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Research Article

Epidemiological findings of Lumpy skin disease outbreak investigations carried out in Cattle Colony Karachi, Pakistan

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Abstract

The objective of this study was to describe the epidemiological findings of Lumpy skin disease (LSD) outbreak in Cattle Colony Karachi. In total nine LSD affected farms were visited. Blood (n=7), skin scabs (n=2) and nasal swabs (n=6) were collected from LSD suspected cattle and data was obtained on pre-designed proforma. The samples were analysed for LSD using PCR. The most commonly observed clinical signs were nodules on the skin, fever, lachrymation, nasal discharge and reduction in milk production. The clinical signs of LSD were only observed in cattle in affected farms. The overall morbidity and mortality rates were 15.3% and 6.8% respectively. Of 15 samples collected from suspected LSD cases two (13.3%) were found positive for LSD using PCR. This is the first study describing epidemiological features of LSD outbreak in Cattle Colony Karachi, Pakistan. This study provides baseline information for further epidemiological studies and establishment of LSD control strategy in Pakistan.

Keywords: Cattle; Epidemiology; Lumpy Skin Disease; Outbreak Investigation; Pakistan.

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1. INTRODUCTION

Lumpy Skin Disease (LSD) is a transboundary animal disease (TAD) affecting cattle (*Bos taurus and B. indicus*) and water buffalo (*Bubalus bubalis*). The disease is caused by LSD virus (LSDV) of genus *Capripoxvirus* and family *Poxviridae* (Namazi & Khodakaram Tafti 2021). The incubation period of the disease may varies from 1-4 weeks. The most common clinical signs of the disease include fever (40-41 °C), formation of nodules in the skin, swelling of superficial lymph nodes, lachrymation, salivation, nasal discharge, difficulty in movement and sometime death of animal. The infection may lead to abortion, infertility in bulls and cows, weight loss, decrease in quality of hides and decrease in milk production (Tuppurainen et al. 2005); (Tuppurainen & Oura 2012). Recent published reports suggest that morbidity of disease may varies from 5%-45% and the

mortality normally less than 10% in cattle (Azeem et al. 2022).

The disease transmission mostly occurs through blood sucking vectors mainly, mosquitoes (*Aedes*) and biting flies (*Stomoxy*). However, ticks have also been reported to be involved in mechanically transmitting the virus from infected to susceptible animals. In addition transmission through artificial insemination and direct contact with infected animals has also been reported (Azeem et al. 2022).

First LSD case was reported from Zambia in 1929 (Namazi & Khodakaram Tafti 2021). Since then disease has been reported from South East Europe and Asian countries including neighboring countries of Pakistan i.e., India, China and Iran (Azeem et al. 2022). Recently, Pakistan notified LSD to World Organization for Animal Health (WOAH) on 4th March 2022. According to this report the initial outbreak was reported in Bahawalpur district of Punjab province, and Jamshoro and Thatta districts of Sindh province. Since then the disease has spread to other districts of Sindh province especially to Cattle Colonies in Karachi despite the implementation of various control measures by livestock authorities. On the request of Livestock Department Sindh and Animal Husbandry Commissioner Office, Islamabad, Pakistan Agricultural Research Council Scientists conducted outbreak investigations in Cattle Colonies in Karachi, Pakistan to understand the epidemiology of disease to inform evidence-based zoo-sanitary and prophylactic measures. In this paper we describe the epidemiological findings of LSD outbreak investigations carried out in Cattle Colony Karachi including clinical findings of the LSD infected animals (cattle and buffaloes), farm management practices relevant to control of LSD, and rationalizing antimicrobial use (AMU) for treatment of secondary bacterial infections.

2. MATERIALS AND METHODS:

2.1. Study area and case definition:

LSD outbreak investigations were carried out on nine cattle and buffalo farms located in Hashim Goth and Super Highway areas of Cattle Colony Karachi, Pakistan (Figure I). An animal exhibiting skin nodules (round, raised nodules of 1- 5 cm diameter) was considered as suspected case of LSD.

2.2. Outbreak investigation:

A proforma developed for LSD outbreak investigation was used to interview farmers during farm visit. The animals were examined carefully to identify the clinical signs of LSD. The animals and farm area were also inspected for presence of potential insect vectors such as mosquitoes, ticks and flies.

2.3. Sample collection:

A total of 15 samples including blood (n=7), skin scabs (n=2), and nasal swabs (n=6) were collected from nine different farms with cattle meeting the criteria of suspected LSD case definition. The samples were brought to Animal Health Research Laboratories, Animal Sciences Institute, National Agricultural Research Centre (NARC), Islamabad in a cool box containing frozen ice gel packs under cold conditions.

2.4. Sample analysis:

DNA was extracted from each of the LSD suspected sample using Invitrogen™ PureLink™ Genomic DNA Mini Kit (ThermoFisher Scientific, Waltham, USA) following manufacturers' instructions. Extracted DNA were analyzed using PCR for presence of p32 gene (Ochwo et al. 2018). The PCR reaction was performed using 25 µl reaction mixture containing 2.5 µl of 10X PCR buffer, 2.5mM $MgCl_2$, 200 nM of dNTPs mix (ThermoFisher Scientific, Waltham, USA), 600nM of each primer (Macrogen, Seoul, South Korea), 1 unit of Taq DNA Polymerase (ThermoFisher Scientific, Waltham, USA) and 3 µl template DNA. The PCR tubes containing the above mixture were transferred into a thermal

cycler (Applied Biosystems, ThermoFisher Scientific USA), and amplification was carried out using the following program: initial denaturation at 94 °C for 5 min, 35 cycles denaturation at 94°C for 1 min, annealing at 50 °C for 30 seconds, extension at 72 °C for 1 min; and a final extension at 72 °C for 5 min.

2.5. Descriptive analysis

Morbidity and mortality rates of LSD affected animals were calculated as follows:

Morbidity rate = Number of animals showing clinical signs of LSD/Total number of animals on affected farms

Mortality rate = Number of animals showed clinical signs of LSD and then died/Total number of animals on LSD affected farms.

3. RESULTS AND DISCUSSION:

Recently LSD has emerged as an important threat to livestock farmers in Pakistan and its neighboring countries (Azeem et al. 2022). Understanding local epidemiology of LSD in Pakistan is paramount to devise evidence-based strategy to prevent and control LSD. This is the first study describing the epidemiology of first-ever LSD outbreak investigations in Cattle Colony Karachi. Cattle Colony Karachi has a geographically confined and unique dairy production system. Approximately 800,000 cattle and buffaloes are being maintained in Karachi to meet the milk and meat requirements of large population of Karachi. The persistent influx of animals from other areas of Pakistan especially districts of Sindh and Punjab has been considered as an important factor responsible for spread of diseases in Cattle Colony Karachi (Afzal et al. 2012). Possibly the introduction of LSD infected animals from southern Punjab and other districts of Sindh where LSD cases were observed for the first time could be the source of transmission of LSD in Cattle Colony Karachi.

All LSD affected farms (n=9) were housing dairy cattle and buffalo and these farms were located in Hashim Goth (3 farms) and Super Highway (6 farms) areas of Cattle Colony Karachi, Pakistan. The farm size ranged from 82-450 dairy animals with average size of 176.4 animals per farm. Nodules were observed in all the LSD affected animals (Figure II). The other most common clinical signs observed were fever, lachrymation, nasal discharge and reduction in milk production. All the clinical signs observed in this study were consistent with previous reports (Namazi & Khodakaram Tafti 2021). In first week of infection 70-80% reduction in milk was observed in affected animals. Previous studies have also reported significant loss in milk production in LSD affected animals causing huge economic losses to the farmers.

Previously, the disease has been reported in buffaloes from various countries (Azeem et al. 2022). However in this study the clinical signs of LSD were only observed in cattle in affected farms while no clinical signs of LSD were observed in buffaloes maintained on those farms. A study carried out in Egypt also reported less occurrence of LSD cases in buffaloes compared to cattle (Elhaig et al. 2017). The overall morbidity and mortality rates were 15.3% and 6.8% respectively. The morbidity rate in crossbred cattle (17%) was higher compared to local breeds of cattle (11.2%). Mortality in affected animals was observed in crossbred cattle only (Table I). A study carried out in Kenya also reported that exotic breeds of cattle were 15 time more likely to get LSD compared to indigenous breeds (Kiplagat et al. 2020). This could be due to variation in vulnerability to ecto-parasites and development of resistance against local diseases in indigenous breeds of cattle due to natural selection.

Mosquitoes and flies were observed in all the affected farms. Ticks could not be found on any affected farm. Presence of biting insects on affected farm may be responsible for

transmission of disease to neighboring farms. Most of the farmers (95%) used smoke as insect repellent while few farmers (5%) used chemical based insect repellents and insecticide sprays to control mosquitoes, flies and other insects. The farmers used smoke mostly in evening and night time only therefore the animals were exposed to biting insects at the day time. These biting insects could be a possibility of spreading the disease from infected farm to other neighboring farms.

Blood, skin scabs and nasal swabs were collected from suspected LSD cases for lab diagnosis using PCR. Of 15 samples collected from suspected LSD cases two (13.3%) were found positive for LSD. All the scab samples (n=2) were found positive for LSD with PCR. In contrast all the blood and nasal swab samples were found negative (Table II). The type of sample and time of collection of sample have been reported to be critical for laboratory diagnosis of LSD. LSD virus can be detected in blood and swab samples in early stages of infection when infected animals have fever, which could explain why LSDV was not detected from blood and nasal swabs. LSD virus replicates in epithelial cells, therefore, concentration of virus is very high in skin scabs which increases the probability of detecting virus using PCR. In addition virus persists in the skin lesions/scabs for long duration therefore, LSD virus can be detected in skin scabs several months after infection (Calistri et al. 2018). However, collection of skin scabs can be painful for animal therefore, local anesthesia and appropriate surgical technique should be used.

LSD lesions put affected animals at a higher risk of secondary bacterial infections, which can aggravate the health and wellbeing of animals. Prevention and control of secondary bacterial infections may necessitate use of antimicrobials. During the outbreak investigation, it was observed that antibiotics were used for the treatment of affected animals on all affected farms with the prescription of veterinarians. However, veterinarians prescribed antibiotics on clinical basis and did not take any help from laboratory testing such as, antimicrobial susceptibility testing (AST).

A holistic approach is required when developing treatment guidelines for LSD including the use of antimicrobials for prophylaxis and treatment of secondary bacterial infections. Irrational use of antimicrobials may exacerbate the already brewing antimicrobial resistance pandemic that resulted in 1.29 human deaths attributable to antibiotic-resistant bacterial infections (Murray et al. 2022). If early action is not taken to rationalize antimicrobial use for prevention and control of secondary bacterial infection, it can lead to excessive use of critically important antimicrobials and other antimicrobials of veterinary importance. Two recent studies quantifying the use of CIAs in corporate and commercial dairy farms of Pakistan has revealed that 42.5% (Umair et al. 2020) and 58% (Mohsin et al. 2021) of all antimicrobials used were CIAs. In light of these findings, there is need for raising awareness of both dairy farmers and veterinary professionals on rational use of antimicrobials and herd health management to rationalize AMU. LSD and other transboundary animal diseases are yet another reminder to improve biosecurity, herd health management and prophylactic vaccination as a tool to reduce AMU as envisioned in the National Action Plan of AMR in Pakistan (National Institute of Health 2017).

To our knowledge this is the first study to describe the epidemiology of LSD in Pakistan. The study indicated that the disease is more prevalent in cattle compared to buffaloes. The crossbred cattle are more

susceptible to disease compared to local breeds. As it is a new disease in Pakistan, therefore, capacity building of farmers, veterinarians, laboratory staff and veterinary paraprofessionals (VPPs) is inevitable for early reporting and diagnosis. In addition to movement restriction on animals from affected farms awareness among farmers regarding biosecurity and ecto-parasite control may help in controlling the disease. The Government of Pakistan should play a role in procurement/development of LSD vaccine and development and implementation of LSD vaccination strategy to control the spread of disease in country to avoid economic losses to farmers, ensuring food security, animal welfare and rational AMU.

Table I: Morbidity and mortality rates of Lumpy skin disease (LSD) affected cattle on LSD affected farms in Cattle Colony Karachi. Clinical signs of LSD were not observed in buffaloes maintained at affected farms

Breed of cattle	Total number of cattle in affected farms	Number of animals showing LSD clinical signs	Number of cattle died	Morbidity (%)	Mortality (%)
Local breed	222	25	0	11.2%	0
Crossbred	523	89	51	17%	9.7%
Total	745	114	51	15.3%	6.8%

Table II: Blood, scab and nasal swab samples (n=15) collected from lumpy skin disease (LSD) suspected cattle maintained at different farms of Cattle Colony Karachi were analyzed using PCR.

Sample type	Number of samples collected	Number of positive samples
Blood	7	0
Scab	2	2
Nasal Swab	6	0
Total	15	2

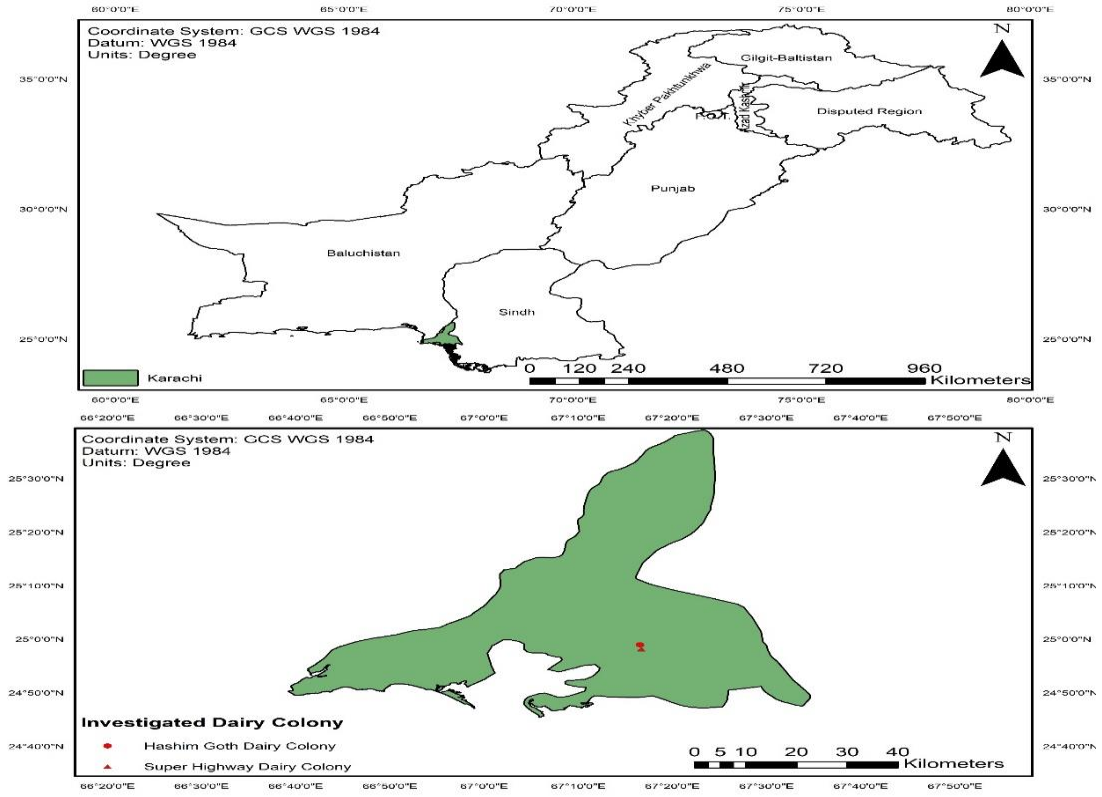

Figure I: A map showing areas in Karachi where lumpy skin disease (LSD) outbreak investigations were carried out.

Figure II: Nodules can be seen on skin of lumpy skin disease affected cattle.

4. REFERENCES:

Afzal, M., M. Hussain, E. Khan, and G. Ferrari. 2012. FMD control in dairy colonies milk production system in Pakistan. Scientific Development and Challenges to Progressive Control of FMD in South Asia. *FAO* New Dehli, India.

Azeem, S., B. Sharma, S. Shabir, H. Akbar, and E. Venter. 2022, Lumpy skin disease is expanding its geographic range: A challenge for Asian livestock management and food security. The Vet. J. 279:105785.

Calistri, P., K. DeClercq, A. De Vleeschauwer, S. Gubbins, E. Klement, and A. Gogin. 2018. Lumpy skin disease: Scientific and technical assistance on control and surveillance activities. 16:05452.

Elhaig, M. M., A. Selim, and M. Mahmoud. 2017. Lumpy skin disease in cattle: Frequency of occurrence in a dairy farm and a preliminary assessment of its possible impact on Egyptian buffaloes. Onderstepoort J. Vet. Res. 84:1-6.

Kiplagat, S. K., P. M. Kitala, J. O. Onono, P. M. Beard, and N. A. Lyons. 2020, Risk factors for outbreaks of lumpy skin disease and the economic impact in cattle farms of Nakuru County, Kenya. Front. vet. sci 7:259.

Mohsin, M., U. M. Zaheer, M. U. Abubakar, A. Rasheed, R. W. Ullah, and M. F. Tahir. 2021. Farm-level study on the trends and quantitates of antimicrobial use in commercial dairy in Pakistan In Annual Conference of Research Workers in Animal Diseases. Chicago, IL, United States 2021. December 3-7.

Murray, C. J., K. S. Ikuta, F. Sharara, L. Swetschinski, G. R. Aguilar, A. Gray, and M. Naghavi. 2022, Global burden of bacterial antimicrobial resistance in 2019. a systematic analysis. The Lancet 399-10325:629-655.

Namazi, F., and Khodakaram Tafti, A. 2021, Lumpy skin disease, an emerging transboundary viral disease: A review Vet. Med. Sci. 7:888-896.

National Institute of Health, 2017, National AMR Action Plan for Pakistan Antimicrobial Resistance National Action Plan Pakistan Ministry of National Health. In. Edited by ministry of National Health Services, Regulations and Coordination, Government of Pakistan.

Ochwo, S., Vander Waal, K, Ndekezi, C., J. Nkamwesiga, A. Munsey, S. G. Witto, and F. N. Mwiine. 2020, Molecular detection and phylogenetic analysis of lumpy skin disease virus from outbreaks in Uganda 2017-2018. BMC Vet. Res. 16:1-10.

Tuppurainen, E. S. M., and C. A. L. Oura. 2012, Review: Lumpy skin disease: an emerging threat to Europe, the Middle East and Asia. Transboundary and Emerging Diseases 59:40-48.

Tuppurainen, E. S., E. H. Venter, and J. A. W. Coetzer. 2005, The detection of lumpy skin disease virus in samples of experimentally infected cattle using different diagnostic techniques. Onderstepoort J. Vet. Res. 72:153-164.

Umair, M., R. M. Abdullah, B. Aslam, M. H. Nawaz, Q. Ali, F. Fatima, and M. Mohsin. 2020. First Case Report on Quantification of Antimicrobial Use in Corporate Dairy Farms in Pakistan. Front. vet. sci. 967.