and Shrestha S.P. (1996) Study of *Khari* disease in buffaloes of Baitadi and Darchula districts of Nepal - Proceeding of the First National Livestock and Fishery Research Workshop 1996, Nepal Agricultural Research Council, pp 176-184.

AN INVESTIGATION OF MAREK'S DISEASE IN CHITWAN DISTRICT

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ABSTRACT

There was an outbreak of a disease in Chitwan district during spring and summer months of 1998. Field investigation was carried out in 16 affected farms. Affected chickens were layers of 12-36 weeks of age. Morbidity ranged from 4.6-35% and mortality rate from 4 to 30%. There were signs of paralysis, wasting and skin tumours in affected birds. Post-mortem findings included tumours of various sizes in ovary, liver, kidney, proventriculus, heart and sternal muscles, and enlarged sciatic nerves and plexus. Histopathological examination revealed heavy infiltration of lymphoid cells disrupting normal structures in sciatic nerve. Proventriculus, kidney and heart were also heavily infiltrated with lymphoid cells. Based on clinical signs, pathological lesions and laboratory findings, Marek's disease was confirmed in this outbreak.

INTRODUCTION

Marek's disease, also known as fowl paralysis or polyneuritis, is one of the ubiquitous avian infection which is identified among chicken flocks worldwide. Every flock except for those maintained under strict pathogen free conditions may be presumed to be infected (Fraser et al., 1991). Marek's disease is a lymphomatous highly contagious disease of poultry caused by DNA herpes virus in which lymphoid cells infiltration and demyelination of peripheral nerves is a common feature. There are three serotypes of the virus; type 1 includes all pathogenic strains, type 2 contains naturally occurring nonpathogenic strains and type 3 contains strains of turkey herpes virus (HVT). Clinical symptoms are emaciation, depression and paralysis starting at about 13 weeks of age (Jordan 1990).

The virus is present in the epithelium of the feather follicles. The virus can survive for months in poultry house litter or dusts. Dusts and dander can remain infective upto 16 weeks after removal of infected chicks and are responsible for transmission of disease. Marek's disease is not a vertically transmitted disease (Mohanty, 1990). The disease was previously suspected in Kathmandu in 20 of the 1433 postmortems conducted on poultry in 1996/97, but has not been confirmed by laboratory investigation (Annual Report CVL, 1997).

During spring and summer months of 1998, there were outbreaks of this disease in different farms of Chitwan and Kathmandu valley. This paper describes the detailed investigation of these outbreaks and histopathological confirmation of Marek's disease in Nepal.

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MATERIALS AND METHODS

Sixteen affected commercial poultry farms of Chitwan district were visited. Information regarding the outbreak were also collected from other sources including district livestock services office (DLSO), Chitwan, poultry forum, Chitwan and from commercial poultry farmers of the district. Records on number of bird population at each farm, number of cases and dead were collected. Sick birds were clinically examined and some of them were brought to DLSO, Chitwan and postmortemed. Some sick birds were also brought to Animal Health Research Division (AHRD), Tripureshwor, Kathmandu for detailed laboratory examination. Serum samples from sick birds were collected for serological tests. Tissue samples like ovary, heart, sciatic nerve and kidney were collected in 10% buffered formalin for histopathological examination. Record of post mortem examination of the Central Veterinary Laboratory during mid April- mid August, especially the cases from Chitwan district, was also were analysed.

RESULTS AND DISCUSSION

Field investigation

Epidemiological and clinical findings

The affected chickens were layer chickens of 10-36 weeks of age and were in heat stress ($>35^{\circ}\text{C}$). In general, there was depression of egg production, and droopiness, depression, recumbency, ataxia, lameness with variable level of leg paralysis was common in almost all the farms visited at the time. Neck paralysis in some of the affected flock was also observed. Weight loss and muscular wasting was also noticed. These chickens were not vaccinated against Marek's disease at the farm but were supposedly vaccinated at day old at the hatcheries.

Mean morbidity rate of 15 farms was 13.2% which ranged from 4.7 to 35% in different flocks. Mean mortality rate was 9.9%, but was as high as 30% in some flocks (Table 1). Morbidity and mortality rates was influenced by the level of management and biosecurity measures applied by the commercial farmers. In general, the disease was more serious in farms with poor biosecurity measures.

Table 1. Morbidity and mortality rate due to Marek's disease in fifteen farm of Chitwan district

Area	No. of farm	Breed	Age (week)	Total population	No. sick	No. dead	Morbidity %	Mortality %
Narayanpur	2	Hyline	18-20	8650	50	370	4.9	4.3
Krislnapur	8	Babcock	12	400	20	120	35.0	30.0
		and Hyline	12-28	7119	564	1282	25.9	18.0
Vijaynagar	4	Hyline	20-24	3750	11	165	4.7	4.4
Rampur	1	Babcock	32	700	40	106	20	15.1
Total	15			20619	685	2037	13.2	9.9

Postmortem findings

Postmortem lesions varied between and within the affected farms. Wasting of sternal muscles, greyish white tumours in skin specially ventral and lateral sides of breast, greyish white tumours in proventriculus, intestine and kidneys in few cases were evident. The majority of the birds had tumours in liver and ovary of up to 1 cm in diameter. Muscular tumors and tumors in heart were evident in the dead birds from only one farm. Typical swollen sciatic nerves with loss of striations (almost 3 times the normal size) and sciatic plexus were noticed. None of the birds had enlarged bursa, rather it was regressed. Trachea and lungs in majority of cases were normal. No significant changes was observed in the bone marrow. These findings were suggestive of Marek's disease. Visceral tumours in proventriculus, liver and spleen were also recorded by Gowda (1990) in cases of Marek's disease.

Laboratory investigation

Histopathology

Sciatic nerve showed heavy infiltration of lymphoid cells disrupting the normal structure. Spleen revealed vacuolation. Heart showed heavy infiltration of lymphoid cells in the myocardium. Proventriculus showed infiltration of lymphoid cells and cancerous tissue below serosa. Mucosal layer was quite normal. Bursa revealed necrotic area in bursal follicles.

Serology

Marek's disease (MD) virus was not detected in feather follicles by agar gel precipitation test. However antibodies against MD were detected in four serum samples collected from the affected areas. Whether these antibodies were from natural infection or in response to vaccination could not be determined.

Egg inoculation

Clinical specimens were also subjected to chicken embryo inoculation for isolation of Newcastle disease virus. All samples were found negative for Newcastle disease virus.

Retrospective analysis of postmortem records of Central Veterinary Laboratory

Postmortem records of Central Veterinary Laboratory between April and August 1998 were analysed. Detailed farm history from flocks that were diagnosed with Marek's disease was analysed to determine the morbidity and mortality and most susceptible age group affected (Table 2). Birds of 12-18 weeks of age were mostly affected, with the overall morbidity of 24.5% and mortality rate of 18%.

Table 2. Morbidity and mortality rate in layer chickens due to Marek's disease

Period	Farms affected	Age	Total population	No. sick	No. dead	Morbidity %	Mortality %
April to August 1998	14	12-18 weeks	16,745	1097	3016	6.4	18.01

Source: CVL (1998)

Though MD seems to have been in Nepal for sometime, factors responsible for influencing outbreak of MD at that time in a wider areas is difficult to outline. Some variant virus may break through the immunity barrier produced by conventional vaccine (Mohanty, 1990). Other factors that might have contributed for a wide scale outbreaks at that time could be

- Poor biosecurity measure or no biosecurity measures in most of the farms of Chitwan
- Severe stress due to high ambient temperature, inadequate space
- Mycotoxin contaminated feeds leading to immunosuppression
- Unawareness of the farmers about the disease, and
- Improper vaccine and vaccination

CONCLUSION AND RECOMMENDATION

Based on clinical signs, pathological lesions and histopathological studies, the outbreak was confirmed to be due to Marek's disease. This outbreak emphasises a need for a national policy on poultry disease especially to prevent introduction of non endemic disease, and proper control of endemic diseases through a strategic vaccination schedules based on the disease epidemiology in the country. Quality control of vaccines and biologicals including regular seromonitoring of vaccinated flocks has to get a priority. Extension activity has to focus on proper biosecurity measures and regular disinfection in the farm.

ACKNOWLEDGMENT

We are very much thankful to Poultry Forum, Chitwan and Technicians of DLSC Chitwan for helping the team in investigation of this disease. We are also thankful to Mrs.T. shrestha and Mrs.A. Jonchhen for preparation of the slides for histopathology. We are also thankful to JICA for providing the vehicle facility during the visit.

REFERENCES

- CVL (1998). Central Veterinary Laboratory, Tripureshwor, postmortem room records, April-August, 1998
- Fraser, C.M.; Bergerson, J.A.; Mays, A. and Aiello, S.E. (1991). *The Merck Veterinary Manual*, Marek's Disease, p 1585.
- Jordan, F.T.W. (1990). *Poultry Disease*, Marek's disease, p96
- Mohanty G.C (1990). Epidemiological survey of poultry diseases in India. *Pasudhan* Vol.5 Issue 1, Jan 1990
- Gowda S. (1990) Broiler Diseases in India. *Pashudhan* Vol.5 Issue1 P5