

Does colonization with MRSA, ESBL – producing *Enterobacteriaceae*, and/or *Acinetobacter baumannii* – increase the risk for postoperative surgical site infection?

Heidrun Gehlen¹ | Katja-Sophia Klein¹ | Roswitha Merle²  |
Antina Lübke-Becker^{3,4} | Sabita D. Stoeckle¹ 

¹Equine Clinic: Surgery and Radiology, Freie Universität Berlin, Berlin, Germany

²Institute for Veterinary Epidemiology and Biostatistics, Freie Universität Berlin, Berlin, Germany

³Institute of Microbiology and Epizootics, Freie Universität Berlin, Berlin, Germany

⁴Veterinary Centre for Resistance Research (TZR), Freie Universität Berlin, Berlin, Germany

Correspondence

Sabita Stöckle, Equine Clinic: Surgery and Radiology, Freie Universität Berlin, Oertzenweg 19b, 14163 Berlin, Germany. Email: SDStoeckle@gmx.de

Abstract

Objective: Evaluation of the role of indicator pathogens in equine surgical site infection (SSI) and other infection-promoting factors.

Study design: Cross-sectional study.

Animals: Horses presenting with an open injury or surgical colic during 1.5 years.

Methods: A nasal swab and a faecal sample were collected from every patient upon admission. Furthermore, a wound swab was collected from wounds of injured horses. Details on the wounds and procedures were documented. Laparotomy incisions and injuries were monitored for signs suggesting infection.

Results: In total, 156 horses presented because of a surgical colic ($n = 48$) or open injuries ($n = 108$). Thirteen surgical colic patients and three injured horses did not survive beyond 24 h, and four injured horses were discharged from the clinic at the day of admission. SSIs occurred in 31 (30.7%) injured horses and 11 (31.4%) horses after laparotomy. Regarding injuries, general anaesthesia increased the risk of developing a WI compared to sedation. Indicator pathogens were cultured from 29/42 SSI. In total, 10/11 infected laparotomy incisions and 19/31 injuries with SSI tested positive for multidrug-resistant pathogens (MDRPs). Indicator pathogens were not detected at admission in any of the horses that developed incisional SSIs after laparotomy but were detected in two of the injured horses that developed SSIs.

Conclusion: MDRPs were identified in almost 70% of the SSI. Less than 5% of the affected animals were colonized with the same pathogen before admission, indicating that colonization with MDR pathogens is only one of the crucial factors for the development of SSI.

Clinical significance: Colonization with MDRP seems not to predispose horses to MDR SSIs.

KEYWORDS

antimicrobial resistance, Equine, Infection, Surgery

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1 | INTRODUCTION

Surgical site infections (SSIs) lead to impaired wound healing and are a common problem in hospitalized horses especially after median laparotomy or traumatic injuries (Mair & Smith, 2005). They are often caused by multidrug-resistant pathogens (MDRPs), including methicillin-resistant *Staphylococcus aureus* (MRSA), extended-spectrum beta-lactamase-producing Enterobacterales (ESBL-E) and *Acinetobacter baumannii* (Bergstrom et al., 2012; Boerlin et al., 2001; Cuny et al., 2006, 2008; Dallap Schaer et al., 2010; Damborg et al., 2012; Dargatz & Traub-Dargatz, 2004; Ekiri et al., 2009; Henninger et al., 2007; Seguin et al., 1999; Van den Eede et al., 2012; Walther, Lübke-Becker et al., 2008; Walther, Wieler, et al., 2008; Weese & Lefebvre, 2007), which cannot only be found in the environment but may also be a part of the body's own microbiota (Weese, 2008). Further examinations of *S. aureus* isolated from equine SSI collected in multiple clinics in Germany revealed that 41% (57/138) actually were MRSA (Vincze et al., 2014). Furthermore, horses are increasingly colonized with ESBL-E (Maddox et al., 2012; Walther et al., 2018a). Various factors influence the risk of developing SSI for an individual animal of which several have been identified previously for small animals (Brown et al., 1997; Eugster et al., 2004). These factors included the clinical status (e.g. weight, immune status and age) and surgical aspects (duration of the operation, surgical skills of the surgeon, clipping of the surgical field, the number of people in the operating room, the condition of the tissue operated on, the degree of contamination and subsequent wound management) as well as the period of hospitalization (Brown et al., 1997; Eugster et al., 2004; Hamilton et al., 1977; Nicholson et al., 2002; Suzuki et al., 1984). In this study, we investigated the hypothesis that colonization with MDRP may predispose horses to SSIs caused by MDRP.

2 | MATERIALS AND METHODS

2.1 | Ethical approval

According to the statement of the responsible regulations for research with animal subjects, which was made before the start of the study, taking swabs from anterior nasal cavity of equine patients at hospital admission in the context of a hygiene evaluation did not require approval.

2.2 | Study population

All horses that presented to the clinic because of a surgical colic or an open injury over a time period of 1.5 years (April 2014–October 2015) were included as study participants. The signalment was recorded for every patient. All horses were examined and treated according to the current standard of the clinic. The horses were not followed after hospital discharge; however, owners were asked to contact the clinic in the case of developing complications.

2.3 | Data extracted

The localization, age, extent of the wound (length, width, depth) and degree of contamination in injured horses and whether the injury was surgically managed standing or under general anaesthesia were extracted.

The length of the incision was recorded in colic patients undergoing laparotomy.

Pretreatment with antibiotics and/or non-steroidal anti-inflammatory drugs was documented for all horses as was the treatment with these drugs during the hospital stay. The length of hospitalization was recorded for each horse. Parts of a day the horse spent at the clinic counted as a whole day, that is the day of admission and day of discharge were also included in the evaluation as a whole day.

2.4 | Surgery

Details of the type of surgery, diagnosis, duration of surgery, surgeon, anaesthesia and intraoperative medication were documented for each patient.

As perioperative antimicrobial prophylaxis, for colic surgery, amoxicillin (10 mg/kg BID) and gentamicin (6.6 mg/kg SID) were prescribed. If no adverse effects were observed, the treatment was continued for a total of 5 days.

In injured horses, the use of a perioperative antibiotic prophylaxis was prescribed at the discretion of the responsible veterinary surgeon depending on extent and contamination of the wound.

2.5 | Surgical site infection

Both the incisions of horses undergoing laparotomy and injuries were clinically examined every 2–3 days for signs suggesting infection (calor, dolor, rubor, tumor, functio laesa, exudation) as part of the active surveillance of postoperative SSIs. Furthermore, the body temperature was monitored at least twice daily. In the case of exudation and/or dehiscence, a sample was taken from the exudate and/or the dehiscent part of the sutures. Regarding SSI, the day of occurrence and the clinical parameters were documented. This included the depth of the infection (skin and subcutaneous tissue, fascia layer and muscle tissue, or organs/body cavities that were opened during the operation), whether the infection occurred during hospitalization, at the home stable or at readmission.

2.6 | Microbiological examinations

A nasal swab and a faecal sample were collected from each horse for microbiological examination within 120 min of arrival at the clinical facilities. A sample was also obtained from the centre of the wound

in injured horses. Samples from infected wounds were collected either from the centre of the wound in case of injuries or directly from the infected part of the laparotomy incision. A sterile swab for nostrils and wounds (Mastaswab, Copan Italia SpA, Via F. Perotti, 10-25125 Brescia Italy) and a conical polypropylene container (120-ml capacity) with a red screw cap and sample sterile spatula (VWR, International bvba, Geldenaaksebaan 464-B-3001 Leuven) for the faecal sample were used for sample collection. Samples were stored between 5 and 7°C and transported to the Institute of Microbiology and Epizootics as fast as possible, that is on the same day or on the following day, at the latest. All samples were screened for ESBL-E, MRSA and *A. baumannii* as indicator pathogens.

All faecal samples, nostril swabs and wound swabs were initially cultured on Columbia agar with 5% sheep blood (bioMérieux, Germany), Brilliance ESBL Agar (Thermo Scientific Germany), CHROMagar Acinetobacter (Mast Diagnostica, Germany) and chromID MRSA (bioMérieux, Germany) agar plates overnight.

Species confirmation was achieved by matrix-assisted laser desorption/ionization-time of flight (MALDI-TOF) mass spectrometry (Bruker, Germany).

Colonies showing characteristic growth signatures of ESBL-E on chromogenic screening plates were further investigated. In case of *Escherichia coli* or *Klebsiella pneumoniae* growing on the plates, the isolates were additionally subjected to the ESBL confirmatory test according to the Clinical and Laboratory Standards Institute (CLSI) recommendations (Clinical and Laboratory Standards Institute (CLSI, 2012).

For all *S. aureus* isolates growing on the screening plates, detection of the *mecA* (Merlino et al., 2002) or *mecC* (Cuny et al., 2011) gene was performed by PCR as described before.

Antimicrobial susceptibility testing using the VITEK2 system (BioMérieux, Germany) was performed according to the standards given by the CLSI VET01-A4 and M100-S21 (CLSI, 2011, 2013).

Bacterial isolates were classified as multiresistant following published definitions (Schwarz et al., 2010).

2.7 | Statistical evaluation

The statistical evaluation was performed with IBM SPSS Version 25 (SPSS Inc., Chicago, Illinois, USA). The chi-square test or Fisher's exact test (if at least 25% of the cells with expected counts <5) was used to compare frequencies and crosstabs across categories. Odds ratios (OR) were calculated, including 95% confidence intervals (95% CI) whenever possible. Distribution of continuous data was assessed by visual inspection (histogram, Q-Q plot, boxplot) and descriptions. Median and interquartile ranges (IQR) are presented for continuous but nonparametric data, and mean and standard deviation for normally distributed data. Quantitative differences between two groups were investigated using the Mann-Whitney *U* test assuming nonparametric distribution or *t* test assuming normal distribution. *p* Values less than 0.05 were considered significant.

3 | RESULTS

3.1 | Study population

In total, 48 patients were admitted because of a surgical colic and 108 because of an injury (total $n = 156$) during the 1.5 years of the study. In total, 13 of the 48 colic patients that required surgical correction of a gastrointestinal problem were euthanized intraoperatively or shortly after surgery (within 24 h). Additionally, three injured horses were euthanized due to a grave prognosis. Four further injured horses were discharged from the clinic after initial treatment, as no surgical procedure was necessary. The following data include the horses presented for open injuries ($n = 101$) and surgical colic ($n = 35$) that survived surgery beyond 24 h/that remained in the clinic for more than 24 h.

The median age of the horses was 8 years (IQR 4–15), and there was neither significant difference between groups (surgical colic: median 8.5 years, IQR 6–15 years; injury: median 8.0 years, IQR 4–15 years; $p = 0.380$, Mann-Whitney *U* test) nor between animals that developed an SSI (7 years, IQR 4–12.5 years) and those whose wounds healed unproblematically (9 years, IQR 4–15 years; $p = 0.253$, Mann-Whitney *U* test).

Horses which were presented for surgical colic were significantly heavier than horses that were presented with an injury (surgical colic: 548.5 kg, IQR 491–587 kg; injury: 471 kg, IQR 371–570 kg; $p = 0.010$, Mann-Whitney *U* test), but there was no significant difference regarding wound healing (SSI: 454.5 kg, IQR 359.5–566.5 kg; unproblematic healing: 524 kg, IQR 420–578 kg; $p = 0.123$, Mann-Whitney *U* test). Most of the horses presented were warmbloods ($n = 74$), and other breeds presented were ponies ($n = 27$), Western breeds (25), Thoroughbreds ($n = 9$) and Standardbreds ($n = 8$). There was no significant difference in breed between the groups ($p = 0.563$, Fisher's exact test), but there was a significant difference in breed ($p = 0.046$, Fisher's exact test) regarding SSI, as Standardbreds and ponies developed SSI more often than other breeds.

Only a few stallions ($n = 14$) compared to mares ($n = 54$) and geldings ($n = 68$) presented due to colic or open injuries. No significant differences regarding group (injured horses vs. colic patients) or infection were detected ($p = 0.587$, $p = 0.727$, respectively, chi-square tests).

The treatment before admission was significantly different between the groups (injured horses vs. colic patients, $p = 0.012$, chi-square test; OR 2.67, 95% CI 1.22–5.82) but not between horses with and without SSI ($p = 0.835$). Before admission, horses presenting for open injuries were treated significantly more often with antibiotics than those presenting for a surgical colic ($p = 0.002$, chi-square test; OR 12.5, 95% CI 1.6–95.4), whereas surgical colic patients were treated significantly more often with non-steroidal anti-inflammatory drugs (NSAIDs) than injured horses ($p < 0.001$, chi-square test; OR 6.2, 95% CI 2.7–14.4). There was no significant difference in the pretreatment with antibiotics ($p = 0.592$) and NSAIDs ($p = 0.822$, chi-square tests each) between horses with and without SSI (Table 1). Furthermore, there was no association between antibiotic treatment pre admission and

TABLE 1 Factors possibly examined for their influence on SSI

| | | WI | Uncomplicated | <i>p</i> |
|--|-------------------------|---------------------|---------------|----------|
| Age in years | | 7 (4–12.5) | 9 (4–15) | 0.253 |
| Weight in kg | | 454.5 (359.5–566.5) | 524 (420–578) | 0.123 |
| Pretreatment | Antibiotics | 10 | 20 | 0.592 |
| | NSAID | 12 | 27 | 0.822 |
| Hospitalization time (days) | Colic | 20 (10–27) | 11 (8.5–14) | 0.001 |
| | Open injury | 9.5 (7–29) | 8 (4–14) | |
| Type of anaesthesia (open injuries only) | General anaesthesia | 19 | 24 | 0.011 |
| | Heavy sedation/standing | 12 | 46 | |
| Duration of surgery (min) | | 90 (60–120) | 110 (60–120) | 0.647 |
| Colonization with indicator pathogens | Nasal vestibule | 4 | 4 | 0.48 |
| | Gastrointestinal tract | 7 | 3 | 0.86 |
| | Central injury | 3 | 7 | 0.312 |

isolation of indicator pathogens upon admission ($p = 0.549$, chi-square test).

Horses were hospitalized for a median of 11 days (IQR 6–18 days, minimum: 1 day, maximum: 61 days). Horses with injuries were hospitalized for a shorter time (median 9.5 days) than those with surgical colic (median 12 days), but this effect was not statistically significant ($p = 0.058$, Mann–Whitney *U* test).

A total of 11/35 (31.4%) surviving colic patients developed SSI after surgery. Due to their SSI, these horses remained hospitalized for 20 days (median; IQR 10–27). This difference was statistically significant (Mann–Whitney *U* test, $p = 0.016$) compared to those surgical colic patients without SSI (median 11, IQR 8.5–14 days).

Injured patients without SSI were hospitalized for 8 days (median, IQR 4–14 days). Injured horses with SSI (31/101; 30.7%) stayed for 9.5 days (median, IQR 4–18). Patients with SSIs remained hospitalized for a significantly longer time (median 18 days, IQR 7–29 days) than patients with uncomplicated wound healing ($p = 0.001$, Mann–Whitney *U* test).

3.2 | Duration of surgery

The duration of surgery was significantly different between the groups ($p = 0.001$, Mann–Whitney *U* test, surgical colic: median 120 [IQR 90–120] min, injuries: median 85 [IQR 60–120] min), but there was no significant difference in the time of surgery in patients with and without SSI ($p = 0.581$, infected: median 90 [IQR 60–120] min, unproblematic: median 110 [IQR 60–120] min).

3.3 | Injuries

Most injuries were located at the distal limb (77/101, 76.2%), followed by truncal (20/101, 19.8%) injuries and head/neck lacerations (11/101, 10.9%). Injured horses were most commonly presented between 1 and 4 h (40/101, 10.0%) or more than 24 h after the insult (32/101, 31.7%).

In total, 25 horses were admitted between 4 and 12 h and 10 animals were admitted 12–24 h after the insult. The time of violation in one horse was unknown to the owner. Of these 101 horses, 43 were operated on under general anaesthesia, and 58 were managed standing. Wounds that were managed under general anaesthesia had a three times higher risk of developing an SSI when compared to wounds that were managed standing ($p = 0.011$, chi-square test; OR: 3.03; 95% CI of 1.26–7.30). The localization and age of the injuries were not significantly different between horses with and without SSI, although older wounds showed a tendency to have SSI more often (chi-square test, $p = 0.091$ for age of injuries, $p = 0.383$ for wound localization).

3.4 | Laparotomy

Forty-eight horses required colic surgery for correction of the intra-abdominal problem. The type of colic and the necessity of enterotomy and/or resection were not considered for statistical analysis. Thirteen of these horses were subjected to euthanasia during or shortly after the surgery (within 24 h). Incisional length was available for 30/35 of the horses (24.7 ± 4.0 cm). It was impossible to measure the incisional length in five horses. There was no significant difference in the incisional length required for laparotomy between horses with and without an SSI ($p > 0.999$, *t* test). A simple wound covering was placed after the laparotomy which in most horses had to be replaced after recovery.

3.5 | Surgical site infection

A total of 42 SSIs occurred. SSIs occurred in 11/35 (31.4%) colic patients after median laparotomy and in 31/101 (30.7%) injured patients.

Most SSIs involved skin and subcutaneous tissue (34/42, 81.0%), and 8/42 (19.0%) involved the fascia and the muscles. The diagnosis of

TABLE 2 Overview of the indicator pathogens cultured from patients upon admission and from surgical site infections

| Sampling site | | MRSA | ESBL-E | MDR <i>Acinetobacter baumannii</i> |
|-------------------------|------------------|------------|------------|------------------------------------|
| Nasal vestibule | All (n = 142) | 4 (2.8%) | 3 (2.1%) | 4 (2.8%) |
| | Colic (n = 34) | 2 (5.8%) | 2 (5.8%) | 2 (5.8%) |
| | Injury (n = 108) | 2 (1.9%) | 1 (0.9%) | 2 (1.9%) |
| Feces | All (n = 108) | 2 (1.9%) | 16 (14.8%) | 0 |
| | Colic (n = 10) | 1 (10.0%) | 5 (50.0%) | 0 |
| | Injury (n = 98) | 1 (1.0%) | 11 (11.2%) | 0 |
| Wound | Injury (n = 108) | 4 (3.7%) | 2 (1.9%) | 1 (0.9%) |
| Surgical site infection | All (n = 42) | 14 (33.3%) | 21 (50.0%) | 2 (4.8%) |
| | Colic (n = 11) | 1 (9.1%) | 9 (81.1%) | 2 (18.2%) |
| | Injury (n = 31) | 13 (41.9%) | 2 (6.5%) | 0 |

Abbreviations: ESBL-E, ESBL-producing *Enterobacteriaceae*; MDR, multidrug-resistant; MRSA, methicillin-resistant *Staphylococcus aureus*.

SSI was most commonly made during hospitalization (37/42, 88.1%). The diagnosis of SSI in two horses occurred after hospital discharge and in three horses at readmission to the hospital. Purulent exudation occurred in 36/42 SSI (85.7%), 31/42 SSI (73.8%) were tender and swollen, whereas increased heat was only present in 3/42 (3.1%) horses. The SSI was accompanied by fever in 10/42 (23.8%) animals. Twenty horses (47.6%) were still under antibiotic cover upon the development of SSI, and in 22 (52.4%) animals, the antimicrobials had already been discontinued.

3.6 | Microbiological examinations

Upon admission, indicator pathogens were cultured from the nasal vestibule and wound and faecal samples (Table 2). Unfortunately, not all samples were available for all patients. A total of 142/143 (99.3%) samples from the nasal vestibule, 129/143 (90.2%) faecal samples and 108/108 (100%) wound samples were available. Table 2 displays the indicator pathogens that were cultured from the horses upon admission.

Indicator pathogens were cultured from 29/42 (69%) SSIs. Regarding the surgical colic patients, 10/11 (90.9%) SSIs tested positive for indicator pathogens. More than 80% of the SSIs after colic surgery tested positive for ESBL-E. None of the colic patients tested positive upon admission for the MDRP that had been cultured from the SSI.

Injuries with SSI tested positive for indicator pathogens in 19/31 (61.3%) cases. Of these horses, two patients had already tested positive for this indicator pathogen upon admission.

In both cases, MRSA was detected both at admission screening and at follow-up after the occurrence of postoperative wound infection. The isolates obtained at admission screening and from the postoperative wound infection were found to be 100% identical based on pulsed-field gel electrophoresis and *S. aureus* protein A (*spa*) typing.

There was no significant difference between the groups regarding the colonization of the nasal vestibule and the gastrointestinal tract with MRSA, ESBL-E and *A. baumannii* upon admission. Further-

more, there was no significant difference in colonization with indicator pathogens upon admission between horses that developed an SSI and those that did not (nasal colonization $p = 0.48$; gastrointestinal colonization $p = 0.86$; colonization of the wound (only injured horses) $p = 0.312$; all chi-square test).

The ESBL-E ($p = 0.041$, chi-square test) and *A. baumannii* ($p = 0.042$, chi-square test) were cultured significantly more often from the SSIs of colic patients than injured horses.

4 | DISCUSSION

The colonization of individuals with MDRP has become a serious challenge in both veterinary and human medicine (Beard, 2010; Bergstrom et al., 2012; Vo et al., 2007). Both our and other studies identified carriers of MRSA, ESBL-E and *A. baumannii* upon admission to the hospital (Baptiste et al., 2005; Weese et al., 2006). In human medicine, in addition to advanced age, extremes in nutritional status, diabetes, coexisting infection at a remote body site, altered immune response and preoperative hospitalization, the colonization of the nasal vestibule with *S. aureus* is defined as a risk factor for an SSI (Bratzler & Houck, 2005; Wacha et al., 2010). Gram-positive and -negative bacteria residing on the horses' skin are often identified in SSIs in horses as well (Ahern & Richardson, 2012). However, in this study, the colonization with indicator pathogens upon admission did not increase the risk of developing an SSI with any organism. Other studies reported on an increased risk of postoperative SSI after pre-treatment with antimicrobials or incorrect antibiotic use (Girou et al., 1998; Safdar & Maki, 2002; Willemsen et al., 2009) as this might increase the risk of colonization with resistant bacteria (Adam & Southwood, 2006; Anderson et al., 2009; Schoster et al., 2020; Walther et al., 2011; Willemsen et al., 2009), which was not the case in our patient material as both injured horses and colic patients received peri- and postoperative antibiotics.

Nevertheless previous research revealed veterinary clinics as 'hotspots' for local MDR pathogen accumulation and transmission

(Apostolakos et al., 2017; Kauter et al., 2021; Walther et al., 2018a; Wright et al., 2005). As our results are in line with recent reports, the development of appropriate hygiene strategies that take into account the special challenges of equine medicine (e.g. large stalls, non-slip floors, bedding, dust and manure) and that differ significantly from conditions in human and small animal medicine is of utmost importance (Gehlen, 2020; Walther et al., 2014, 2017).

MacDonald et al. identified 4.6 times higher risk for the development of wound infections in orthopaedic surgery if the patient was operated under antibiotic cover. They assumed that the surgeons identified patients at risk for the development of wound infections and prescribed antibiotics for these procedures. In total, they reported on an SSI rate of 8.1% for clean ($n = 433$) and 52.6% for clean-contaminated surgeries ($n = 19$) (MacDonald et al., 1994). In our patient material, 31/101 (30.7%) injured horses developed SSI. Commonly, injuries that require general anaesthesia are more complicated to operate than those that do not. Wounds that required surgery under general anaesthesia had a three times higher risk for infection than injuries that were managed in the standing horse.

In this study, 11/35 (31.4%) horses developed SSI after a median laparotomy which is similar to the SSI rate reported in the literature. Previous studies on wound infection after colic surgery research reported SSI in 15% to 50% of the horses (Anderson et al., 2015; Colbath et al., 2014; Darnaud et al., 2016; Durward-Akhurst et al., 2013; Stöckle et al., 2021; Tnibar et al., 2013; Torfs et al., 2010).

MDRPs were commonly cultured from infected laparotomy incisions (90.9%). Additionally, infected laparotomy incisions were infected significantly more often with ESBL-E and *A. baumannii* than the infected wounds of injured horses. The protection of the incisional wound from residing MDRP from the mucosal surfaces and/or the environment seems crucial to prevent SSI, especially because earlier studies identified 10.7% of admitted horses being positive for ESBL-E and 3.5% for MRSA (Kauter et al., 2021; Walther et al., 2018a; Walther et al., 2018b). A simple wound covering was placed after the laparotomy and renewed after recovery. This only minimally reduces the entry of environmental germs, especially when the horses lie down in their stable. In light of the multidrug-resistant bacteria associated with SSI in this study, improvement of the management of postoperative bandaging seems advisable, particularly as another study reports on a significant reduction in SSI after recovery utilizing a sterile stent bandage sewn over the incision which was subsequently replaced by an abdominal bandage (Tnibar et al., 2013).

Various studies show that an increased period of hospitalization also increases the risk of nosocomial infection (hospitalism) (Brown et al., 1997; Eugster et al., 2004; Kauter et al., 2019; Nicholson et al., 2002). However, this has to be rated as a codependency as the treatment of an SSI requires additional hospitalization time (Brown et al., 1997; Eugster et al., 2004; Nicholson et al., 2002; Green & Wenzel, 1977), which is also reflected by our study results.

An increased time of surgery is mentioned in other studies as a risk factor for the development of SSI. In human surgeries, it is assumed that the estimated risk of postoperative SSI in a 90-min surgery is

twice as high as that for a 60-min surgery (Brown, 2011). In dogs and cats, a study reports on a 1.01× increased risk of postoperative SSI per minute of surgery, which means that the risk of postoperative SSI doubles every 70 min. In equine surgery, an increasing risk of the development of SSI with the increasing time of surgery was described for both laparotomy and management of injuries (Adam & Southwood, 2006; Wilson et al., 1995; Macdonald et al., 1994). However, another study on canines as well as this study did not identify the duration of operations as a risk factor for SSI (Whittem et al., 1999). Both in these and in our study, this might be attributed to an insufficient number of cases with surgery time available.

The length of the wound/incision is commonly mentioned as an additional risk factor for SSI (Darnaud et al., 2016; Bartmann et al., 2003). This study, however, identified no statistical difference in the SSI development regarding incisional length which may be attributed to the various kind of wounds (surgical vs. nonsurgical, clean vs. dirty) that were included in the study.

The necessity of a surgical intervention under general anaesthesia was identified as a risk factor for the development of SSI in injured horses in this study. This may be attributed to the fact that deeper and more complicated wounds are commonly managed under general anaesthesia compared to more superficial and uncomplicated wounds.

A clear limitation of the study is that we only sampled a limited number of horses compared to all horses admitted to the clinic during the study period. The non-sampled horses may have been a source of contamination with MDR bacteria in the environment. Furthermore, information on the environmental contamination with MDR pathogens is unavailable. It therefore seems possible that horses may have acquired infections with MDR bacteria from the hospital environment. This again highlights the necessity of a constant environmental surveillance of clinical environments and the aspiration of high hygienic standards for both the staff and the environment.

5 | CONCLUSION

Although we identified MDRP in almost 70% of the SSIs, endogenous infections caused by colonizing MDR bacteria (MRSA; ESBL-E, *A. baumannii*) played a minor role in the equine patients of this study. Nevertheless, our results indicate the urgent need for improved hygiene strategies especially concerning the wound management to prevent exogenous nosocomial infections.

AUTHOR CONTRIBUTION

Study design: Heidrun Gehlen and A. Lübke-Becker. *Data acquisition:* Katja-Sophia Klein and A. Lübke-Becker. *Statistical analysis:* Roswitha Merle, Katja-Sophia Klein and Sabita D. Stoeckle. *Writing – first draft:* Heidrun Gehlen, Katja-Sophia Klein and Sabita D. Stoeckle. *Writing – review and editing:* Heidrun Gehlen, Katja-Sophia Klein, Roswitha Merle, A. Lübke-Becker and Sabita D. Stoeckle. *Final approval of the version to be published:* Heidrun Gehlen, Katja-Sophia Klein, Roswitha Merle, A. Lübke-Becker and Sabita D. Stoeckle.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

ETHICS STATEMENT

According to the statement of the responsible regulations for research with animal subjects, which was made before the start of the study, taking swabs from anterior nasal cavity of equine patients at hospital admission in the context of a hygiene evaluation did not require approval.

ORCID

Roswitha Merle  <https://orcid.org/0000-0002-8688-2926>

Sabita D. Stoeckle  <https://orcid.org/0000-0002-9763-5816>

REFERENCES

- Adam, E. N., & Southwood, L. L. (2006). Surgical and traumatic wound infections, cellulitis, and myositis in horses. *Veterinary Clinics of North America-Equine Practice*, 22, 335–361.
- Ahern, B. J., & Richardson, D. W. (2012). Surgical site infection and the use of antimicrobials. In J., Auer, J., Stick(Eds.), *Equine surgery*, vol. 4, Elsevier Saunders.
- Anderson, M. E., Lefebvre, S. L., Rankin, S. C., Aceto, H., Morley, P. S., Caron, J. P., Welsh, R. D., Holbrook, T. C., Moore, B., Taylor, D. R., & Weese, J. S. (2009). Retrospective multicentre study of methicillin-resistant *Staphylococcus aureus* infections in 115 horses. *Equine Veterinary Journal*, 41, 401–405.
- Anderson, S. L., Devick, I., Bracamonte, J. L., Hendrick, A., Barber, S. M., Carmalt, J. L., & Wilson, D. G. (2015). Occurrence of incisional complications after closure of equine celiotomies with USP 7 polydioxanone. *Veterinary Surgery*, 44, 521–526.
- Apostolakos, I., Franz, E., van Hoek, A. H., Florijn, A., Veenman, C., Sloet-van Oldruitenborgh-Oosterbaan, M. M., Dierikx, C., & van Duijkeren, E. (2017). Occurrence and molecular characteristics of ESBL/AmpC-producing *Escherichia coli* in faecal samples from horses in an equine clinic. *Journal of Antimicrobial Chemotherapy*, 72, 1915–1921.
- Baptiste, K. E., Williams, K., Williams, N. J., Wattret, A., Clegg, P. D., Dawson, S., Corkill, J. E., O'Neill, T., & Hart, C. A. (2005). Methicillin-resistant staphylococci in companion animals. *Emerging Infectious Diseases*, 11, 1942–1944.
- Bartmann, C. P., Bubeck, K., Georgiadis, S., & Deegen, E. (2003). Reduction of complications of the wound healing following ventral median celiotomy in horses. *Pferdeheilkunde*, 19, 351–358.
- Bergstrom, K., Aspan, A., Landen, A., Johnston, C., & Grönlund-Andersson, U. (2012). The first nosocomial outbreak of methicillin-resistant *Staphylococcus aureus* in horses in Sweden. *Acta Veterinaria Scandinavica*, 54, 11.
- Beard, L. A. (2010). Multiple drug resistant bacteria in equine medicine: An emerging problem. *Equine Veterinary Education*, 22, 287–289.
- Boerlin, P., Eugster, S., Gaschen, F., Straub, R., & Schawalder, P. (2001). Transmission of opportunistic pathogens in a veterinary teaching hospital. *Veterinary Microbiology*, 82, 347–359.
- Brown, D. C. (2011). Wound infections and antimicrobial use. In S.A, Tobias KMJ. (Ed.), *Veterinary surgery* (pp. 135–139). Small Animals Elsevier, Vol Elsevier.
- Brown, D. C., Conzemius, M. G., Shofer, F., & Swann, H. (1997). Epidemiologic evaluation of postoperative wound infections in dogs and cats. *Journal of the American Veterinary Medical Association*, 210, 1302–1306.
- Bratzler, D. W., & Houck, P. M. (2005). Antimicrobial prophylaxis for surgery: An advisory statement from the National Surgical Infection Prevention Project. *The American Journal of Surgery*, 189, 395–404.
- CLSI. (2012). *Performance standards for antimicrobial susceptibility testing. Supplement M100-S22*. Clinical and Laboratory Standards Institute.
- CLSI. (2011). *Clinical and Laboratory Standards Institute: Performance standards for antimicrobial susceptibility testing: Twenty-first Informational Supplement M100-S21*. Clinical and Laboratory Standards Institute.
- CLSI. (2013). *Vet01-S2 Performance Standards for Antimicrobial Disk and Dilution Susceptibility for Bacteria isolated from Animals. Second International Supplement*. Clinical and Laboratory Standards Institute.
- Colbath, A. C., Patipa, L., Berghaus, R. D., & Parks, A. H. (2014). The influence of suture pattern on the incidence of incisional drainage following exploratory laparotomy. *Equine Veterinary Journal*, 46, 156–160.
- Cuny, C., Kuemmerle, J., Stanek, C., Willey, B., Strommenger, B., & Witte, W. (2006). Emergence of MRSA infections in horses in a veterinary hospital: Strain characterisation and comparison with MRSA from humans. *Eurosurveillance*, 11, 44–47.
- Cuny, C., Strommenger, B., Witte, W., & Stanek, C. (2008). Clusters of infections in horses with MRSA ST1, ST254, and ST398 in a veterinary hospital. *Microbial Drug Resistance*, 14, 307–310.
- Cuny, C., Layer, F., Strommenger, B., & Witte, W. (2011). Rare occurrence of methicillin-resistant *Staphylococcus aureus* CC130 with a novel mecA homologue in humans in Germany. *PLoS One*, 6, e24360.
- Dallap Schaer, B. L., Aceto, H., & Rankin, S. C. (2010). Outbreak of Salmonellosis Caused by *Salmonella enterica* Serovar Newport MDR-AmpC in a Large Animal Veterinary Teaching Hospital. *Journal of Veterinary Internal Medicine*, 24, 1138–1146.
- Damborg, P., Marskar, P., Baptiste, K. E., & Guardabassi, L. (2012). Faecal shedding of CTX-M-producing *Escherichia coli* in horses receiving broad-spectrum antimicrobial prophylaxis after hospital admission. *Veterinary Microbiology*, 154, 298–304.
- Dargatz, D. A., & Traub-Dargatz, J. L. (2004). Multidrug-resistant *Salmonella* and nosocomial infections. *Veterinary Clinics of North America-Equine Practice*, 20, 587–600.
- Darnaud, S. J., Southwood, L. L., Aceto, H. W., Stefanovski, D., Tomassone, L., & Zarucco, L. (2016). Are horse age and incision length associated with surgical site infection following equine colic surgery? *Veterinary Journal*, 217, 3–7.
- Darnaud, S. J. M., Southwood, L. L., Aceto, H. W., Stefanovski, D., Tomassone, L., & Zarucco, L. (2016). Are horse age and incision length associated with surgical site infection following equine colic surgery? *The Veterinary Journal*, 217, 3–7.
- Durward-Akhurst, S. A., Mair, T. S., Boston, R., & Dunkel, B. (2013). Comparison of two antimicrobial regimens on the prevalence of incisional infections after colic surgery. *The Veterinary Record*, 172, 287.
- Ekiri, A. B., MacKay, R. J., Gaskin, J. M., Freeman, D. E., House, A. M., Giguère, S., Troedsson, M. R., Schuman, C. D., von Chamier, M. M., Henry, K. M., & Hernandez, J. A. (2009). Epidemiologic analysis of nosocomial *Salmonella* infections in hospitalized horses. *Journal of the American Veterinary Medical Association*, 234, 108–119.
- Eugster, S., Schawalder, P., Gaschen, F., & Boerlin, P. (2004). A prospective study of postoperative surgical site infections in dogs and cats. *Veterinary Surgery*, 33, 542–550.
- Gehlen, H. (2020). Umgang mit Antibiotikaresistenzen in Praxis und Klinik. *Leipziger Blaue Hefte*, 174.

- Girou, E., Stephan, F., Novara, A., Safar, M., & Fagon, J. Y. (1998). Risk factors and outcome of nosocomial infections: Results of a matched case-control study of ICU patients. *American Journal of Respiratory and Critical Care Medicine*, *157*, 1151–1158.
- Green, J. W., & Wenzel, R. P. (1977). Postoperative Wound Infection: A Controlled Study of the Increased Cost of Hospitalization. *Annual Surgery*, *185*, 264–268.
- Hamilton, H. W., Hamilton, K. R., & Lone, F. J. (1977). Preoperative hair removal. *Canadian Journal of Surgery*, *20*, 269–271, 274–275.
- Henninger, R. W., Reed, S. M., Saville, W. J., Allen, G. P., Hass, G. F., Kohn, C. W., & Sofaly, C. (2007). Outbreak of neurologic disease caused by equine herpesvirus-1 at a university equestrian center. *Journal of Veterinary Internal Medicine*, *21*, 157–165.
- Kauter, A., Epping, L., Ghazisaedi, F., Lübke-Becker, A., Wolf, S. A., Kannapin, D., Stoeckle, S. D., Semmler, T., Günther, S., Gehlen, H., & Walther, B. (2021). Frequency, local dynamics, and genomic characteristics of ESBL-producing *Escherichia coli* isolated from specimens of hospitalized horses. *Frontiers in Microbiology*, *12*, 843.
- Kauter, A., Epping, L., Semmler, T., Antao, E. M., Kannapin, D., Stoeckle, S. D., Gehlen, H., Lübke-Becker, A., Günther, S., Wieler, L. H., & Walther, B. (2019). The gut microbiome of horses: Current research on equine enteral microbiota and future perspectives. *Animal Microbiome*, *1*, 14.
- MacDonald, D. G., Morley, P. S., Bailey, J. V., Barber, S. M., & Fretz, P. B. (1994). An examination of the occurrence of surgical wound infection following equine orthopaedic surgery (1981-1990). *Equine Veterinary Journal*, *26*, 323–326.
- Macdonald, D. G., Morley, P. S., Bailey, J. V., Barber, S. M., & Fretz, P. B. (1994). An examination of the occurrence of surgical-wound infection following equine orthopedic-surgery (1981-1990). *Equine Veterinary Journal*, *26*, 323–326.
- Maddox, T. W., Clegg, P. D., Diggle, P. J., Wedley, A. L., Dawson, S., Pinchbeck, G. L., & Williams, N. J. (2012). Cross-sectional study of antimicrobial-resistant bacteria in horses. Part 1: Prevalence of antimicrobial-resistant *Escherichia coli* and methicillin-resistant *Staphylococcus aureus*. *Equine Veterinary Journal*, *44*, 289–296.
- Merlino, J., Watson, J., Rose, B., Beard-Pegler, M., Gottlieb, T., Bradbury, R., & Harbour, C. (2002). Detection and expression of methicillin/oxacillin resistance in multidrug-resistant and non-multidrug-resistant *Staphylococcus aureus* in Central Sydney, Australia. *Journal of Antimicrobial Chemotherapy*, *49*, 793–801.
- Mair, T., & Smith, L. (2005). Survival and complication rates in 300 horses undergoing surgical treatment of colic. Part 2: short-term complications. *Equine Veterinary Journal*, *37*, 303–309.
- Nicholson, M., Beal, M., Shofer, F., & Brown, D. C. (2002). Epidemiologic evaluation of postoperative wound infection in clean-contaminated wounds: A retrospective study of 239 dogs and cats. *Veterinary Surgery*, *31*, 577–581.
- Safdar, N., & Maki, D. G. (2002). The commonality of risk factors for nosocomial colonization and infection with antimicrobial-resistant *Staphylococcus aureus*, *Enterococcus*, gram-negative *Bacilli*, *Clostridium difficile*, and *Candida*. *Annals of Internal Medicine*, *136*, 834–844.
- Seguin, J. C., Walker, R. D., Caron, J. P., Kloos, W. E., George, C. G., Hollis, R. J., Jones, R. N., & Pfaller, M. A. (1999). Methicillin-resistant *Staphylococcus aureus* outbreak in a veterinary teaching hospital: Potential human-to-animal transmission. *Journal of Clinical Microbiology*, *37*, 1459–1463.
- Schwarz, S., Silley, P., Simjee, S., Woodford, N., van Duijkeren, E., Johnson, A. P., & Gaastra, W. (2010). Assessing the antimicrobial susceptibility of bacteria obtained from animals. *Veterinary Microbiology*, *141*, 1–4.
- Schoster, A., van Spijk, J. N., Damborg, P., Moodley, A., Kirchgassner, C., Hartnack, S., & Schmitt, S. (2020). The effect of different antimicrobial treatment regimens on the faecal shedding of ESBL-producing *Escherichia coli* in horses. *Veterinary Microbiology*, *243*, 108617.
- Suzuki, A., Namba, Y., Matsuura, M., & Horisawa, A. (1984). Airborne contamination in an operating suite – Report of a 5-Year Survey. *Journal of Hygiene*, *93*, 567–573.
- Stöckle, S. D., Kannapin, D. A., Kauter, A. M. L., Lübke-Becker, A., Walther, B., Merle, R., & Gehlen, H. (2021). A pilot randomised clinical trial comparing a short-term perioperative prophylaxis regimen to a long-term standard protocol in equine colic surgery. *Antibiotics*, *10*, 587.
- Torfs, S., Levet, T., Delesalle, C., Dewulf, J., Vlamincq, L., Pille, F., Lefere, L., & Martens, A. (2010). Risk factors for incisional complications after exploratory celiotomy in horses: Do skin staples increase the risk? *Veterinary Surgery*, *39*, 616–620.
- Tnibar, A., Grubbe Lin, K., Thurøe Nielsen, K., Christophersen, M. T., Lindegaard, C., Martinussen, T., & Ekstrøm, C. T. (2013). Effect of a stent bandage on the likelihood of incisional infection following exploratory coeliotomy for colic in horses: A comparative retrospective study. *Equine Veterinary Journal*, *45*, 564–569.
- Tnibar, A., Grubbe Lin, K., Thuroe Nielsen, K., Christophersen, M. T., Lindegaard, C., Martinussen, T., & Ekstrøm, C. T. (2013). Effect of a stent bandage on the likelihood of incisional infection following exploratory coeliotomy for colic in horses: a comparative retrospective study. *Equine Veterinary Journal*, *45*, 564–569.
- Van den Eede, A., Hermans, K., Van den Abeele, A., Floré, K., Dewulf, J., Vanderhaeghen, W., Crombé, F., Butaye, P., Gasthuys, F., Haesebrouck, F., & Martens, A. (2012). Methicillin-resistant *Staphylococcus aureus* (MRSA) on the skin of long-term hospitalised horses. *The Veterinary Journal*, *193*, 408–411.
- Vincze, S., Stamm, I., Kopp, P. A., Hermes, J., Adlhoch, C., Semmler, T., Wieler, L. H., Lübke-Becker, A., & Walther, B. (2014). Alarming proportions of methicillin-resistant *Staphylococcus aureus* (MRSA) in wound samples from companion animals, Germany 2010-2012. *PLoS One*, *9*, e85656.
- Vo, A. T., van Duijkeren, E., Fluit, A. C., & Gaastra, W. (2007). Characteristics of extended-spectrum cephalosporin-resistant *Escherichia coli* and *Klebsiella pneumoniae* isolates from horses. *Veterinary Microbiology*, *124*, 248–255.
- Walther, B., Lübke-Becker, A., & Wieler, L. H. (2008). Wundinfektionen durch methicillinresistente *Staphylococcus* spp. (MRS) bei Kleintieren und Pferden: klinische Bedeutung, Therapie und Prophylaxe. *Tierärztliche Praxis, Ausgabe K: Kleintiere/Heimtiere*, *36*, 5–10.
- Walther, B., Wieler, L. H., Friedrich, A. W., Hanssen, A. M., Kohn, B., Brunnerberg, L., & Lübke-Becker, A. (2008). Methicillin-resistant *Staphylococcus aureus* (MRSA) isolated from small and exotic animals at a university hospital during routine microbiological examinations. *Veterinary Microbiology*, *127*, 171–178.
- Walther, B., Klein, K. S., Barton, A. K., Semmler, T., Huber, C., Wolf, S. A., Tedin, K., Merle, R., Mitrach, F., Guenther, S., Lübke-Becker, A., & Gehlen, H. (2018). Extended-spectrum beta-lactamase (ESBL)-producing *Escherichia coli* and *Acinetobacter baumannii* among horses entering a veterinary teaching hospital: The contemporary “Trojan Horse”. *PLoS One*, *13*, e0191873.
- Wacha, H., Hoyme, U., Isenmann, R., Kujath, P., Lebert, C., Naber, K., & Salzberger, B. (2010). Perioperative Antibiotika-Prophylaxe: Empfehlungen einer Expertenkommission der Paul Ehrlich-Gesellschaft für Chemotherapie e.V. *Journal of Chemotherapie*, *19*, 70–84.
- Walther, B., Lübke-Becker, A., Brunnerberg, L., Kohn, B., & Wieler, L. H. (2011). Einsatz von Antibiotika in der Kleintiermedizin: Quo vadis? *Der Praktische Tierarzt*, *92*, 1047–1049.
- Walther, B., Janßen, T., Gehlen, H., Vincze, S., Borchers, K., Wieler, L., Barton, A., & Lübke-Becker, A. (2014). Infektionsprävention und Hygiene management in Pferdekliniken. *Berliner und Münchener Tierärztliche Wochenschrift*, *127*, 448–497.
- Walther, B., Tedin, K., & Lübke-Becker, A. (2017). Multidrug-resistant opportunistic pathogens challenging veterinary infection control. *Veterinary Microbiology*, *200*, 71–78.
- Walther, B., Klein, K. S., Barton, A. K., Semmler, T., Huber, C., Merle, R., Tedin, K., Mitrach, F., Lübke-Becker, A., & Gehlen, H. (2018). Equine methicillin-resistant sequence type 398 *Staphylococcus aureus* (MRSA) harbor mobile genetic elements promoting host adaptation. *Frontiers in Microbiology*, *9*, 2516.

- Weese, J. S., & Lefebvre, S. L. (2007). Risk factors for methicillin-resistant *Staphylococcus aureus* colonization in horses admitted to a veterinary teaching hospital. *Canadian Veterinary Journal*, *48*, 921–926.
- Weese, J. S. (2008). A review of multidrug resistant surgical site infections. *Veterinary and Comparative Orthopaedics and Traumatology*, *21*, 1–7.
- Weese, J. S., Rousseau, J., Willey, B. M., Archambault, M., McGeer, A., & Low, D. E. (2006). Methicillin-resistant *Staphylococcus aureus* in horses at a veterinary teaching hospital: Frequency, characterization, and association with clinical disease. *Journal of Veterinary Internal Medicine*, *20*, 182–186.
- Whittem, T. L., Johnson, A. L., Smith, C. W., Schaeffer, D. J., Coolman, B. R., Averill, S. M., Cooper CW, & Merkin, G. R. (1999). Effect of perioperative prophylactic antimicrobial treatment in dogs undergoing elective orthopedic surgery. *Journal of the American Veterinary Medical Association*, *215*, 212–216.
- Wilson, D. A., Baker, G. J., & Boero, M. J. (1995). Complications of celiotomy incisions in horses. *Veterinary Surgery*, *24*, 506–514.
- Willemsen, I., Bogaers-Hofman, D., Winters, M., & Kluytmans, J. (2009). Correlation between antibiotic use and resistance in a hospital: Temporary and ward-specific observations. *Infection*, *37*, 432–437.
- Wright, J. G., Tengelsen, L. A., Smith, K. E., Bender, J. B., Frank, R. K., Grendon, J. H., Rice, D. H., Thiessen, A. M. B., Gilbertson, C. J., Sivapalasingam, S., Barrett, T. J., Besser, T. E., Hancock, D. D., & Angulo, F. J. (2005). Multidrug-resistant *Salmonella* Typhimurium in four animal facilities. *Emerging Infectious Diseases*, *11*, 1235.

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