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Short communication

Isolation of Moraxella spp. strains in horses

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Abstract

Two species of *Moraxella* were isolated from swabs obtained from two symptomatic horses suffering from conjunctivitis and rhinitis. The isolates were identified via MALDI-TOF as *Moraxella osloensis* and *Moraxella equi* respectively, with a good genus identification and a probable species identification score. The first one was also confirmed by BLAST sequence analysis with a high homology (99%). Antimicrobial susceptibility testing obtained by minimum inhibitory concentration method showed that the two isolates were sensitive to all the tested molecules. The therapeutic protocols applied had an excellent clinical outcome in both cases with no relapses. Notwithstanding the difficulty in assessing the pathologic role of commensal microorganisms such as *Moraxella*, this genus should be taken into account in the differential etiologies of conjunctivitis and/or rhinitis in horses. Given also the scarce information and the difficulties in the identification of this species, it is important to report clinical cases in order to extend the knowledge in the veterinary field.

Keywords

Moraxella spp., horse, conjunctivitis, rhinitis

Introduction

Members of the genus *Moraxella* are glucose non-fermenting gram-negative rods or cocci belonging to the family Moraxellaceae (Markey et al., 2013). As of June 2026, 37 species, 29 of which have validly published names, have been reported as members of the genus *Moraxella.* Many of these have been found as commensals in several mammalian species and a few of them are potentially zoonotic, such as *Moraxella (M.) canis* (Li et al., 2023b; LPSN, 2026; Wang et al., 2022).

Regarding their health impact, *Moraxella*-associated infections, are well known in both in veterinary and human medicine, however, in the latter, they are rarely associated with severe syndromes (Azevedo et al., 2023; Ioannou et al., 2022; Janoušková et al., 2022).

The main agents of disease in animals are *M. bovis*, *M. ovis* and *M. bovoculi*, considered the most relevant aetiological agents of infectious bovine keratoconjunctivitis (IBK) (Postma et al., 2008; Pugh and Hughes, 1975; Zheng et al., 2019). Antibiotic treatment is usually effective, with low minimum inhibitory concentration (MIC) values, suggesting effective antimicrobial activity (Angelos et al., 2011, Maboni et al., 2015). Moraxella spp. have occasionally been isolated from pathological lesions in goats, chamois, elk, common eland, camels, cats, dogs, marine mammals, and macaques (Embers et al., 2011; Lavin et al., 2000; Li et al., 2023a; Shotts et al., 1990; Tagawa et al., 2017; Tejedor-Junco et al., 2010; Wang et al., 2022). A detailed description of these findings is shown in Table I.

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Affected species	Lesion/Syndrome	Isolated pathogen	Reference
Goat	Respiratory disease	*M. nasicaprae*	Li et al., 2023b
Chamois	Broncopneumonia	*M. bovis*	Lavin et al., 2000
Elk and common eland	Pulmonary abscesses	*M. lacunata*	Kim et al., 2018
Camel	Keratoconjunctivitis	*M. canis*	Tejedor-Junco et al., 2010
Cat	Pericarditis	*M. osloensis*	Tagawa et al., 2017
Dog	Corneal ulcer	*M. canis*	Wang et al., 2022
Bowhead whale	Skin lesions	*Moraxella* spp.	Shotts et al., 1990
Rhesus Macaque	Epistaxis	*M. macacae*	Embers et al., 2011
Horses	Conjunctivitis, keratoconjunctivitis and pharyngitis	*Moraxella* spp., *M. bovoculi*	Hoquet et al., 1985; Seeger et al., 2021

Table I. Isolation of Moraxella spp. from pathological lesions in mammalian species: affected species, main findings, isolated pathogens, and bibliographic references.

In equines, this microorganism has been described as commensal of the pharyngeal (Zak et al., 2018) and conjunctival microflora (Andrew et al., 2003; Cattabiani et al., 1976). A few reports have also identified *Moraxella* strains in horses suffering from conjunctivitis, keratoconjunctivitis and pharyngitis (Hoquet et al., 1985; Hughes and Pugh, 1970; Huntington, 1987; Liu et al., 2014; Seeger et al., 2021). The description of *Moraxella* spp. as conjunctival commensals in horses (Cattabiani et al., 1976) has also been reported in Italy too but, to date, no evidence of pathological cases has been reported. In the present communication, the identification of two species of *Moraxella* from two symptomatic horses suffering from conjunctivitis and rhinitis is described.

Materials and methods

Cases presentation

Case 1: A conjunctival swab was obtained from a 5-year-old mare suffering from an acute, bilateral conjunctivitis, in the absence of other clinical signs. This mare was housed in a paddock with two other horses that did not show any ocular and/or other clinical sign.

Case 2: A nasal swab was collected from a 12-year-old mare affected by a severe catarrhal unilateral rhinitis.

The horses were housed on different farms, located in the hilly area of the province of Udine, in the Friuli Venezia Giulia region. Both horses showed watery/mucous conjunctivitis during the summer of 2023 and swab samples were collected by two different veterinary clinicians in June (Case 2) and July (Case 1). Besides the ocular and respiratory symptoms described, both veterinary clinicians reported no other concurrent symptoms or syndromes. Both cases were reported to have an acute onset and no treatments were reported at the time of sampling.

Laboratory diagnostics

Both swabs were submitted to the diagnostic laboratory of the Istituto Zooprofilattico Sperimentale delle Venezie (Udine). They were plated onto blood agar (BA) and eosin methylene blue agar plates, inoculated into tryptic soy broth and aerobically incubated. In addition, another BA plate was incubated under microaerophilic conditions (Campygen, Thermo Scientific) in both cases. All plates were incubated at 37 ± 2 °C.

Following the isolation of the bacterial colonies, species identification was performed by mass spectrometry using MALDI-TOF (MALDI Biotyper® Sirius System; Bruker Daltonics GmbH, Bremen, Germany) and the associated libraries (MALDI Biotyper library version BDAL 2023 and software version 3.4).

In addition, bacterial DNA was extracted using the Magmax Core Nucleic Acid Purification Kit and a 16S rRNA gene assay was performed using primers described by Davidovich et al. (2022). PCR was performed in 50 µL reaction volumes, containing 1× buffer, 0.2 mM dNTPs, 2 mM MgCl2, 0.5 U of Taq polymerase (Platinum Taq DNA Polymerase, Invitrogen, Carlsbad, CA, USA), 0.2 µM of each primer, 2 µl of extracted DNA (100 ng/µl) and Milli-Q water to the final volume. Amplification was carried out using a ProFlex PCR System thermal cycler (Applied

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Biosystems, Life Technologies, Foster City, CA, USA) and the following protocol: 94 °C for 1 min, 35 cycles of 94 °C for 1 min, 60 °C for 1 min, 72 °C for 1 min and a final extension at 72 °C for 10 min. The expected amplicon lenght was 1,060 bp. The amplified products were analysed by capillary electrophoresis using a QIAxcel Advanced instrument (QIAGEN GmbH, Hilden, Germany). The amplicon was then sequenced in-house using a Sanger sequencing protocol, sequencing both strands with the BDT Cycle Sequencing Kit 3.1 and determining the nucleotide sequence with a SeqStudio Genetic Analyser (Applied Biosystems, Thermo Fisher Scientific, Waltham, MA, USA). The resulting chromatograms were evaluated manually and assembled into a consensus using BioEdit Sequence Alignment Editor v.7.2.6.1 software. The consensus was finally aligned against sequences available in GenBank to identify a possible match.

Moreover, MICs were determined using the broth microdilution method with a commercial 96-well microtitre plate (Thermo Fisher Diagnostic, Waltham, MA, USA). The antimicrobial susceptibility testing was performed in accordance with the guidelines of the Clinical and Laboratory Standards Institute (CLSI).

Results

From both swab samples, after 24 h of incubation at 37 ± 2 °C on blood agar plates, flat, rounded, whitish colonies were observed. No other relevant bacterial pathogens were isolated under either aerobic or microaerophilic conditions.

MALDI-TOF analysis identified *M. osloensis* from the conjunctival swab and provided a presumptive identification of *M. equi* from the nasal swab. Specifically, the identification score was 2.13 for *M. osloensis* (conjunctival swab), and 2.09 for *M. equi* (nasal swab). In both cases, the scores obtained indicated good genus-level identification and probable species-level identification.

In addition, *M. osloensis* was further analysed using molecular methods. The sequence obtained using the16S rRNA gene assay confirmed the MALDI-TOF identification by BLAST sequence analysis (GenBank accession number: PZ658180) showing high sequence identity (99%).

The antimicrobials tested using the MIC plates were ampicillin, amoxicillin/clavulanic acid, doxycycline, enrofloxacin, trimethoprim/sulfamethoxazole, and tetracycline. The interpretive criteria, the MIC values obtained and the results are summarised in Table II.

Both horses had an excellent clinical outcome following antibiotic treatment. Specifically, in the first case (conjunctivitis), a combination of tobramycin and dexamethasone was administered, while in the case of rhinitis, the clinician prescribed treatment with thrimethoprim/sulfamethoxazole.

In both cases, no relapse was observed.

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	M. osloensis		*M. equi*		
	VALUE	S/I/R	VALUE	S/I/R	Reference
Amikacin	≤1	S	≤1	S	CLSI M100 ED35:2025 Acinetobacter sp
Amoxicillin/ Clavulanic Acid	≤0.06/0.03	S	≤0.06/0.03	S	EUCAST Clinical Breakpoint Tables v. 15.0, valid from 2025-01-01 M catarrhalis. https://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Breakpoint_tables/v_15.0_Breakpoint_Tables.pdf
Ampicillin	≤0.06		≤0.06		
Ceffalexin	≤0.5		≤0.5		
Cefazolin	2		≤0.5		
Cefovecin	0.12		≤0.06		
Cefpodoxime	≤0.5		≤0.5		
Doxycycline	0.12	S	0.12	S	EUCAST Clinical Breakpoint Tables v. 15.0, valid from 2025-01-01 M catarrhalis
Enrofloxacin	≤0.06		≤0.06		
Gentamicin	≤0.5	S	≤0.5	S	CLSI M100 ED35:2025 Acinetobacter sp
Kanamycin	≤8		≤8		
Pradofloxacin	≤0.12		≤0.12		
Tetracycline	≤2	S	≤2	S	EUCAST Clinical Breakpoint Tables v. 15.0, valid from 2025-01-01 M catarrhalis
Trimethoprim/ Sulfamethoxazole	≤0.5	S	≤0.5	S	EUCAST Clinical Breakpoint Tables v. 15.0, valid from 2025-01-01M catarrhalis

Table II. MIC values and interpretative criteria for *Moraxella* spp. isolates. Legenda. MIC: minimum inhibitory concentration; CLSI: Clinical and Laboratory Standards Institute; EUCAST: European Committee on Antimicrobial Susceptibility Testing.

Discussion

The genus *Moraxella* spp. includes several species that may colonise different mucosal surfaces as commensals, as well as other species that have pathogenic potential (Andrew et al., 2003; Azevedo et al., 2023). This distinction applies to both veterinary and human medicine.

In detail, in the latter, *Moraxella* spp. are considered a commensal organisms living on skin and mucosal surfaces of the human respiratory tract. *Moraxella* associated infections have rarely been implicated in human disease and, indeed, are mainly reported in immunocompromised patients. Moreover, isolation of *Moraxella* spp. have also been isolated from hospital environments, suggesting a putative role in hospital-acquired infections (Koleri et al., 2022).

In the veterinary context, most of the research and clinical data derive from cases of IBK in domestic ruminants, mainly cattle. Apart from *M. bovis*, *M. ovis* and *M. bovoculi*, reported as causal agents of pinkeye, occasional reports of *Moraxella* spp. as causative agents of disease in other mammalian species have been published, as described in Table I.

In the present report, the first isolation of *Moraxella* spp. from two symptomatic horses in Italy is described. While in other countries *Moraxella* spp. have previously been described in cases of keratoconjunctivitis and pharyngitis in equids, to the best of the author's knowledge, they have not yet been reported in Italy (Liu et al., 2014; Seeger et al., 2021).

In Case 1, *M. osloensis* was isolated from a horse suffering from bilateral conjunctivitis. The bacterial isolate was identified by MALDI-TOF (score 2.13), and this identification was further confirmed by 16S sequencing. Data on the identification of *M. osloensis* in horses are currently lacking in Italy, although *M. osloensis* has previously been isolated from healthy canine newborns (Rota et al., 2021) and from ectoparasites, namely *Lipoptena*, feeding on wild cervids (Andreani et al., 2023). Moreover, both *Moraxella* spp. and *M. bovoculi* were isolated from clinically affected

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horses with conjunctivitis in China (Liu et al., 2014) and South America (Seeger et al., 2021), respectively.

In Case 2, the presumptive identification of *M. equi* (score 2.09) in a case of rhinitis is noteworthy. Accordingly, *Moraxella* spp. are not considered primary respiratory pathogens in veterinary medicine (Hughes and Pugh, 1970; Loy et al., 2021). Recently, *M. nasibovis* was isolated from the nasal cavity of a cow with respiratory disease (Li et al., 2023b), while in horses, *Moraxella* spp. were isolated in cases of grade II and grade III pharyngitis (Hoquet et al., 1985). The presuntive identification of *M. equi* in a case of equine catarrhal rhinitis, in the absence of apparent ocular involvement, is worth mentioning. Considering that members of this genus have been identified as commensals of the nasal mucosa of horses (Cattabiani et al., 1976), other authors have suggested that an imbalance in the flora may underlie pathological conditions at this anatomical site (Hoquet et al., 1985). From a diagnostic perspective, both the microbiological culture and the species identification of *Moraxella* spp. are challenging. Growth is favoured by the addition of serum or blood to the medium and the routine identification methods may not be able to identify species belonging to the genus *Moraxella*. (Pimenov et al., 2024; Wilkes et al., 2024). In fact, as observed in the present case report, *the presumptive identification of M. equi was obtained by* MALDI-TOF (score 2.09), which also yielded a similar score (2.08) for *M. bovis* too. This was probably due to the similarity between the bacterial spectral profiles and the limited amount of data deposited in the reference libraries for the species considered (MALDI Biotyper library version BDAL 2023). Since it was not possible to further characterise this bacterial isolate, *M. equi* was considered the most likely identification, given the animal species involved.

With regards to AMR, the breakpoints for *Moraxella* species and the data on susceptibility patterns among clinical isolates in veterinary medicine are scarce. In human medicine, *M. osloensis* strains isolated from blood cultures showed that all strains were susceptible to amikacin, trimethoprim-sulfamethoxazole, ciprofloxacin, imipenem, and some cephalosporins. They were also β-lactamase negative, which is one of the key features of *Moraxella* species (Han and Tarrand, 2004). Moreover, there is not a standard antibiotic regimen for the treatment of bacteraemia caused by *M. osloensis* (Richards et al., 2022).

For both cases, considering the limited clinical information, we cannot demonstrate a definitive aetiological role for the *Moraxella* isolates. However, no other relevant bacterial agents were isolated and Moraxella colonies were obtained in pure culture. After the antibiotic treatment, both animals rapidly recovered. Although not conclusive from a clinical point of view, an *ex- juvantibus* diagnosis of an infection due to *Moraxella* spp. may be suggested.

Furthermore, in the cases presented herein, both horses were in good general condition. The affected horses did not show any relevant concurrent infections or diseases, and no known predisposing environmental factors and conditions were identified. Even though it is not possible to completely rule out underlying immunosuppressive conditions that could potentially have favoured these two pathological conditions, given the good prognosis following antibiotic treatment, these seem unlikely.

With regard to the response to antibiotic treatment, the *M. osloensis* strain was susceptible to all the antimicrobials tested. A previous study evaluated the antimicrobial susceptibility for several *Moraxella* strains collected from horses (Bzdil et al., 2018). As in the present case report, *Moraxella* strains were susceptible to tetracycline (100%) and enrofloxacin (100%), with lower susceptibility to amoxicillin/clavulanic acid (83.3%). However, unlike in our cases, the strains showed lower or absent susceptibility to other antimicrobials, i.e., gentamicin (0%), erythromycin (66.7%), clindamycin (33.3%), bacitracin (50%), and cephalothin (83.3%). This reduced susceptibility pattern is of great concern for treatment, with regard to the potential risk of antimicrobial resistance developing. Although resistant strains are rarely isolated, *M. bovis* isolated recovered during IBK episodes showed higher MICs for oxytetracycline compared with previous studies, suggesting that this may be related to the widespread use of this drug in cases of IBK (Maboni et al., 2015).

Conclusion

In conclusion, horses may display conjunctivitis and/or rhinitis associated with the isolation of *Moraxella* spp. These findings do not necessarily imply a direct/primary pathogenic role of *Moraxella* spp., since the distinction between pathogenicity and colonisation may be difficult when dealing with commensal microorganisms. In this context, it is beyond the scope of this short communication to draw conclusions about the potential impact of *Moraxella* spp. as pathogens in equids. Instead, the main aim is to draw the attention of both practitioners and microbiologists to the potential pathogenic role of *Moraxella* spp. in horses, as these bacteria are mainly considered commensals. Given the difficulties in bacterial identification and the limited data in the literature, an underestimation of the pathological role of *Moraxella* spp. in this species cannot be excluded.

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Ethical approval

Ethical approval was not required.

Conflict of interest

Authors declare no conflict of interest.

Author Contributions

Conceptualisation: LG, GDZ, MC; Methodology: SD, NG, LC, MU; Formal analysis: SD, NG, LC, MU; Investigation: FC, PL; Writing original draft preparation: LG, GDZ, MC; Writing, review and editing: LG, GDZ, MC; Supervision: MC; Project administration: MC.

All authors have read and agreed to the published version of the manuscript.

Data availability

Not applicable, as no new data were created or analysed in this study.

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