



***Enterobacter hormaechie*: An Etiological Agent for Bovine Mastitis Identified through PCR and Sequencing**

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Authors' contributions

This work was carried out in collaboration among all authors. Author SKD collected the data. Author PK did the conceptualization of the manuscript. Author NK did the conception, designed the study and wrote the original draft. Author SKR did formal analysis and revised the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Mastitis is an important disease at the global level. With the steep rise in cases of bovine mastitis, it has become a matter of concern. It is responsible for nearly 70 per cent of the losses in milk production in India and also poses a significant public health risk. Diagnostic methods in field conditions are rather too crude, relying mainly on the apparent symptoms. Diagnostic kits available for field conditions for mastitis fail to give accurate indications about the causative agents, thus increasing antibiotic resistance. Although, molecular methods are not economical, they are reliable diagnostic tool for early and accurate assessment of etiological agents. Current study deals with the usage of PCR-based diagnostic method coupled with sequencing to find out the causative agent for mastitis in milk sample. In the present study, presence of *Enterobacter hormaechei* (*E. hormaechei*), was confirmed using PCR method. Sequencing was used to find the base sequence and phylogenetic tree of the causative agent. The 16S rRNA gene-based DNA Barcoding of the bacteria found in milk revealed that cattle are infected with *E. hormaechei* bacteria. The presence of *E. hormaechei* is also a matter of concern due to public health importance. More research should be done in future with greater number of samples for prevalence of microorganisms from the same region.

Keywords: Mastitis; primer; *Enterobacter hormaechei*; sequencing; PCR; phylogenetic tree.

1. INTRODUCTION

Mastitis refers to inflammation of the mammary gland. It includes a wide variety of causative agents (Kumari et al., 2018). The etiological agents include gram-positive, gram-negative bacteria, viruses and mycoplasma or can also be environmental (for example, *Escherichia coli*, coagulase-negative *Staphylococcus*, *Enterococcus* spp., *Streptococcus uberis*) in nature (Cheng et al., 2020). Depending upon the degree of inflammation, bovine mastitis can be classified as clinical, sub-clinical, and chronic mastitis. Clinical mastitis can further be sub-divided into per-acute, acute, and sub-acute depending on degree of the inflammation. Ironically, sub-clinical mastitis, which shows no visible abnormality in the udder or milk, but decreased milk production with an increase in the somatic cell count (Abebe et al., 2016) accounts for more financial losses in the herd than do clinical cases (Wani et al., 2022). Therefore, early diagnosis of mastitis is necessary so that the quarters of udder don't get fibrosed. Environmental mastitis is mainly associated with the gram-negative bacteria *Enterobacteriaceae*. The *Enterobacteriaceae* family has more than 50 genera and over 200 species (Schalm, Carroll, & Jain, 1971). Out of 50 genera, *Escherichia coli*, *Klebsiella*, *Enterobacter*, *Serratia* and *Proteus* are frequently isolated from dairy environment. Thus, identification and characterization of *Enterobacteriaceae* in dairy production system is necessary to understand the importance of these bacteria as causative agent for mastitis.

Diagnosis of mastitis based on clinical observations is the most common diagnostic method in field condition. Other commonly used methods are identification of the infectious agent, somatic cell count/somatic cell score and other. However, culture and polymerase chain reaction are the modern method for the diagnosis of an intramammary infection, although, both methods have their advantages and disadvantages (Adkin et al., 2018; Singh et al., 2014). Disadvantage in PCR method is, high cost while the advantage is the accurate and early diagnosis which is necessary in the wake of increasing antibiotics resistance due to its indiscriminate use and also to ensure complete treatment of animal (Khan & Khan, 2006). Nowadays, molecular techniques such as 16S rRNA sequencing is the gold standard technique for identification of bacteria. Therefore, the quality of microbial identification is the key task to control mastitis in veterinary clinical management. With this hypothesis, present study aimed to identify the causative agent of bovine mastitis using PCR technique and sequencing.

2. MATERIALS AND METHODS

The animals were screened and selected from organized dairy farm in and around Hazaribagh and Ramgarh District, Veterinary Clinical Complex (RVC) and Instructional Livestock Farm Complex, College of Veterinary Science and Animal Husbandry, Kanke, Ranchi. Selections of the animals were done on the basis of history, physical and clinical examination of udder. Indirect tests were

performed after collections of milk samples from selected animals on the basis of the above-mentioned criteria for the diagnosis of mastitis. Screening of mastitis cow was done by California Mastitis Test (CMT)), strip cup test and modified white side test. Out of the milk samples of selected animals, four random samples were taken for PCR-based analysis and sequencing. This was a part of M.V.Sc. work in the Department of Veterinary Medicine, CoVSc. & A.H on the topic "Therapeutic efficacy of *Psidium guajava* and *Punica granatum* against coliform mastitis in cattle". The Current research article is based on the molecular analysis part of the research. The purpose of PCR-based assay was to emphasize on molecular diagnostic techniques and to be able to decipher the holistic phylogenetic detail of the causative organism of the mastitis in current experimental units. The following work was done by outsourcing in Akriti Biotech Research & Development Centre, Kathal More, Ranchi, Jharkhand.

Four milk samples were received and labeled as LF, RF, RH1 and LH1 (Fig. 1). After serial dilution they were inoculated (Fig. 2a, 2b, 2c and 2d). Milk samples were serially diluted and the highest dilution was spread on EMB agar plates and incubated at 37°C overnight. No bacterial growth was observed in sample RF and RH1. Single colony obtained on the EMB agar plates of samples LF1 and LH (Fig. 3a, 3b, 3c and 3d) was inoculated in Lysogeny Broth (LB) and incubated at 37°C for overnight. Genomic DNA was isolated from the respective bacteria cultures (Fig. 4a and 4b) using Hi Media bacterial DNA isolation kit (Fig. 5). Universal 16S rRNA gene primer set was used to amplify the 16S rRNA gene and sent for

sequencing. Two PCR reactions failed for unknown reasons, so our internal primer set was used (Fig. 6).

The composition of the PCR Mix was as follows-

PCR Composition (for 50 µl reaction):

Master Mix (Promega)– 25 µl
Forward Primer– 2.5 µl
Reverse Primer– 2.5 µl
Template DNA– 2 µl
Nuclease-free water for final volume – 18 µl

PCR programme was run in the following steps-

Initial denaturation	94 °C for 5minutes
Denaturation	94°C for 40seconds
Annealing	45°C for 45seconds
Extension	72°C for 1minute
Final Extension	72°C for 10minutes

Sequencing of the PCR amplicon thus obtained was performed on the Applied Biosystems platform. Sequence analysis was done online on the NCBI Database.

3. RESULTS

Total four milk samples were received and labeled as LF, RF, RH1 and LH1. The milk samples LH1 and LF were found to be contaminated with bacteria as evidenced by the growth on EMB agar plates, however sample LH1 being highly infected as shown in Fig. 3c and 3d. Milk samples RH1 and RF had no bacterial contamination as no growth of bacteria was observed on the plate (Fig. 3a and b).



Fig. 1. Milk Samples



Fig. 2a



Fig. 2b



Fig. 2c

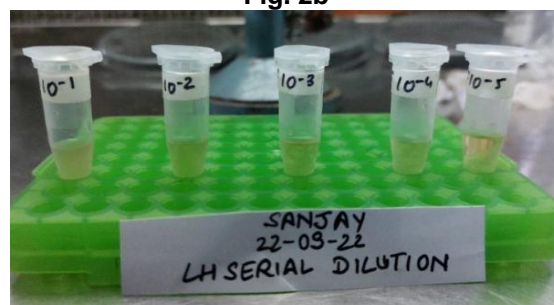


Fig. 2d

Fig. 2. Serial dilution technique

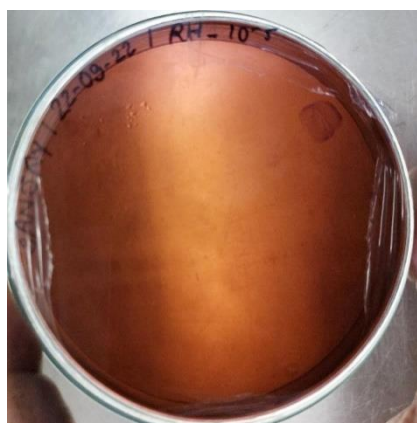


Fig. 3a. Milk sample from RH1 on culture media



Fig. 3b. Milk sample from RF on culture media

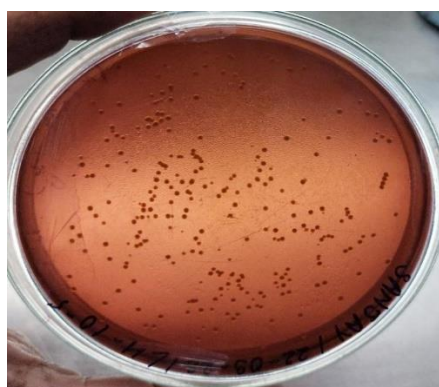


Fig. 3c. Milk sample from LH1 on culture media

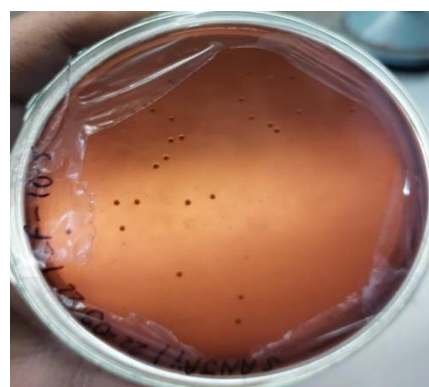


Fig. 3d. Milk sample from LF on culture media

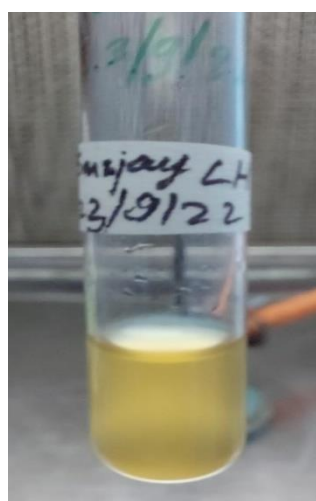


Fig. 4a. Pure culture Bacteria from LH1

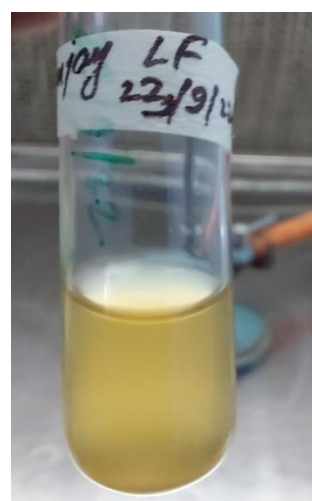


Fig. 4b. Pure culture of Bacteria from LF



Fig. 5. DNA Extracted

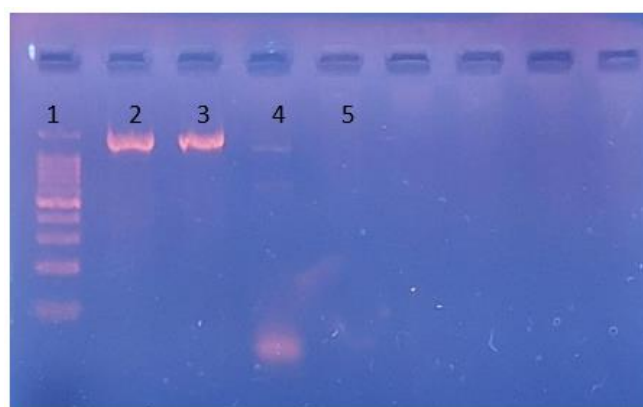


Fig. 6. PCR amplification of 16S rRNA gene. Lane1:100bp Ladder, Lane 2: LH1; Lane 3: LF, Lane 4: Positive control, Lane 5: Negative control

Genomic DNA yield: The genomic DNA yield from the respective pure bacterial cultures (Fig. 4a and 4b) was found to be 10 µg (Fig. 5) as analyzed by spectrophotometry analysis.

PCR Amplification: The PCR reaction of 16S rRNA gene gave an amplification of 1.5 kb (Fig. 6)

Sequencing: The DNA sequence obtained after sequencing, when analyzed using the NCBI Database, revealed that both the milk samples were infected with *E. hormaechei*, a common pathogen that is a recognized contaminant of raw milk and dairy products.

Sequencing Result:

Sample: LF bacteria

Identity: Enterobacter *hormaechei*

Sequence:

GCTGAGGCGGCTACACATGCAGTCGAACGGTAACAGGAAGCAGCTATGCTAGCTATTGCTAGAC
GAGTGGCGGACGGGTGAGTAATGTCTGGGAAACTCGCCTGATGGAGGGGGATAACTACTGGAA
ACGGTAGCTAATACCGCATAACGTGCGCAAGACCAAAGAGGGGGACCTTCGGGCCTCTTGCCATC
GGATGTGCCCAGATGGGATTAGCTAGTAGGTGGGGTAACGGCTCACCTAGGCGACGATCCCTA
GCTGGTCTGAGAGGATGACCAGCCACACTGGAAGTGGGACACGGTCCAGACTCCTACGGGAGG
CAGCAGTGGGGAATATTGCACAATGGGCGCAAGCCTGATGCAGCCATGCCGCGTGTATGAAGA

AGGCCTTCGGGTTGTAAAGTACTTTTCAGCGGGGAGGAAGGTGTTGAGGTTAATAACCTCAGCAAT
TGACGTTACCCGCAGAAGAAGCACCGGCTAACTCCGTGCCAGCAGCCGCGGTAATACGGAGGGT
GCAAGCGTTAATCGGAATTACTGGGCGTAAAGCGCACGCAGGCGGTCTGTCAAGTCGGATGTGA
AATCC
CCGGGCTCAACCTGGGAACTGCATTGCAAACTGGCAGGCTAGAGTCTTGTAGAGGGGGGTAGA
ATTCCAGGTGTAGCGGTGAAATGCGTAGAGATCTGGAGGAATACCGGTGGCGAAGGCGGCCCC
CTGGACAAAGACTGACGCTCAGGTGCGAAAGCGTGGGGAGCAAACAGGATTAGATACCCTGGT
AGTCCACGCCGTAAACGATGTGCACTTGGAGGTTGTGCCCTTGAGGCGTGGCTTCCGGAGCTAA
CGCGTTAAGTCGACCGCCTGGGGAGTACGGCCGCAAGGTTAAACTCAAATGAATTGACGGGG
GCCCCGACAAGCGGTGGAGCATGTGGTTTAATTGATGCAACGCGAAGAACCTTACCTACTCTT
GACATCCAGAGAACTTACCAGAGATGCTTTGGTGCCTTCGGGAACTCTGAAACAGGTGCTGCAT
GGCTGTCGTCAGCTCGTGTGTTGTGAAATGTTGGGTTAAGTCCCGCAACGAGCGCAACCCTTATCA
TTTGTGCGCGGGGTGCGCCGGGAACTCAAAGGAGACTGCCAGTGATAACTGGAGGAAGGGGG
GGATGACGTCAAGTCATCAGGCCCTTACGAGTAGGGCTCCCCCTGCTAAATGGCGCTTCAAGA
AAAAGCGACCT

Sample: LH bacteria

Identity: *Enterobacter hormaechei*

Sequence:

CCGGGGCGGGCAGGCCTAACACATGCAAGTCGAACGGTAACAGGAAGCAGCTTGCTGCTTTGC
TGACGAGTGGCGGACGGGTGAGTAATGTCTGGGAACTGCCTGATGGAGGGGGATAACTACTG
GAAACGGTAGCTAATACCGCATAACGTCGCAAGACCAAAGAGGGGGACCTTCGGGCCTCTTGCC
ATCGGATGTGCCAGATGGGATTAGCTAGTAGGTGGGGTAACGGCTCACCTAGGCGACGATCCC
TAGCTGGTCTGAGAGGATGACCAGCCACACTGGAAGTGAAGACACGGTCCAGACTCCTACGGGAG
GCAGCAGTGG
GGAATATTGCACAATGGGCGCAAGCCTGATGCAGCCATGCCGCGTGTATGAAGAAGGCCTTCG
GGTTGTAAAGTACTTTTCAGCGGGGAGGAAGGTGTTGAGGTTAATAACCTCAGCAATTGACGTTAC
CCGCAGAAGAAGCACCGGCTAACTCCGTGCCAGCAGCCGCGGTAATACGGAGGGTGCAAGCGT
TAATCGGAATTACTGGGCGTAAAGCGCACGCAGGCGGTCTGTCAAGTCGGATGTGAAATCCCCG
GGCTCAACCTGGGAACTGCATTGCAAACTGGCAGGCTAGAGTCTTGTAGAGGGGGGTAGAATTC
CAGGTGTAGCGGTGAAATGCGTAGAGATCTGGAGGAATACCGGTGGCGAAGGCGGCCCCCTGG
ACAAAGACTGACGCTCAGGTGCGAAAGCGTGGGGAGCAAACAGGATTAGATACCCTGGTAGTCC
ACGCCGTAAACGATGTGCACTTGGAGGTTGTGCCCTTGAGGCGTGGCTTCCGGAGCTAACGCG
TTAAGTCGACCGCCTGGGGAGTACGGCCGCAAGGTTAAACTCAAATGAATTGACGGGGGGCCC
GCACAAGCGGTGGAGCATGTGGTTTAATTGATGCAACGCGAAGAACCTTACCTACTCTTGACAT
CCAGAGAACTTACCAGAGATG
CTTTGGTGCCTTCGGGAACTCTGAGACAGGTGCTGCATGGCTGTCGTCAGCTCGTGTGTTGTGAAA
TGTTGGGTAAAGTCCCGCAACGAGCGCAACCCTTATCTTTGTTGCCAGCGGTTCCGGCCGGGAAC
TCAAGGGAACTGCCAGTGATAAACTGGAGGAAGGTGGGGATGACGTCAAGTCATCATGGCCCT
TACGAGTAGGGCTACCACCTGCTACATGGGCCATACAAGAAGAAGCGACTTCGCGA
AAACAAGCGGACCTCATAAATGCTTCCAATCCGAATGAAAT

Phylogenetic Analysis: Bacteria from milk sample LF clearly depicts that the bacteria found in the milk sample is closest to *E. hormaechei* in terms of evolutionary tree or distance. Whereas, bacteria from milk sample LH1 was closest to *Enterobacter cloacae*. On the contrary, the sequence alignment presented *E. hormaechei* in the first match.

distance tree. In case of bacteria from milk sample LH1 the sequence alignment analysis shows 98.73% identity with *E. hormaechei*. However, in the phylogenetic tree the query sequence shows closest relation to *Enterobacter cloacae*. But, since the query sequence matches first with *E. hormaechei*, we consider it to be the correct identity.

The 16S rRNA gene based DNA Barcoding of the bacteria found in milk revealed the cattle is infected with *E. hormaechei* bacteria. The 16S rRNA gene sequence of bacteria isolated from LF milk sample, on sequence alignment analysis matched 97.95% with *E. hormaechei* and in accordance with the phylogenetic tree of

4. DISCUSSION

Enterobacter is an oxidase-negative, gram-negative bacterium that belongs to the family *Enterobacteriaceae* formerly known as Enteric group 75 (O'hara et al., 1989). *E. hormaechei* is a common pathogen that infects both cattle and

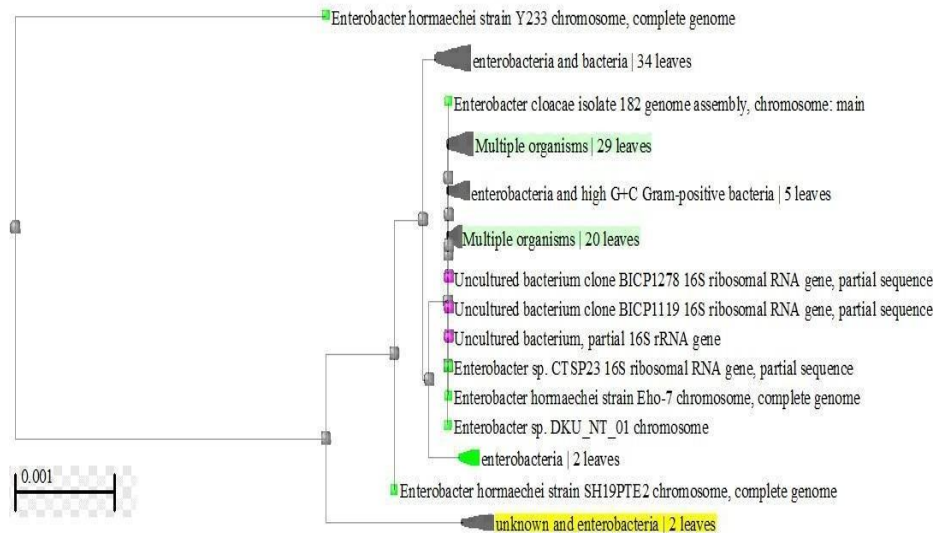


Fig. 7. Phylogenetic tree of bacteria isolated from LH1 milk sample

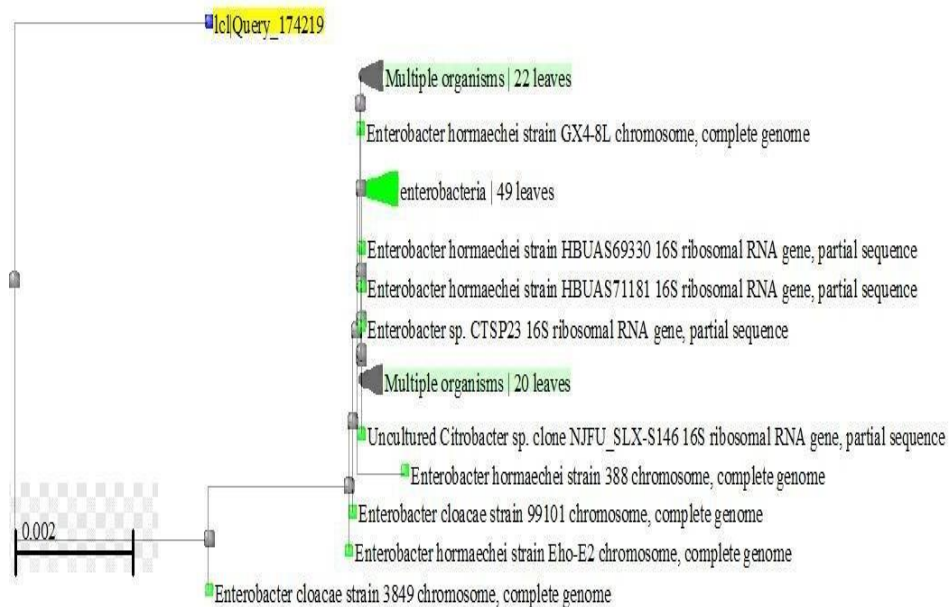


Fig. 8. Phylogenetic tree of bacteria isolated from LF milk sample

humans (O'hara et al., 1989). *Enterobacter hormaechei* infection can cause mastitis in cattle as reported earlier in a number of cases (Aslantaş et al., 2022; Mokgaotsi, 2019; Rodrigues et al., 2017; Zhong et al., 2023) as observed in present study also. Enterobacter sp. exists in broad range of environments like soil, water, food processing factories, plantations etc. They are also widely considered as potential human pathogens. Several clinical cases of *E. hormaechei* milk infection have been reported in almost every region of the world. The case of *E. hormaechei* infection was first reported in a 2-

months old severely ill calf (Wang et al., 2020). It also infects the buffalos as reported by Fagiolo & Lai, (2007). *E. hormaechei* has also been isolated from animals suffering from polymicrobial bovine pneumonia (Choudhary et al., 2019). In recent times, *E. hormaechei* has evolved as an emerging pathogenic bacteria due to its potential to cause bloodstream infection in livestock linked with morbidity, mortality and financial loss (Qian et al., 2020).

Incidence of multi drug resistant strains of *E. hormaechei* being isolated from cow with

reproductive system infection (Zaitsev et al., 2022). It belongs to the Cluster VI of *Enterobacter cloacae* complex (ECC) that is a group of common nosocomial pathogens that are capable of cause a wide variety of infections (Annavaiah et al., 2019). One of the primary reasons for Multi Drug Resistance or Antimicrobial Resistance among pathogens is due to the ability of pathogens to produce biofilm that prevents the drug to reach the bacteria cell (Liu et al., 2022). Since, *E. hormaechei* belong to the biofilm-forming group of pathogens, the reason for its drug resistance is its ability to formation of biofilm (Edris et al., 2023).

5. CONCLUSION

The finding of the present revealed that milk samples were found to contain *Enterobacter hormaechei* bacteria hitherto not very common causative agent for bovine mastitis with zoonotic importance, so it raises alarming public health concerns in the wake of milk being global source of food. Further and future research is needed with a large number of samples to ascertain and confirm *Enterobacter hormaechei* as well as to find other causative agent in mastitic milk using PCR method.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author (s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

- Abebe, R., Hatiya, H., Abera, M., Megersa, B., & Asmare, K. (2016). Bovine mastitis: prevalence, risk factors and isolation of *Staphylococcus aureus* in dairy herds at Hawassa milk shed, South Ethiopia. *BMC Vet. Res.*, 12, 1-11.
- Adkins, P. R., & Middleton, J. R. (2018). Methods for diagnosing mastitis. *Vet. Clin: Food Anim. Pract.*, 34(3): 479-491.
- Annavaiah, M. K., Gomez-Simmonds, A., & Uhlemann, A. C. (2019). Multidrug-resistant *Enterobacter cloacae* complex emerging as a global, diversifying threat. *Front. Micro.*, 10, 44.
- Aslantaş, Ö., Yılmaz, E. Ş., Büyükalay, K., & Kocabağ, H. D. (2022). Whole-genome sequencing of extended-spectrum β -lactamase (ESBL)-producing *Enterobacteriaceae* from retail raw milk. *Int. J. Dairy Tech.*, 75(1), 77-82.
- Cheng, W. N. & Han, S. G. (2020). Bovine mastitis: risk factors, therapeutic strategies, and alternative treatments - A review. *Asian-Aus. J. Anim. Sci.*, 33(11), 1699–1713.
- Choudhary, M., Choudhary, B. K., Ghosh, R. C., Bhojar, S., Chaudhari, S., & Barbuddhe, S. B. (2019). Cultivable microbiota and pulmonary lesions in polymicrobial bovine pneumonia. *Microbiol. Path.*, 134, 103577.
- Edris, S. N., Hamad, A., Awad, D. A. B., & Sabeq, I. I. (2023). Prevalence, antibiotic resistance patterns, and biofilm formation ability of *Enterobacteriales* recovered from food of animal origin in Egypt. *Vet. World*, 16(2),403–413.
- Fagiolo, A., & Lai, O. (2007). Mastitis in buffalo. *Italian J Anim Sci* 6 (sup2), 200-206.
- Khan, M. Z., & Khan, A., 2006. Basic facts of mastitis in dairy animals: A review. *Pakistan Vet. J.*, 26(4): 204.
- Kumari, T., Bhakat, C., & Choudhary, R. K. (2018). A review on subclinical mastitis in dairy cattle. *Int. J. Pure Appl. Biosci.* 6 (2): 1291-1299.
- Liu, S., Chen, L., Wang, L., Zhou, B., Ye, D., Zheng, X., ... & Ye, J. (2022). Cluster differences in antibiotic resistance, biofilm formation, mobility, and virulence of clinical *Enterobacter cloacae* complex. *Front. Microbiol.*, 13, 814831.
- Mokgaotsi, G. F. (2019). Assessment of bacteriological quality and handling of bovine raw milk collected from communal areas in Mahikeng, South Africa (Doctoral

- dissertation, North-West University (South Africa).
- O'hara, C. M., Steigerwalt, A. G., Hill, B. C., Farmer 3rd, J. J., Fanning, G. R. & Brenner, D. J. (1989). *Enterobacter hormaechei*, a new species of the family Enterobacteriaceae formerly known as enteric group 75. *J Clin. Microbiol.*, 27(9), 2046-2049.
- Qian, W., Yang, M., Wang, T., Sun, Z., Liu, M., Zhang, J., ... & Li, Y. (2020). Antibacterial mechanism of vanillic acid on physiological, morphological, and biofilm properties of carbapenem-resistant *Enterobacter hormaechei*. *J. Food Prot.* 83(4), 576-583.
- Rodrigues, N. M. B., Bronzato, G. F., Santiago, G. S., Botelho, L. A. B., Moreira, B. M., Coelho, I. D. S., & Coelho, S. D. M. D. O. (2017). The Matrix-Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometry (MALDI-TOF MS) identification versus biochemical tests: a study with enterobacteria from a dairy cattle environment. *Brazilian J. of Microbiol.*, 48, 132-138.
- Schalm, O. W., Carroll, E. J., & Jain, N. C. (1971). *Bovine Mastitis*. Lea and Febiger, Inc., Philadelphia, U.S.A. Google Scholar.
- Singh, D., Kumar, S., Singh, B., & Bardhan, D. (2014). Economic losses due to important diseases of bovines in central India. *Vet. World*, 7(8).
- Wang, Z., Duan, L., Liu, F., Hu, Y., Leng, C., Kan, Y., Yao L. A. & Shi, H. (2020). First report of *Enterobacter hormaechei* with respiratory disease in calves. *BMC Vet. Res.*, 16, 1-4.
- Wani, S.A., Ul Haq, R.I., Parray, D.R., Nazir, Q.U., Mushtaq, M., Bhatt, R.A., Parah, J.U., Chakrabarty, S., Dhama, K. and Yatoo, M.I. (2022). A brief analysis of economic losses due to Mastitis in Dairy cattle. *Indian Vet. J.*, 99 (02), 27-31
- Zaitsev, S. S., Khizhnyakova, M. A., & Feodorova, V. A. (2022). First Case Report of Detection of Multidrug-Resistant *Enterobacter hormaechei* in Clinical Sample from an Aborted Ruminant. *Microorganism*, 10(5), 036.
- Zhong, H., Zheng, N., Wang, J., & Zhao, S. (2023). Isolation and pan-genome analysis of *Enterobacter hormaechei* Z129, a ureolytic bacterium, from the rumen of dairy cow. *Front. Microbiol.*, 14, 1169973.

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