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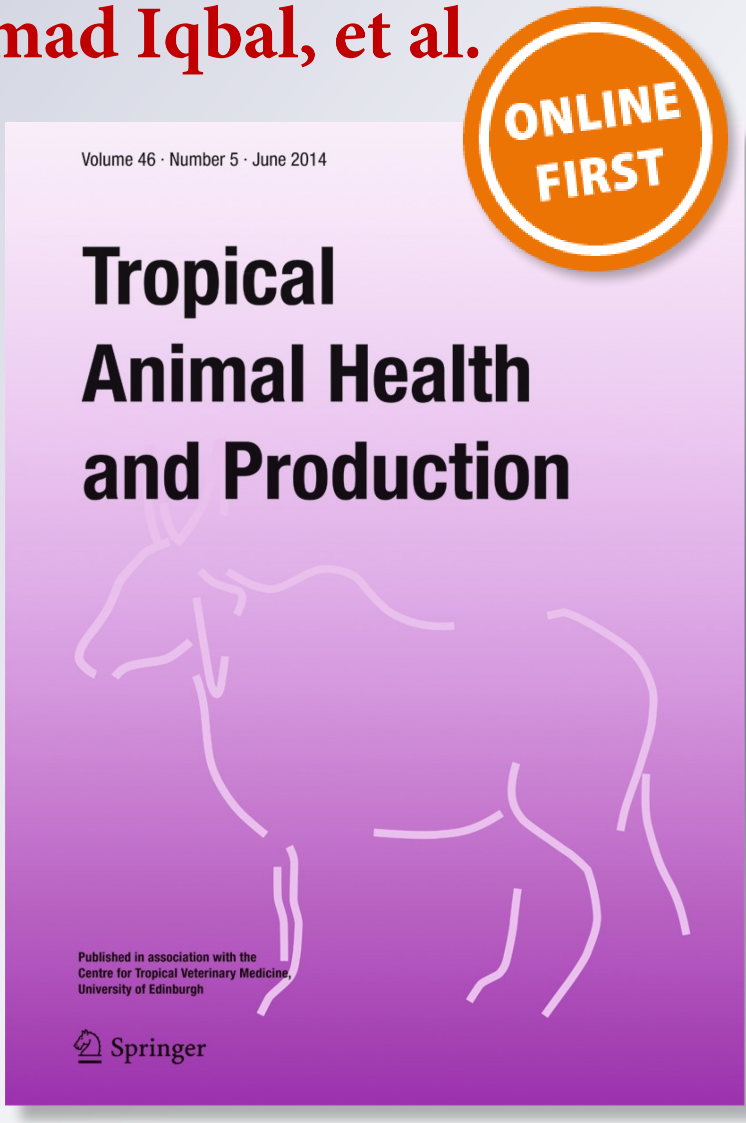

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REGULAR ARTICLES

Effect of management practices and animal age on incidence of mastitis in *Nili Ravi* buffaloes

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Abstract Buffalo is an economically important dairy animal in South Asia but mostly ignored in research priorities. In this retrospective study, the effect of management practices and age of animal on the incidence of mastitis in Nili Ravi buffaloes was investigated. A total of 1,560 quarters of buffaloes (n=390) were screened by visual examination of the udder and milk (clinical mastitis) and California mastitis test (subclinical mastitis). Household data was collected on a predesigned questionnaire and analyzed. The prevalence of subclinical mastitis, clinical mastitis, and blind quarters was 41.8, 13.6, and 9.7 %, respectively. The highest prevalence was noted in the hind quarters and left side as compared to that in the forequarters and right side. This data significantly ($p<0.05$) supported the idea that larger herd size has more chances of mastitis, with the highest prevalence (40, 32, and 27 %) in the large, medium, and small herds, respectively. Stage of lactation was significantly ($p<0.01$) involved in mastitis, and the highest incidence (43.3 %) was noted in early lactation. Milk production of lactating buffaloes that ranged 6–10 l/day showed a higher rate of mastitis occurrence ($p<0.05$). The cleanliness condition of a farm also contributed significantly. Animal age significantly affected the incidence of mastitis. Results revealed that age of the animal has a positive correlation (R^2=0.772) with mastitis. This study concluded that some factors alone or in combination with other factors influence significantly the occurrence of mastitis, and to minimize the infection, these factors should be considered. The outcome of the study will be valuable for policy-making for positive management practices and implementation of preventive measures.

Keywords Buffalo · Mastitis · Lactation · Age · Management practices

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Introduction

Buffalo (*Bubalus bubalis*) is acknowledged as the black gold of Asia and is a major dairy animal in Pakistan. Pakistan is considered as the top 2 largest buffalo milk-producing country in the world (F.A.O. 2008). Pakistan's dairy industry is composed of mostly buffaloes and cattle, contributing 95 % of total milk production, and the share of buffalo in total milk is more than 75 % (Jamil et al. 2011). In addition, buffalo milk is more preferred by consumers, and buffalo farmers are getting higher prices relative to cattle milk (Javaid et al. 2009). Currently, Pakistan harbored three breeds of buffaloes, namely Nili Ravi, Kundi, and Azekhili. The Nili Ravi breed of buffalo is contributing about 75 % of the total milk produced in the country (Sharif et al. 2009) and is a dynamic breed under field situation. Additionally, this breed is also used to provide beef, traction, and tilling power. Domesticated water buffalo population is around 172 million in the world and 27.33 million in Pakistan (Anonymous 2006). However, buffalo farming is integrated with the agricultural system, and the majority (85 %) of herd size is very small (Khan 2002). In Europe, Italy is the only country where buffalo production has fetched

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popularity in the recent years, and the reasons are the attractiveness of *Mozzarella* cheese from buffaloes and buffalo milk production is free of quotas in the European community (Moroni et al. 2006; Fagiolo and Lai 2007).

Bovine mastitis is considered as the most costly, common, and complicated disease in dairy animals worldwide (DeVliegher et al. 2005). Mastitis is the inflammation of the mammary glands with physical, chemical, and microbiological changes, characterized by an increase in somatic cells, especially the leukocytes, in the mammary tissues (Radostits et al. 2000). Clinical mastitis is characterized by abrupt onset, inflammation of the udder, pain, and reduced and altered milk secretion from the affected mammary tissues. However, in subclinical form, there are no apparent physiological changes in the udder and no visible defects in its secretion, and it is more prevalent (15 to 40 times) than the clinical one (Khan and Muhammad 2005; Mekibib et al. 2010; Hussain et al. 2013). Mastitis adversely affects milk quality and quantity and comprises a reservoir of microorganisms that spread the infection to other animals within the herd. This disease is also a risk for the spread of zoonotic diseases like tuberculosis, brucellosis, etc., where milk acts as a medium of pathogens causing these diseases (Shoshani et al. 2000), and contributes to antimicrobial residues as a result of drug therapy (Andrew et al. 2009). A higher rate of mastitis is a great threat to the dairy industry, especially the subclinical type, badly affects milk production and magnitude which eventually impart losses to the dairy farmers (Halasa et al. 2007), increases the risk of getting clinical mammary infection in the succeeding lactation, and increases the risk of premature culling in the herd (Parker et al. 2007). In addition, bovine mastitis in early lactation imparts long-term production losses to the dairy industry (Ahmadzadeh et al. 2009; Sharif and Muhammad 2009). The reported annual losses of the dairy industry due to mastitis are approximately $2 billion and $526 million in the USA and India, respectively (Varshney and Naresh 2004; Donovan et al. 2005). Similar economic losses are also observed in Europe and other countries (Denis et al. 2009).

Three major factors are involved in this disease: the microbes as the contributory agent, the host animal, and the environment, which can influence both the animal and the microbes. The infectious agent enters through the milk canal, interacts with the mammary tissue cells, and multiplies. The mammary tissues react to these agents and become inflamed. Environmental and management factors influence the incidence of mastitis in dairy animals (Nyman et al. 2007; Steeneveld et al. 2008). In the environment, a certain risk factor exists with a high probability to affect the prevalence of mastitis in buffaloes; however, the degree of influence by these factors is unknown in buffaloes. Most of the published work about these risk factors is pertaining to dairy cattle, and information about buffaloes is scarce (Salvador et al. 2012). Therefore, the present study was conducted with the following objectives: (1) to investigate the effects of different management practices on the incidence of mastitis in Nili Ravi buffalo, (2) to assess the effect of age on mastitis, and (3) to study the interaction effects of various management practices on the occurrence of mastitis.

Materials and methods

Before the initiation of the live phase of this research work, a study protocol was approved by the technical and supervisory committee of the Faculty of Animal Husbandry and Veterinary Sciences, The University of Agriculture Peshawar, Pakistan.

Study setting, universe, and study population

The current study was carried out in the district of Dera Ismail Khan (D.I. Khan), Khyber Pakhtunkhwa, Pakistan, where six villages were randomly selected among 30 villages with high buffalo density. Topographically, D.I. Khan is situated at the brim of the Indus River and is considered to be one of the hottest areas in Pakistan, where summer temperature ranged from 30 to 48 °C. Its area could be divided into three zones, viz irrigated areas, rain-fed areas, and adjacent areas to the Indus River. A total of 1,560 quarters of apparently healthy and lactating Nili Ravi buffaloes (n=390) were screened for the detection of subclinical mastitis, clinical mastitis, and blind quarters. Excluding the clinical mastitis and blind quarters, samples of 1,465 quarters were collected for the detection of subclinical mastitis. Animals in the first week of lactation and those in the last month were not included due to the possibility of false-positive results (Radostits et al. 2000).

In this study, the effect of different husbandry practices regarding the management of herd on the incidence of mastitis was assessed in Nili Ravi buffaloes, through conducting interviews in the survey phase of the study. The actual milk samples taken from the selected households were analyzed in the laboratory at Livestock Research and Development Station, D.I. Khan for actual confirmation and investigation of the management effects. Different husbandry practices were studied such as herd sizes classified into small herd sizes (1–5 buffaloes), medium herd sizes (6–10 buffaloes), and large herd sizes (≥11 buffaloes); type of farms (*kaccha*—made of mud, semi-*pakka*—made of bricks and mud, and *pakka* farms—made of bricks and concrete); stage of lactation (early 1–3 months, mid 4–7 months, and late 8–10 months); average milk yield/day (≤5, 6–10, and ≥11 l), washing of udder (simple water, disinfectant, and none); hygiene conditions of herds (poor, moderate, and good); ventilation status of the shed (poor, moderate, and good); milking methods (*stripping* and *fisting* or *knuckling*); and age of the animals based on lactation number (first, second… sixth lactation). Equal numbers of samples were studied from each category and all the information were collected on a predesigned questionnaire. Nili Ravi buffaloes (n=22) from the Livestock Research and Development Station, D.I. Khan were also included in this study.

Laboratory work

Clinical mastitis was determined by visual symptoms of the mammary glands and changes in milk morphology, while subclinical mastitis was identified by the California mastitis test (CMT). CMT was used for the detection of subclinical mastitis as it is the easiest and superior diagnostic cowside test for detection of subclinical mastitis (Suojala et al. 2011).

Preparation of sample bottles

Sample bottles (25 ml) with screw-tight caps were used for the collection of milk samples. The bottles were first washed with tap water, air dried, and sterilized in a hot air oven at 160 °C for 1 h. The bottles were then labeled as LF (left front), LH (left hind), RF (right front), and RH (right hind), and the label also showed the owner's name, identification of the animal, and collection date.

Collection of milk samples

The procedure described by the National Mastitis Council Inc., USA (1990) was followed for the collection of quarter milk samples. Prior to the collection of milk samples, the udder was bathed with tap water and dried with separate paper towel. Subsequently, the quarter was disinfected with cotton swabs soaked in 75 % ethyl alcohol. The vial was placed horizontally and the cap was removed without touching the inner surface. Mixed milk samples of 5 ml were collected separately from all four quarters after discarding the first few streams.

California mastitis test

On-farm CMT was performed for detection of subclinical mastitis. The test was carried out under a clean and isolated environment in a farm separated from the shed and its contamination. The CMT kit (Techni. Vet., Inc. USA), composed of alkyl aryl sulfonate (3 %), sodium hydroxide (1.5 %), and bromocresol purple (1:10,000) as indicator, was used in this study. CMT was carried out and scored as described by Schneider and Jasper (1964).

Statistical analysis

Raw data was entered in Microsoft Excel 2007. The data was statistically analyzed by using Statistical Package for Social Sciences (SPSS) version 16. The prevalence data was calculated using simple percentage values and frequencies, while the effect of different management practices and age (independent variables) on incidence of mastitis (dependent variable) was analyzed by using chi-square test (Pearson's chi-square test). The interaction effects of different parameters were analyzed by multiple variant regression. Correlation of age with mastitis incidence was also analyzed. Statistical significance level was set at p value <0.05.

Results

Prevalence of mastitis in Nili Ravi buffaloes

The overall and quarterwise prevalence of mastitis in Nili Ravi buffaloes is shown in Table 1. The overall prevalence of subclinical mastitis, clinical mastitis, and blind quarter was found to be 41.8, 13.6, and 9.7 %, respectively. A total number of 330 quarters were affected with subclinical mastitis, and out of these, 64 (19.39 %) in right front, 66 (20.00 %) in right hind, 88 (26.67 %) in left front, and 112 (33.93 %) in left hind quarters were noted. The results of clinical mastitis revealed that out of the total of 57 affected quarters, right front quarters were 11 (19.29 %), right hind 15 (26.32 %), left front 7 (12.28 %), and left hind 24 (42.11 %). Similarly, the prevalence of blind quarters in Nili Ravi buffaloes showed that out of the total of 38 blind quarters, right front were 6 (15.79 %), right hind 10 (26.32 %), left front 9 (23.68 %), and left hind quarters 13 (34.21 %).

Effect of management practices on mastitis

The effect of different management practices is presented in Table 2. In this study, we investigated the significant effect of herd size on the incidence of mastitis, with the highest prevalence in large herds (40.3 %) than in medium herds (32.2 %) and small herds (27.5 %). Although in *kaccha* types of farms the prevalence of mastitis was the highest (38.6 %) followed by semi-*pakka* (32.7 %) and *pakka* types of farms (28.7 %), the disease occurrence was not significant. Stage of lactation had a significant ($p<0.01$) effect on the prevalence of mastitis. The highest incidence (43.3 %) was noted in the early lactation, followed by late lactation (38.6 %) and mid lactation (18.1 %). Furthermore, the milk yield in buffaloes had shown significant effects on the incidence of mastitis. The highest rate (63.7 %) was found in high milk-producing animals having 6–10 l yield/day. This investigation lacked the incidence in the highest group of production (≥11 l) because buffaloes in this group were scarce. Hygienic measures in the herds also influenced significantly the rate of mastitis. Farms with poor hygiene conditions had the highest rate of

Table 1 Overall and quarterwise prevalence of mastitis in Nili Ravi buffaloes (sample population=390)

Quarter	Subclinical mastitis (%)	Clinical mastitis (%)	Blind quarters (%)
Overall prevalence	41.8	13.6	9.7
Right front quarter	19.39	19.29	15.79
Right hind quarter	20.00	26.32	26.32
Left front quarter	26.67	12.28	23.68
Left hind quarter	33.93	34.21	34.21

disease (46.8 %), whereas 31.0 and 22.2 % cases were investigated in moderate and good hygiene condition farms, respectively (Table 2). Although the effect of milking methods on the incidence was not significant ($p>0.05$), however, in the two commonly used milking methods, the incidence of mastitis was high (53.8 %) in *stripping*, and in *fisting*, mastitis was low (46.2 %) as shown in Table 2.

Effect of age on mastitis occurrence

In this section of investigation, we found the effect of age/lactation number on the incidence of mastitis in buffaloes. The linear regression line has given the best fit as the value of correlation coefficient (R^2) was high (R^2=0.772) as shown in Fig. 1. This model clearly revealed that as the number of lactation or age of buffaloes increased, the incidence of mastitis also increased. Correlation between the age of the animal and the incidence of the disease showed a significant effect ($p<0.01$) and was positively interlinked. This demonstrated that with the increase in lactation number, there is also an increase in the incidence of mastitis.

Table 2 Effect of different management practices on the incidence of mastitis in Nili Ravi buffaloes

Parameters	Percentage	p value
1.1 Small herds	27.50	0.033
1.2 Medium herds	32.20	
1.3 Large herds	40.30	
2.1 *Kaccha* farms	38.60	0.769
2.2 Semi-*pakka* farms	32.70	
2.3 *Pakka* farms	28.70	
3.1 Early lactation	43.30	0.05
3.2 Mid lactation	18.10	
3.3 Late lactation	38.60	
4.1 Low milk yielder (≤5)	27.50	0.02
4.2 Medium milk yielder (6–10)	63.70	
4.3 High milk yielder (≥11)	08.80	
5.1 Poor hygienic condition	46.80	0.049
5.2 Moderate hygienic condition	31.00	
5.3 Good hygienic condition	22.20	
6.2 Stripping method	53.8	0.916
6.3 Fisting method	46.2	

Interaction effects of different management practices

Table 3 presents the interaction effects of various management practices contributing to mastitis. Some of the management practices alone (like ventilation in shed and milking methods) did not show significant results in influencing the disease; however, certain parameters when looked in connection to certain other parameters also showed significant ($p<0.01$) interaction effects.

Discussion

The present retrospective study was an attempt to investigate the prevalence of mastitis, effect of management practices, and animal age on mastitis in Nili Ravi buffaloes. Firstly, it addresses the quarterwise prevalence of mastitis in Nili Ravi buffaloes, and secondly, it deals with the effects of management practices on the incidence rate of mastitis. In this regard, we are able to assess and document the potential risk factors for the incidence of mastitis. Thirdly, the effect of age on mastitis is studied and investigated, and it was found that with the increase in age, the incidence rate of mastitis also increased in buffaloes. Mastitis is the most common disease in

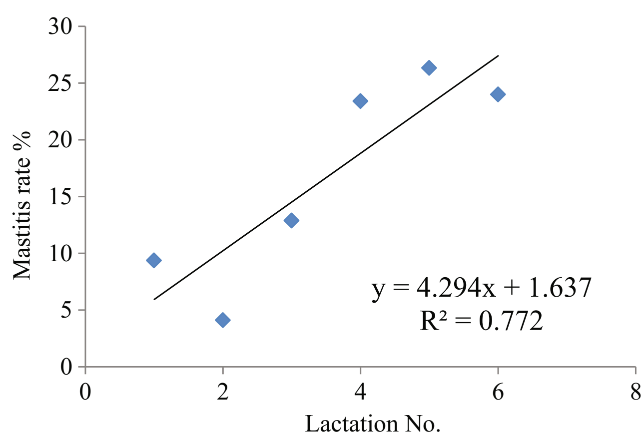

Fig. 1 Animal age relationship with incidence of mastitis in Nili Ravi buffaloes at D.I. Khan. The *linear regression line* has given the best fit and predicts a significant linear relationship between animal age/lactation number (*x* component) and mastitis incidence (*y* component) in Nili Ravi buffaloes. The value of correlation coefficient (R^2=0.772) suggested that with the increase in age, the incidence of mastitis also increased

Table 3 Interaction effects of different management practices observed in the studied farms at D.I. Khan

Parameters	Model fitting criteria	Likelihood ratio tests		
	−2 log likelihood of reduced model	Chi-square	*dof*	*p* value
Ventilation × average yield	194.65	26.11	4	0.002
Washing of udder × average yield	196.34	27.80	4	0.000
Type of farm × average yield	219.31	50.77	4	0.000
Stage of lactation × average yield	192.42	23.88	4	0.000
Hygiene × ventilation	207.59	39.04	4	0.000
Washing of udder × ventilation	186.73	18.18	4	0.001
Type of farm × ventilation	206.35	37.81	4	0.000
Stage of lactation × ventilation	193.25	24.71	4	0.000
Ventilation × herd size	201.75	33.21	4	0.000
Washing of udder × hygiene	186.78	18.24	4	0.001
Type of farm × hygiene	193.53	24.99	4	0.000
Hygiene × herd size	204.74	36.19	4	0.000
Type of farm × washing of udder	184.00	15.46	4	0.004
Stage of lactation × washing of udder	190.26	21.71	4	0.000
Washing of udder × milking methods	179.89	11.35	2	0.003
Washing of udder × herd size	181.07	12.52	4	0.014
Type of farm × stage of lactation	189.06	20.52	4	0.000
Type of farm × herd size	194.91	26.37	4	0.000
Stage of lactation × milking methods	179.52	10.97	2	0.004
Stage of lactation × herd size	202.05	33.51	4	0.000

dof degrees of freedom

dairy animals and subclinical mastitis prevalence is found multifold to clinical mastitis. Due to the presence of strong muscles at the opening of the teat canal, buffaloes are considered less susceptible to mastitis than cattle. However, we found a higher prevalence of mastitis in Nili Ravi buffaloes. Khan and Muhammad (2005), by using Surf field mastitis test (SFMT) and bacteriological examination of quarter milk samples from 50 buffaloes, also found similar results, reporting 27, 4, and 10 % prevalence of subclinical, clinical mastitis, and blind quarters, respectively. The results of other studies regarding the prevalence of mastitis in buffaloes showed a variation ranging from 6.0 to 87 % (Khan 2002; Liaqat 2009; Salvador et al. 2012; Hussain et al. 2013). This variation in occurrence depends upon several factors which contribute to the incidence of mastitis such as age of the animal, stage of lactation, cleanliness at the farm, and management of the owners. The risk of mastitis may be increased by lactational ketosis in periparturient immunosuppressed dairy animals because many types of immune cells are negatively affected by altered concentrations of metabolites, i.e., low concentrations of glucose and high concentrations of ketone bodies (Suriyasathaporn et al. 2000). The current study also showed that there was a higher occurrence in hind left quarters as compared to that in front right quarters. Some studies on mastitis also presented the same results of higher frequency in left hind quarters (Khan 2002; Khan and Muhammad 2005; Hussain et al. 2013). This higher incidence may be due to the exposure of hind quarters more frequently to dung, insects, urine, and other excreta and also the effect of direct sunlight and probable traumas onto the hind quarter's skin. Secondly, the sitting habit of the animal also exerts more pressure on the hind and left side. It has also been observed that normally the milkmen sit on the left side of the cow and start milking from the hind left teat first because of more milk in the hind quarters. This might exert more pressure on the left hind teat and, as a result, caused mastitis more often in that quarter.

Larger herd size (≥10 animals) significantly affected the prevalence of mastitis in our study. In a large herd size, management is more difficult as compared to that of small and medium herds. The authors had also observed that larger herds have more chances of infectious diseases and their spread. That might lead to cause the disease. Larger herd size has a positive effect on mastitis both in cattle and buffaloes, and this has been supported by the several scientists (Bilal et al. 2004). During the early stage of lactation, the effect of physiological stresses and various contaminations predispose the animal to different pathogens including mastitis-causing organisms during parturition. At the early stage of lactation (soon after parturition), milk production gradually increases and causes stress in the newly calved animal which resultantly led to mastitis. These conditions collectively made the animal prone to get the infections. In our study, the stage of lactation showed a highly significant ($p<0.01$) influence on the incidence of mastitis. Most of the scientists supported our results because mastitis was recorded more frequently in the early stage of lactation (Lalrintluanga et al. 2003; Almaw et al. 2008; Petrovski et al. 2009; Salvador et al. 2012). However, Liaqat (2009) has reported that mastitis occurrence is higher in later lactation. Similarly, milk yield in buffaloes had shown significant effects on the incidence of mastitis. In high milk-producing animals, physiological stresses negatively influence the immune system, and the physiological and anatomical condition of the teats and teat canal may make the quarter prone to injury and microbe exposure, which may lead to mastitis. The findings of several studies (Almaw et al. 2008; Petrovski et al. 2009; Gordon et al. 2013; Hussain et al. 2013) are in agreement with our study, and the authors of these studies also observed that with the increase of animal milk production, the incidence of mastitis was increased. Cleanliness and good hygienic measures are key factors to prevent diseases. These measures minimize the chances of pathogen survival. Inadequate sanitary measures and unhygienic conditions pave the way for the growth of microbes in the vicinity which can cause mastitis more frequently. The present study also reported that in herds where poor hygiene conditions prevailed, the incidence rate of mastitis was also higher than in farms where good hygiene measurements were practiced. The present findings are also supported by other researches (Biffa et al. 2005; Liaqat 2009).

Our study showed that the age of buffaloes had a positive correlation and a highly significant effect on the incidence of mastitis in buffaloes. Animal age presented a linear relationship, and the correlation coefficient ($R^2=0.772$) suggested that with increase in age, the incidence of mastitis also increased. Generally, with advancing age of the animal, there is a gradual suppression of the immune system, and also anatomical changes in the mammary glands are observed. These all contribute to increase chances of mastitis with increase in age. A similar positive link of age with mastitis was also previously reported (Ahmed and Mohammed 2009; Liaqat 2009; Salvador et al. 2012). However, the study of Bilal et al. (2004) was in discordance, as they reported maximum incidence of mastitis by third lactation, and after then, a decline was observed in mastitis incidence. They furnished the reason that most of the animals achieved maximum production up to third lactation and then a decrease was observed in milk. This affects the incidence pattern of mastitis.

Mastitis is a multifactorial problem, and various intrinsic and extrinsic factors are involved in the development of the infection. There were substantial differences in the identified risk factors in the various models. Some of the factors were in agreement with the reported literature, while others were not. This highlights the multifactorial nature of the disease and the differences in the risks for both mastitis manifestations. Attempting to understand these multifactorial associations for mastitis within larger management groups continues to play an important role in mastitis control programs. Some of the husbandry practices alone, such as ventilation in the shed and milking methods, did not show significant results in influencing the disease; however, certain parameters when looked in connection to certain other parameters also showed significant ($p<0.01$) interaction effects. Significant ($p<0.05$) interaction effects were also observed between the type of farm and washing of the udder, washing of the udder and milking methods, and stage of lactation with milking methods.

In conclusion, mastitis should be properly monitored and prevented in Nili Ravi buffaloes, and different husbandry practices should be kept in consideration while combating this economically devastating disease. The interaction effects of various management practices, such as the interaction of washing of the udder and ventilation status of the farm, produced significant effects on mastitis, thus supporting the idea of interdependency of various parameters in causing mastitis in buffaloes. The control strategies should be based on the improvement of management practices since these factors predispose animals to mastitis. Knowledge of the potential risk factors is vital for the control of bovine mastitis. In general, mastitis can be prevented by implementation of various hygienic/preventive measures.

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Conflict of interest The authors declare that they have no competing interest.

References

Ahmed Abdel-Radyand, Mohammed Syed, 2009. Epidemiological studies on subclinical mastitis in dairy cows in Assiut governorate, Veterinary World, 2(10), 373-380

Ahmadzadeh, A., Frago, F., Shafii, B., Dalton, J.C., Price, W.J., McGuire, M.A., 2009. Effect of clinical mastitis and other diseases on reproductive performance of Holstein cows, Animal Reproduction Science, 112, 273-282

Almaw, G., Zerihun, A., Asfaw, Y., 2008. Bovine mastitis and its association with selected risk factors in small holder dairy farms in and around Bahir Dar, Ethiopia, Tropical Animal Health and Production, 40, 427-432

Andrew, S. M., Moyes, K.M., Borm, A., Fox, L.K., Leslie, K.E., Hogan, J.S., Oliver, S.P., Schukken, Y.H., Owens, W.E., Norman, C., 2009. Factors associated with the risk of antibiotic residues and intramammary pathogen presence in milk from heifers administered prepartum intramammary antibiotic therapy, Veterinary Microbiology, 134, 150-156

Anonymous, 2006. Livestock census report of Pakistan, published by the Statistical Department, Government of Pakistan, Islamabad

Biffa, D., Debela, E., Beyene, F., 2005. Prevalence and risk factors of mastitis in lactating dairy cows in Southern Ethiopia, International Journal of Applied Research in Veterinary Medicine, 3(3), 189-198

Bilal, M.Q, Iqbal, M.U., Muhammad, G., Avaisand, M., Sajid, M.S., 2004. Factors affecting the prevalence of clinical mastitis in buffaloes around Faisalabad district (Pakistan). International Journal of Agriculture and Biology, 6(1), 185-187

Denis, M., Wedlock, D.N., Lacy-Hulbert, S.J., Hillerton, J.E., Buddle, B.M., 2009. Vaccines against bovine mastitis in the New Zealand context: What is the best way forward?, New Zealand Veterinary Journal, 57, 132-140

DeVliegher S., Barkema, H.W., Opsomer, G., Kruif, A.D., Duchateau, L., 2005. Association between somatic cell count in early lactation and culling of dairy heifers using Cox frailty models, Journal of Dairy Science, 88, 560-568

Donovan, D.M., Kerr, D.E., Wall, R.J., 2005. Engineering disease resistant cattle, Transgenic Research, 14, 563-567

Fagiolo, A., Lai, O., 2007. Mastitis in buffalo, Italian Journal of Animal Science, 6 (2), 200-206

F.A.O., 2008. The Economic and Social Department the Statistical Division, Food and Agriculture Organization of United Nations, Rome

Gordon, P.F., van den Borne B.H, Reist, M., Kohler S, Doherr M.G., 2013. Questionnaire-based study to assess the association between management practices and mastitis within tie-stall and free-stall dairy housing systems in Switzerland, BMC Veterinary Research, 9, 200. doi: 10.1186/1746-6148-9-200

Halasa T., Huijps K., Osteras O., Hogeveen H., 2007. Economic effects of bovine mastitis and mastitis management: A review, Veterinary Questions, 29, 18-31

Hussain, R., Javed, M.J., Khan, A., Muhammad, G., 2013. Risks factors associated with subclinical mastitis in water buffaloes in Pakistan, Tropical Animal Health and Production, 45, 1723-1729

Jamil, H., Samad, H.A., Rehman, N., Qureshi, Z.I., Lodhi, L.A., 2011. Effect of somatic cell types and culture medium on in vitro maturation, fertilization and early development capability of buffalo oocytes, Pakistan Veterinary Journal, 31, 105-108

Javaid, S.B., Gadahi, J.A., Khaskeli, M., Bhutto, M.B., Kumbher, S., Panhwar, A.H., 2009. Physical and chemical quality of market milk sold at Tandojam, Pakistan, Pakistan Veterinary Journal, 29, 27-31

Khan, A.Z., Muhammad, G., 2005. Quarter-wise comparative prevalence of mastitis in buffaloes and crossbred cows, Pakistan Veterinary Journal, 25, 9-12

Khan, M.S., 2002. In: Water buffaloes for food security and sustainable rural development in Pakistan. Regional workshop on water buffaloes for Food Security and Sustainable Development (Thailand, Saran). pp 111-116

Lalrintluanga, C, Ralte, E.L., Hmarkunga, 2003. Incidence of mastitis, bacteriology and antibiogram in dairy cattle in Aizawl, Mizoram, Indian Veterinary Journal, 80, 931-932

Liaqat Ali, 2009. Study on epidemiology of mastitis in dairy buffalo and cow in Tehsil Samundri of district Faisalabad. Ph.D thesis, Dept. of Vet. Clinical Medicine and Surgery, Univ. Agri. Faisalabad

Mekibib, B., Furgasa, M., Abunna, F., Megersa, B., Regassa, A., 2010. Bovine mastitis: Prevalence, risk factors and major pathogens in dairy farms of Holeta Town, Central Ethiopia, Veterinary World, 3, 397-403

Moroni, P., Sgoifo Rossi, C., Pisoni, G., Bronzo, V., Castiglioni, B., Boettcher, P.J., 2006. Relationships between somatic cell count and intramammary infection in buffaloes, Journal of Dairy Science, 89, 998-1003

National Mastitis Council Inc., 1990. Microbiological procedure for the diagnosis of udder infection, National Mastitis Council Inc. Wilson Boulevard Arlington, USA

Nyman, A-K., Ekman, T., Emanuelson, U., Gustafsson, A.H., Holtenius, K., Waller, K.P., Sandgren, C.H., 2007. Risk factors associated with the incidence of veterinary-treated clinical mastitis in Swedish dairy herds with a high milk yield and a low prevalence of subclinical mastitis, Preventive Veterinary Medicine, 78, 142-160

Parker, K.I., Compton, C., Anniss, F.M., Weir, A., Heuer, C., McDougall, S., 2007. Subclinical and clinical mastitis in heifers following the use of a teat sealant pre-calving, Journal of Dairy Science, 90, 207-218

Petrovski, K.R., Heuer, C., Parkinson, T.J., Williamson, N.B., 2009. The incidence and aetiology of clinical bovine mastitis on 14 farms in Northland, New Zealand. New Zealand Veterinary Journal, 57,109-115

Radostits, O.M., Gay, C.C., Blood, D.C., Hincheliff, K.W., 2000. Mastitis. In: Veterinary Medicine: A text book of diseases of cattle, sheep, pig, goat and horses. 9th ed., London: W.B. Saunders, pp. 603-700

Salvador, R.T., Beltran, J.M.C., Abes, N.S., Gutierrez, C.A., Mingala, C.N., 2012. Prevalence and risk factors of subclinical mastitis as determined by the California Mastitis Test in water buffaloes (*Bubalis bubalis*) in Nueva Ecija, Philippines, Journal of Dairy Science, 95(3), 1363-1366

Schneider, R, Jasper, D.E., 1964. Standardization of the California Mastitis Test, American Journal of Veterinary Research, 25, 163-165

Sharif A, Ahmad, T., Umer, M., Bilal, M.Q, Muhammad, G., Sharif, M.A., 2009. Quarter based determination of milk lactose contents and milk somatic cell count from dairy buffaloes under field conditions in Pakistan, Pakistan Journal of Zoology, 9 (Suppl), 313-32

Sharif, A., Muhammad, G., 2009. Mastitis control in dairy animals. Pakistan Veterinary Journal, 29, 145-148

Shoshani, E., Leitner, G., Hanochi, B., Saran, A., Shpigel, N., Berman, A., 2000. Mammary infection with *Staphylococcus aureus* in cows: progress from inoculation to chronic infection and its detection, Journal of Dairy Research, 67,155-157

Steeneveld, W., Hogeveen, H., Barkema, H.W., Van Den Broek, J.R., Huirne, B.M., 2008. The influence of cow factors on the incidence of clinical mastitis in dairy cows, Journal of Dairy Science, 91, 1391-1402

Suojala, L., Pohjanvirta, T., Simojoki, H., Myllyniemi, A.L., Pitkala, A., Pelkonen, S., Pyorala, S., 2011. Phylogeny, virulence factors and antimicrobial susceptibility of *Escherichia coli* isolated in clinical bovine mastitis, Veterinary Microbiology, 147, 383-388

Suriyasathaporn, W., Heuer, C., Noordhuizen-Stassen, E.N., Schukken, Y.H., 2000. Hyperketonemia and the impairment of udder defense: a review. Veterinary Research, 31, 397-412

Varshney, J.P., Naresh, R., 2004. Evaluation of homeopathic complex in the clinical management of udder diseases of riverine buffaloes, Homeopathy, 93(1): 17-20