# RISK FACTORS ANALYSIS OF FASCIOLOSIS IN BUFFALOES IN AZAD JAMMU AND KASHMIR

I. Ahmad, A. Z. Durrani[*], K. Ashraf[**], K. Hameed[***], O. Khan, A.A. Abbasi[***], A. Shakar[*], N. Hameed, I. Haq[*] and A. Safdar[*]

Department of Veterinary Clinical Sciences, University of the Poonch, Rawalakot, (AJK)
[*]Department of Clinical Medicine and Surgery.
[**]Department of Parasitology, University of Veterinary and Animal Sciences, Lahore.
[***]Department of Zoology, Mirpur University of Science and Technology (MUST) Mirpur-10250 (AJK)
Corresponding author's email: vetmedicine2015@gmail.com

**ABSTRACT:** A study was conducted in districts Poonch and Mirpur, Azad Jammu and Kashmir to analyze the prevalence of Fasciolosis in buffaloes and its relation to some epidemiological factors on the basis of altitude. The faecal samples of buffaloes (n=852) were collected randomly from April 2012 to March 2013. The samples were examined by sedimentation technique while egg per gram (EPG) count was performed through McMaster technique. From 852 samples, 106 (12.44%) were found positive. The risk factors like altitude, season, age and feeding pattern were tested through Chi square and quantified through generalized linear model. Prevalence and mean EPG was significantly different ($p<0.05$) at altitudes 3000-6000 feet as compared to <3000 and >6000 feet. When compared, significant differences ($P<0.05$) in prevalence was observed in seasons, age groups and feeding patterns. The study revealed more prevalence of the disease in buffaloes at high altitude as compared to low lying areas of AJK.

**Key words**: Fasciolosis, Buffaloes, Azad Jammu and Kashmir and Altitudes.

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## INTRODUCTION

Azad Jammu and Kashmir (AJK) is located at the foothill of Himalayas, lies between 730º-750º longitude and 330º-360º latitude (Ahmad *et al*., 2012). The state has unique topographic distribution and is comprised of mountainous ranges with heights of 14202 feet in the north with an average annual rainfall of 1500 mm (Afshan *et al*., 2011). Livestock contributes 30-40% to the livelihood of the rural population in high altitudes as in Himalayas (Mir *et al*., 2013). Poor management and hygiene leads to various parasitic diseases in animals of the area. Fasciolosis is one of those known globally as neglected tropical zoonosis (Musotsi *et al*., 2017). *Fasciola hepatica (F. hepatica)* and *F. gigantica* are two causative agents of which first is more common in temperate while second in tropical regions (Amer *et al*., 2016). Information on the prevalence of parasitic diseases in different climatic conditions provides a basis for strategic control measures (Mir *et al.*, 2013) as fasciolosis is highly susceptible to climatic factors (Fuentes *et al*., 2001) and climate change affects the transmission (Mas-Coma *et al*., 2008).

In cattle and buffaloes, the disease appears in subclinical form (Schweizer *et al*., 2005) which causes heavy economic losses due to poor weight gain, reduced milk yield and mortality (Ejeh *et al*., 2015). The disease is endemic in Pakistan (Shahzad *et al*., 2012) and unique in having areas that range up to 13123 feet above sea level (Mas-Coma *et al*., 2008). Studies from Pakistan reported 41.83%, 30.83% and 53.50% from Larkana, Hyderabad and Badin, Sindh (Bhutto *et al*., 2012), 13.06% from Punjab (Khan and Maqbool, 2012), 27% from Rajanpur (Farooq *et al*., 2015), 21.7%, 16.7%, 15.6%, 14.4% and 10.6% from Yazman, Bahawalpur, Khairpur, Hasilpur and Ahmadpur, respectively (Hasanat *et al*., 2014) prevalence in buffaloes. The disease has been reported from AJK in sheep and goats (Afzal *et al*., 1997) but data are lacking regarding buffaloes. Hence, this study was designed to estimate the prevalence and associated risk factors of Fasciolosis in buffaloes at high altitudes of AJK.

## MATERIALS AND METHODS

The study was carried out in district Mirpur and Poonch during 2012-13. Fresh fecal samples from 852 buffaloes were collected randomly. The sample size was determined through Epi Info$^{TM7}$. Information regarding each buffalo was altitude, season, age and feeding pattern. Depending on climatic conditions of the two districts, the area was categorized into below 3000 (A), 3000-6000 (B) and above 6000 feet (C) on the basis of altitude. Keeping in view, grazing pattern of the area, the buffaloes were divided in to two age groups as young (≤2 year) and adult (>2 year) animals. Duration of the seasons at high altitudes was adjusted with low lying areas as winter (October-February), spring (March-

April), summer (May-July) and autumn (August-September). Feeding pattern like stall feeding, grazing and mixed grazing (buffaloes grazing with cattle and sheep) were analyzed for association with disease. Freshly passed fecal samples were collected, labeled and transported to the Parasitology laboratory, University of the Poonch, Rawalakot, AJK. The samples were analyzed using sedimentation technique while egg per gram (EPG) count was performed by McMaster technique according to Foreyt, (2001).

**Statistical analysis:** Chi square analysis was used on EpiInfo™ 7 (Centers for Disease Control, Atlanta, GA, USA). The association of epidemiological factors with disease was estimated through odds ratio and the corresponding 95% confidence intervals. Generalized Linear Model was used for further processing of the significant epidemiological factors. P-value less than 0.05 was considered significant. Analysis of variance was used to compare mean egg per gram of feces with reference to altitude, season, age group and feeding pattern.

## RESULTS

Overall prevalence of fasciolosis in buffaloes was 12.44%. Prevalence of the disease was 07.77%, 18.64% and 11.95% at altitudes A, B and C (Table-1). Significant difference (P<0.05) was observed among different altitudes. Odds ratio (OR) value indicated that buffaloes were 2.72 times more prone to fasciolosis. Seasonal prevalence of disease noted highest in winter (19.86%) followed by autumn (9.25%), summer (8.71%) and spring (7.42%). Statistical analysis revealed that prevalence was significantly higher (P<0.05) in winter season. The prevalence of the disease in young animals was found significantly different (P<0.05) from adults. Intensity of infection revealed a significant difference (p<0.05) in EPG among three different altitudes (Table 2). The EPG at altitude B showed a higher mean value 302.7273± 21.64 as compared to 175.8621±15.81 and 193.1818±18.98 at A and C, respectively. All other variables like season (winter 271.19±20.63, spring 226.32±26.59, summer 226.92±37.81 and autumn 183.33±23.24), age (young 231.58±35.91 and adult 248.28±15.06) and feeding pattern (stall feeding 204.17±21.48, grazing 235.71±32.21 and mixed grazing 268.52±13.87) showed non-significant (p>0.05) variation in EPG count (Table-2). Mixed grazing of buffaloes with sheep and cattle appeared to be the most significant factor in the generalized linear model (Table-3) contributing maximum variations in disease with highest Odds (4.30), followed by altitude B (2.51), age group adult (1.59) and season. Risk factors for fasciolosis in buffaloes were mixed grazing, altitude 3000-6000 feet, adult age group and winter season.

**Table 1. Prevalence of fasciolosis in buffaloes in Azad Jammu and Kashmir.**

| Variable | Type | Total no. of Samples | No. of samples positive | Prevalence% | Odds ratio | $\chi^2$value | p-value |
|---|---|---|---|---|---|---|---|
| **Altitude** | -<3000 | 373 | 29 | 07.77 | Reference Category | | |
| | -3000-6000 | 295 | 55 | 18.64 | 2.72 | 17.7 | 0.00002 |
| | >6000 | 184 | 22 | 11.95 | 1.62 | 2.59 | 0.11 |
| **Season** | Winter | 297 | 59 | 19.86 | Reference Category | | |
| | Spring | 175 | 13 | 07.42 | 0.32 | 13.17 | 0.00028 |
| | Summer | 218 | 19 | 08.71 | 0.38 | 12.16 | 0.0005 |
| | Autumn | 162 | 15 | 09.25 | 0.41 | 8.71 | 0.003 |
| **Age** | Young | 226 | 19 | 08.40 | 1.76 | 4.59 | 0.032 |
| **Groups** | Adults | 626 | 87 | 13.89 | | | |
| **Feeding** | Stall feeding | 286 | 24 | 08.39 | Reference Category | | |
| **pattern** | Separate grazing | 379 | 28 | 07.38 | 1.15 | 0.23 | 0.63 |
| | Mixed grazing | 187 | 54 | 28.87 | 4.43 | 34.46 | 0.000 |

**Table 2. Analysis of variance of intensity with fasciolosis in buffaloes in Azad Jammu and Kashmir.**

| Variable | Degree of Freedom | Sum of Square | Mean Square | F-value |
|---|---|---|---|---|
| **Altitude** | 2 | 5.362 | 2.68 | $8.31^{***}$ |
| **Season** | 3 | 0.867 | 0.289 | $0.89^{ns}$ |
| **Age group** | 1 | 0.233 | 0.233 | $0.707^{ns}$ |
| **Feeding Pattern** | 2 | 0.82 | 0.42 | $1.24^{ns}$ |
| **Residuals** | 97 | 31.98 | 0.32 | |

Signif..codes: '***' 0.001 '**' 0.01 '*' 0.05 $^{ns}$Non Signif

**Table 3. Generalized linear model for risk factors assessment of fasciolosis in buffaloes in Azad Jammu and Kashmir.**

| Epidemiological Factors | Estimate | Std. Error | OR value | Z value |
|---|---|---|---|---|
| **Intercept** | -2.66 | 0.37 | 0.07 | -7.09*** |
| **Altitude 3000-6000** | 0.92 | 0.26 | 2.51 | 3.57*** |
| **Altitude>6000** | 0.26 | 0.31 | 1.30 | 0.84ns |
| **Season spring** | -0.82 | 0.29 | 0.44 | -2.80** |
| **Season summer** | -1.06 | 0.34 | 0.34 | -3.13** |
| **Season autumn** | -0.92 | 0.32 | 0.40 | -2.84** |
| **Age group adult** | 0.46 | 0.28 | 1.59 | 1.67ns |
| **Feeding pattern grazing** | -0.15 | 0.30 | 0.86 | -0.49* |
| **Feeding mixed grazing** | 1.46 | 0.28 | 4.30 | 5.27*** |

Signif..codes: '***' 0.001 '**' 0.01 '*' 0.05 nsNonSignif
Null deviance: 640.07 on 851degrees of freedom
Residual deviance: 552.10 on 843 degrees of freedom
AIC: 570.1.

## DISCUSSION

Incidence of fasciolosis in highland area was influenced by hosts, parasites and environmental effects. The epidemiological control measures depended upon an effective forecast of the disease which included causative agents and their intermediate and final hosts (Bhutto *et al*., 2012). Study of seasonal trends and influence of geographic and climatic factors were very important to know the epidemiology of a disease for establishment of data for disease control strategy. The data revealed highest prevalence (18.64%) and mean EPG (302.7273± 21.64) of the disease at higher altitude between 3000-6000 feet when compared to low lying areas. These findings were found in agreement with Gebreyohannes *et al*., (2013); Walker *et al*., (2008) and Ahmad *et al*., (2005) where they reported high incidence of the disease at highlands as compared to low-lying areas. The highest prevalence of the disease at an altitude of 3000-6000 feet showed the climatic suitability for liver flukes and snails intermediate host during the months of June to September in the area. During these months animals harbour infection and after completion of the life cycle of the parasite shed ova during winter. Such differences of variation of the disease pattern appear to favour transmission and adaptation to high altitudes (Mas-Coma *et al*., 2001).

Seasonal trend of the disease was highest during winter and lowest during spring. Adult animals were found more affected. These findings correlate with the study of Qureshi *et al*., (2012) who reported 14.69% prevalence with highest during the month of September-October and lowest in May. Pandya *et al*., (2015) also reported highest prevalence in December and lowest in May from India. In a study from Bahawalpur, Punjab Hasanat *et al*., (2014) reported 15.8% disease with highest in September and lowest in May. Contrarily a very high rate of prevalence of fasciolosis in buffaloes (Bhutto *et al*., 2012 and Farooq *et al*., 2015) was also reported from different areas of the country. The difference might be due to difference in climate, temperature and humidity. The environment and management have strong relationship with fasciolids and its intermediate host snail population (Mas-coma *et al*., 2001). Adult animals showed more disease than in young ones. These findings were in agreement with the results of Hasanat *et al*., (2014); Qureshi *et al*., (2012); Maqbool *et al*., (2002) and Bhutto *et al*., (2012). An important epidemiological factor noticed in the present investigations was mixed grazing of buffaloes with cattle and sheep. The disease was significantly higher $p<0.05$) in mixed grazing pattern which elaborated the fact that fasciolosis was primarily a disease of sheep reported by (Urquhart *et al*., 1996). In mixed grazing system sheep sheds ova of the flukes which caused infection in other animals. These results were in agreement with the findings of Abebaw *et al*., (2012) and Sangma *et al*., (2012). These findings seem to be directly associated with number and species of animals kept under one roof or sharing water ponds and grazing fields.

**Conclusion:** It was concluded that this disease was more prevalent at high altitudes as compared to low lying areas. Mixed grazing of different species of animals with buffaloes aggravated the situations. Adult age of animals and winter season were the important risk factors of fasciolosis in buffaloes in AJK.

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