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# Epidemiology and Risk Factor Analysis of Fasciolosis in Buffaloes in District Bagh, Azad Kashmir, Pakistan

# EPIDEMIOLOGY AND RISK FACTOR ANALYSIS OF FASCIOLOSIS IN BUFFALOES IN DISTRICT BAGH, AZAD KASHMIR, PAKISTAN

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## ABSTRACT

Fasciolosis is a cosmopolitan parasitic disorder cause great economic decline in cattle and buffaloes in terms of poor carcasses quality decrease in meat and milk production, malfunctioning of livers and weight loss. This research work was carried out in district Bagh, Azad Jammu and Kashmir, Pakistan, extending from December 2016 to May 2017. During this study 200 fecal sample from Buffaloes (*Bablus bubalis*) of the study area were collected, it was found that 40 percent fecal samples contain eggs of *Fasciola hepatica* and 60 percent fecal samples were negative. Liver samples of slaughtered buffaloes were also examined during study period and it was found that 74 samples have faciola, out of 200 samples, so 37 per cent samples contain *F. hepatica* and 63 per cent buffaloes liver samples were negative. It was observed that 19.25% animals of the area have parasites in their body, which are causing economic losses to the owners, in the form of less production of milk and meat and more mortality among the livestock and damages to farmers.

**Keywords:** Epidemiology, buffaloes, faciola, livestock, fasciolosis.

## INTRODUCTION

Fasciolosis, a cosmopolitan parasitic disorder responsible for great economic decline in cattle and buffaloes in terms of poor carcasses quality, decrease in meat and milk production, malfunctioning of livers and weight loss (Maqbool et al., 2002). This disease cause major losses in terms of mortality and morbidity in fluky regions. Data collected from some countries of Asia has displayed that buffaloes suffer more frequently from Fasciolosis, than any other parasitic disease (Balasingam, 1962). In the world among all parasitic disorders of ruminants fascioliasis is one of the most common (Massoud et al., 2012). In advanced regions of the world data on epidemiology of several helminthiasis has been circulated and remedial measures have been taken in these areas for the control of this disease but in developing countries little research work has been done on Fasciolosis (Maqbool et al., 2002). Two liver flukes commonly reported to causes Fasciolosis in ruminants are *F. hepatica* and *F. gigantica*. The life cycle of these trematodes involves snail as an intermediate host. Infected cattle show poor weight increase and dairy cattle have lesser milk yield (Mihreteab et al., 2010). The geographical distribution of trematode species is dependent on the distribution of suitable species of snails (Rahmeto, 2010). Symptoms of Fasciolosis in buffaloes will be imminent after eighty four days eggs will be found in fecal samples have infection after thirteen to fourteen weeks for *F. hepatica* and *F. gigantica*

respectively by shedding eggs of flukes in excreta and up to this time most part of liver may be damaged.

The disease is responsible for obvious financial drop in livestock by declining milk production, weight gain, liver condemnation, fertility and disease is very commonly found in rainy areas of maximum parts of the world and absent where environmental circumstances are critical for the growth of secondary host (Mazhar, 2005). Fasciolosis is the major cause of economic losses in parasitized groups because of limited yield of meat and milk, mortality, abortions and rejection of livers in slaughterhouses, growth retardation in young animals and expenditure on monitoring of this disease. Alves et al. 2011 described that in Brazil, the growing topographical distribution of *F. hepatica* is due to transfer and trade of animals (parasitized) from enzootic disease areas where the localities are not affected but have satisfactory epidemiological surroundings. The encouraging conditions of area are suitable and ensure occurrence of snails (genus *Lymnae*) which are intermediate hosts for *F. hepatica* (Martins et al., 2012). Thus these situations such as adequate and high humidity temperature and rainfall also include the environmental and climatic features that deliver a favorable habitation for these snails, Feasibility of eggs upcoming from naturally infected buffaloes and cattle liability of *L. columella* collected and studied on the farmhouses, to infection by liver fluke (Dracz and Lima, 2014).

Major source of animal protein is derived from Meat of goats, cattle and sheep. There are various species of these but the economically important ones in tropics are *F. gigantic* and in the temperate region is *F. hepatica* (Usip et al., 2014). Numeral surveys have been done by veterinary research groups concerning the occurrence of the disease and studied in many parts of the world (Ozung et al., 2011). The climatic changes that have occurred in Bungoma in the recent past such as prolonged season of rains may have had an effect on the prevalence and economic importance of Fasciolosis on the rural population (Usip et al., 2014). For great milk yield among the livestock buffalo is thought to be an important kind. Total contribution of milk in country due to buffalo is seventy two percent (Mahadevan, 1978). Buffaloes are subject to the number of significant parasitic diseases. Field reports reveled that more than 27,000 buffaloes lost every year because of different diseases (Junejo and Qureshi, 1993). Milk is mainly produced by the smallholders in Pakistan (Afzal, 2010). Chaudhry et al. 1984 reported that in the alimentary tract, due to combination of many species of helminthic parasites gastrointestinal helminthiasis ailment is caused. Irfan observed in 1984 that in Pakistan where the sanitary situation is very low, the massive financial damages in livestock industry is because of parasites. In Pakistan, Fasciolosis is one of the major factor that is responsible for reduced livestock development (Kendall, 1954).

*Bablus bubalis* (Asian buffalo) belongs to the class Mammalia (Hussain et al., 2010; Afzal, 2010) as compared to other domesticated bovines species well thought out to be an advantageous animal, and agonized a huge number of internal and external parasitic bacterial, fungal and viral diseases (Raza et al., 2010; Mahfooz et al., 2008).

Helminthiasis, among all parasitic infections has long been documented and still is a criminal principal to deaths in ruminant growth in all across the globe (Alawa et al., 2010). Fascioliasis caused by genus Fasciola, is economically most important helminthes ailments of the farm-ruminants (Santos et al., 2015). It is an emerging parasitic pollution, impacts threateningly on both human health and veterinary worldwide (Lazara et al., 2010). It roots huge economic losses in terms of decrease in meat, milk and higher rate of

deaths in all forms of animals (Saleha, 1991).

Two extremely infectious species are identified as and *F. gigantica* and *F. hepatica* first species is normally prevailing in humid regions of various states of the biosphere while the other one endures in a diversity of climatic circumstances (Urquhart et al., 1988). These parasites mainly cause infection in the liver, where they exist in bile duct and hepatic parenchyma and lesion on mucosa and subsequent huge tissue damage (Shaikh et al., 2004, 2005). Proofs indicated that most deaths in bovine is because of fascioliasis (Losos, 1986).

*B. bubalis*, meat and the prime dairy producing animal not filling the demand and supply gap of milk and meat of the Pakistani people is due to variety of diseases being faced by domesticated animals (Bhutto et al., 2012). Universally, above seven hundred million household ruminants are at threat and economic loss go above 3 billion US dollar per year. Peoples do not gave due concentration to the Fasciolosis in the human population in tropical regions of the world where more than 50 million of the population has been affected. The prevalence rates in Bangladesh in live animals has been reported to vary from 19 to 51% in 8.4 to 31% in sheep (Sangma et al., 2012), buffaloes (Roy, 2016), 10 to 32% in goats (Sammadar et al., 2015) and 21 to 53% in cattle (Islam et al., 2015). In human fascioliasis was traditionally considered as a secondary disease. The explanation of many human endemic regions and the emergence of human reports in many countries, including both prevalence and intensity increases and topographical expansion, have drastically changed the global scenario from the middle of 1990s. Fascioliasis is at present known to be the vector-borne disease presenting the altitudinal, widest latitudinal, longitudinal and distribution known (McAllister and Sultana, 2011).

The global estimation of seventeen million people infected may be an underestimation due to the situation in many countries was previously not known (Mas-Coma et al., 2012). In temperate climatic zones, *F. hepatica* is tremendous significant parasitic of cattle (Bennema et al., 2011). Heavy infection was found in buffaloes as compared to cow and goat (Swarnaka and Sanger, 2014). Production of bovines yield about the earth is majorly affected by trematode invasion (Vercruysse and Claerebout, 2001). But Fasciolosis increases public to fear not only due to its zoonotic feature. But also its frequency and economic implication to live stock in many regions of the world Ruminants' Fasciolosis is an inhibition in beneficial bovine-farming and for slaughterers and customers too. Parasite of genus Fasciola i.e. *F. gigantica* and *F. hepatica* is the causal agent of disease (Fasciolosis) which occur in a wide range of ultimate hosts. It is encouraged on by both environmental changes (warmer, wetter climate) and man-made changes such as an increase in animal travels and growth of livestock farming (Gul, 2017).

## MATERIALS AND METHOD

### *Geographical area of Study*

The State Azad Jammu and Kashmir consist of 10 districts and Bagh is one of them. Climate of Bagh has been categorized as warm and temperate. In Bagh the normal annual temperature is 17.3°C. My study area was Bagh.

### *Fecal Samples*

200 stool samples of buffaloes were collected from different localities of city Bagh (During December, 2016 to May, 2017) for the determination of liver fluke eggs, stool samples were taken with the help of forceps. These samples were packed into labelled polythene bags and transferred to the lab. All the collected

samples were examined within 12 hours (Yildirim et al., 2007).

The presence of Fasciola was determined in these collected samples by following the sedimentation and floating technique (Urquhart et al., 1988). One gram of stool sample was taken and mixed with water (15 to 20ml) to make solution in cylinder and stir well with glass rod and strained with the help of strainer then put a drop of strained solution on the slide with the help of dropper and place the cover slip on the glass slide. This slide was examined under microscope for the presence of *F. hepatica*. The eggs were counted in each slide. Mask and gloves were used as a hygienic measure.

### *Liver Samples*

A total of 200 samples of liver were collected of slaughtered buffaloes from different slaughter houses of the study area located in Bagh The liver of freshly slaughtered domestic buffaloes were collected for the study. Firstly the liver was collected and then it was observed keenly, in the beginning, the bile ducts were opened for obtaining flukes. For generalized liver fluke infections incisions were made in different parts of the bile duct to find the fluke in the liver, gall bladder was also incised and then bile duct opened, first common bile was observed and then smaller ones. Flukes were with care taken up with the help of forceps and needles and thoroughly washed with tap water and then these flukes transferred in saline solution and observed with the help of hand lens.

## RESULTS AND DISCUSSION

During the present study of Fasciolosis in buffaloes of district Bagh, extending from December 2016 to May 2017. The presence of Fasciola was determined in these collected samples by following the sedimentation and floating technique (Urquhart et al., 1988). Among the collected samples 120 were found negative and 80 were positive having eggs.

During the present study liver samples were also collected from freshly slaughtered animals and these were examined for the presence or absence of Fasciola species. Among these 200 samples *F. hepatica* was found in seventy four samples. All the collected samples were examined within 12 hours (Yildirim et al., 2007). Overall prevelance of both liver and fecal samples is 19.25 percent.

**Table 1: Data collected through fecal examination.**

| Sr. No. | Place | Total | Male | Female | Hypher | Non Hypher | Fasoiola Present | Fasiola Absent |
|---|---|---|---|---|---|---|---|---|
| 1 | Arja | 25 | 1 | 24 | 2 | 23 | 8 | 17 |
| 2 | Dhuli | 30 | 2 | 28 | 3 | 25 | 13 | 17 |
| 3 | Baglor | 10 | 2 | 8 | 3 | 7 | 3 | 7 |
| 4 | Chatter | 13 | 0 | 13 | 1 | 12 | 5 | 8 |
| 5 | Hular | 20 | 1 | 19 | 1 | 18 | 8 | 12 |
| 6 | Neela butt | 20 | 0 | 20 | 0 | 20 | 8 | 12 |
| 7 | Dheerkot | 25 | 0 | 25 | 0 | 25 | 9 | 16 |
| 8 | Chamyati | 20 | 1 | 19 | 0 | 20 | 9 | 11 |
| 9 | Jhala | 7 | 0 | 7 | 0 | 7 | 3 | 4 |
| 10 | Mallot | 17 | 0 | 17 | 4 | 13 | 9 | 8 |
| 11 | Bypass | 13 | 2 | 11 | 3 | 10 | 5 | 8 |

**Table 2: The percentage of *F. hepatica* in fecal samples collected from Bagh.**

| Place | Fasciola present | Percentage |
|---|---|---|
| Arja | 8 | 32% |
| Dhuli | 13 | 43% |
| Baglor | 3 | 30% |
| Chatter | 5 | 38.46% |
| Hular | 8 | 40% |
| Neela butt | 8 | 40% |
| Dheerkot | 9 | 36% |
| Chamyati | 9 | 45% |
| Jhlala | 3 | 42.85% |
| Mallot | 9 | 53% |
| Bypass | 5 | 38.46% |

**Table 3: The percentage of *F. hepatica* collected from slaughter houses.**

| Month | Total | Male | Female | Hypher | Non Hypher | Fasciola Present | Fasciola Absent |
|---|---|---|---|---|---|---|---|
| December | 30 | 2 | 28 | 2 | 28 | 13 | 17 |
| January | 40 | 4 | 36 | 6 | 34 | 19 | 21 |
| February | 32 | 2 | 30 | 2 | 30 | 12 | 20 |
| March | 35 | 3 | 32 | 3 | 32 | 11 | 24 |
| April | 33 | 2 | 31 | 3 | 30 | 9 | 24 |
| May | 30 | 1 | 29 | 3 | 27 | 10 | 20 |

**Table 4: The percentage collected through monthly basis.**

| Month | Fasciola present | Percentage |
|---|---|---|
| December | 13 | 56.66% |
| January | 19 | 47.5% |
| February | 12 | 37.5% |
| March | 11 | 31.42% |
| April | 9 | 27.7% |
| May | 10 | 33.33% |
| **Total** | **74** | **100** |

(A) **(B)**

**Figure 1** (A, B)**: *F. hepatica* collected from liver of slaughtered animals.**

During the present study period, 200 samples of liver were collected from the freshly slaughtered animals (figures 2&3) and 200 stool samples from house hold buffaloes from December 2016 to May 2017. Among these 200 fecal samples, 80 buffaloes fecal samples were found positive for liver fluke, prevalence rate of Fasciolosis is 40%. It is found that Fasciolosis is caused by *F. hepatica* in the study area. This study shows that domestic ruminants are highly infected with *F. hepatica*.

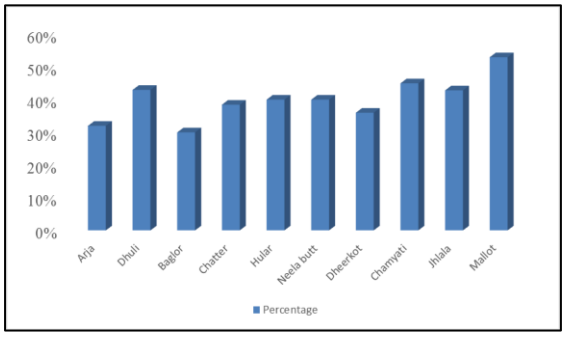


**Figure 2: The percentage of fecal sample collected from Bagh.**

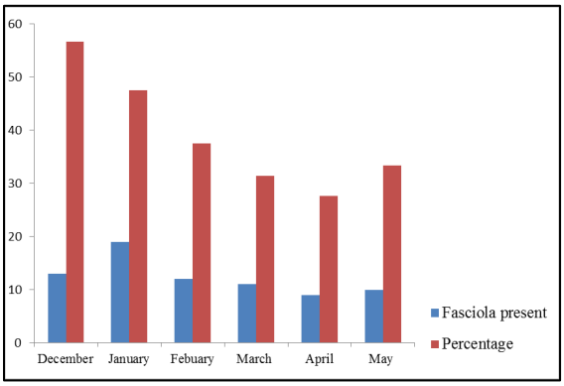


**Figure 3: The percentage of f. hepatica from liver of slaughtered animals collected on monthly basis.**

Table (1) reveals that *F. hepatica is* most prevalent in buffaloes of the study area. Fecal samples were collected from different places of Bagh like Arja, Dhuli, Baglor, Chatter, Hular, Neela Butt, Dheerkot, Chamyati, Jhala, Mallot and Bypass. From Arja 25 fecal samples (non hypher 23, hypher 2) during the examination of theses fecal samples it was found that Fasciola is present only in eight samples; 30 fecal samples were collected from area of Dhulli and it was found that 13 buffaloes were infected with liver fluke while 17 were not affected; 3/10 buffaloes

of Baglor were found positive, having Fasciola eggs in their excreta; From Chatter 13 fecal samples were examined and in 5 samples Fasciola was found and 8 samples were free of Fasciola; From Hular 20 fecal samples collected, Fasciola was present in 8 samples and absent in 12; twenty fecal samples were collected from Neelabutt and it was found that eight samples were positive for liver fluke and twelve were negative; From Chamyati 20 samples were examined and only nine found positive; from Jhala 7 fecal samples were collected and found three positive for Fasciola; From Mallot 17 fecal samples of buffaloes were collected and 9 positive. Total 13 fecal samples from Bypass, 5 were positive for Fasciola and 8 were free. Out of 200 buffaloes 80 were found infected by *F. hepatica* and prevalence rate is 40%, eggs of liver flukes were found in only 80 samples out of 200 and it is close to the results of Pfukenyi and Mukaratirwa, who reported 37.1% in Zimbabwe and Abraham and Jude found the prevalence rate 44.8% in Nigeria. So prevalence of liver fluke was thus higher in domestic ruminants. (Maqbool et al., 2002; Aliyu et al., 2014). 200 liver samples were also examined for the determination of rate Fasciolosis in buffaloes, only 74 samples found positive during the present study, showing overall 37%. For the determination of Fasciolosis prevalence in the animals of the study area, liver samples of slaughtered animals were collected from December to May, 2017. Month wise Prevalence rates from December to May 2017 are as 43.33%, 47.55%, 37.5%, 31.42%, 27.27% and 33.33% respectively. The highest prevalence rate is in the month of January, 2017 and lowest in the month of April, 2017. This variation may be due to de-worming of animals in different months; highest rate in the month of January, 2017 may be due to lowest cold temperature and less availability of food, weakness of animals and immune system. May be due to non-drenching of in the month of December and January. These are assumptions for the determination of accurate reason more detailed studied is needed. Carneiro et al., in 2010 reported natural infection by *F. hepatica* in buffaloes for the first time in the south of the state of Espirito Santo, with a positivity rate of 46.67 % among 15 samples. Higher incidences of fascioliasis have been recorded by Pfukenyi and Mukaratirwa, who reported 37.1 % in Zimbabwe and Abraham and Jude recorded 44.8 % in Nigeria.

Marques and Scroferneker conducted a study in 2003 in the state of Rio Grande do Sul and prevalence rate recorded by them was 10.34 % among 377 livers samples of cattle and 20 % in 105 livers samples of buffaloes. The result of the fecal samples are 37 and 40 % liver samples percent which is consistent with the result of Pfukenyi and Mukaratirwa 37.1 % in Zimbabwe, Abraham and Jude 44.8 % in Nigeria.

Alemu et al., (2015) described that Southern Ethiopia using post-mortem examination of liver of each slaughtered animal shows that A total of 500 cattle were examined for the presence of Fasciolosis using both coprological and post mortem examination .The prevalence under coprological and post mortem examinations were found to be 19.4% and 28.6% respectively. This result is lower than my results. Carneiro et al., studied in 2010 that the Fasciolosis in buffaloes of south state of Espirito Santo and found the infection rate 46.6% which is slightly higher to my results. Marques and Scroferneker 2003 carried out in study in the state of Rio Grando Sul and found Fasciolosis rate in fecal and liver samples 10.34% and 20% respectively which is not consistent to my results and manifold lower than my results. Rahman and Mondol in 1983 collected the 762 fecal samples in Bangladesh and found infection rate 66.14% which is higher than my results.

Karim, et al., (2015) in Bangladesh examined 762 fecal samples and found 504 (66.14%) positive with Fasciola species. Rahman and Mondol in 1983 and Gupta et al. in 2002 recorded 53 and 70% Fascioliasis in cattle in Bangladesh and India, respectively. Gupta et al. 2002 collected samples and found the prevalence rate 53% which is slightly higher than my results and 70% is too much higher than my findings. Epidemiological studies were undertaken by Maqbool et al., 2002 explained that in Punjab Lahore at slaughter houses and on household buffaloes under the different climatic conditions existing in Punjab province. Infection rate was 25.59 and 10.5 percent, respectively in slaughtered buffaloes and house hold buffaloes. These results are lower than my results.

## CONCLUSION

A study was conducted in districts Poonch and Mirpur, Azad Jammu and Kashmir by Ahmad et al., in 2017. They study the prevalence of Fasciolosis in buffaloes from 852 samples, 106 (12.44%) were found positive. The findings of this study are much lower than my results. The variations in the prevalence of fascioliasis may be attributed to the meteorological factors and grazing habits of the host (Shaikh et al., 2004). The inconsistence of my results with the neighboring areas studies may be due to difference of topographical variations.

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