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Prevalence of MRSA in canine and feline clinical samples from one-third of veterinary practices in Germany from 2019–2021

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Background: MRSA is a major contributor to AMR-related deaths. The WHO's global action plan emphasizes a One Health approach, acknowledging the connection between humans and their companion animals. It is agreed on that comprehensive AMR surveillance is needed.

Objectives: This study provides a large-scale overview of MRSA occurrence in cats and dogs in Germany, serving as a foundation for continuous surveillance.

Methods: The study analysed all results of canine and feline bacterial diagnostic samples from a large laboratory, encompassing samples received from veterinary practices between January 2019 and December 2021. MRSA prevalence between host species, sample types and geographical distribution were compared. Additionally, data were contrasted with human MRSA surveillance data from Germany.

Results: Samples originated from 3491 German veterinary practices, representing 33.1% of practices and clinics nationally. Bacterial examination results from 175 171 samples were analysed, identifying *S. aureus* in 5526 of these samples (3.2% isolation rate). *S. aureus* in clinical samples was more prevalent in cats (5.6%) than dogs (2.0%). Methicillin resistance was found in 17.8% of *S. aureus* samples and was higher in dogs (20.4%, 95%CI 18.9–22.0) than cats (15.6%, 95%CI 14.3–17.0). The highest MRSA prevalence was found in canine wound samples (32%), compared to skin/soft tissue, respiratory tract and other (<23% respectively).

Conclusion: The study reveals a 17.8% MRSA prevalence, which is higher than the human outpatient MRSA prevalence (5.4%). Restriction and regulation of veterinary antibiotic use should be validated with AMR surveillance. Our study shows that this is feasible in companion animals with significant coverage.

Introduction

Antimicrobial resistance (AMR) is a silent pandemic, directly responsible for at least 1.27 million people's deaths each year. In 2019, methicillin-resistant *Staphylococcus aureus* (MRSA) was the most common pathogen-drug combination for deaths attributable to AMR globally.[1] While MRSA rates are trending down in the last decade in Europe, MRSA still remains the second largest burden of disease in the EU/EEA between 2016 and 2020 in terms of attributable deaths and disability-adjusted life years.[2] MRSA causes infections ranging from mild to life-threatening, such as skin and soft tissue infections, bloodstream infections, pneumonia and endocarditis.[3] It is rated as a high priority pathogen on WHO's antibiotic-resistant 'priority pathogens' list.[4]

AMR is a natural phenomenon, but antimicrobial substance usage promotes its selection.[5] With humans and animals sharing

a close microbial environment, tackling AMR requires an interdisciplinary strategy. The One Health approach, which acknowledges the intricate connection between people, animals and their shared environment, emerges as an essential framework for addressing this issue.[3,5,6] The WHO's global action plan for AMR leverages this One Health perspective to enhance understanding of AMR through comprehensive surveillance and research. These strategies aim to maintain treatment efficacy and enable efficient infection control.[7]

AMR surveillance data from human diagnostics are available in Europe through the European Antimicrobial Resistance Surveillance Network (EARS-Net). Germany is part of EARS-Net through the national 'Antibiotika-Resistenz-Surveillance' (ARS) system, which collects approximately 1.5 million antimicrobial susceptibility testing (AST) results yearly, which represents data of 37.6% of hospitals and 31.7% of medical practices in Germany (2021).[8] GERM-Vet, the veterinary AMR surveillance counterpart in Germany, which uses an active surveillance approach, meanwhile only reported on 3736 bacterial isolates for 2020, 80% of which come from livestock animals.[9] An active surveillance approach collects isolates from participating laboratories and performs an in-depth testing, for GERM-Vet including genome sequencing. By contrast, we present a passive surveillance approach, which collects all available clinical data and provides more representative results. Pets are an important risk factor for MRSA transmission due to close contact and shared living spaces with their owners and transmission between dogs and their owners has been shown.[10] This is especially relevant since 25% of European households owned at least one cat or dog in 2021.[11]

The lack of data for veterinary care, specifically on companion animals, is partly addressed by the enactment of the EU Regulation 2019/6, which requires mandatory reporting of consumption and dispensing quantities of antimicrobial agents for dogs, cats and horses.[12] The regulation (EU) 2019/6 also harmonizes the rules on veterinary antimicrobial products in the EU and adds additional regulation for the use of antibiotics in animals,[12] but the monitoring of AMR in bacterial species in animals is not yet organized at European level. AMR monitoring is needed to evaluate the success of the new regulations and experts have therefore brought the proposal to establish the European Antimicrobial Resistance Surveillance network in Veterinary medicine (EARS-Vet), which could represent the current AMR situation in bacterial pathogens of animals in Europe.[13]

Given the significance of collecting and comparing AMR data from companion animals, we conducted a large-scale, Germany-wide analysis of routine diagnostic samples. MRSA prevalence was examined using data from different sample types from 2019–2021 for cats and dogs. To our knowledge, this represents the worldwide largest analysis of the occurrence of MRSA in companion animals to date.[9,14–16] The objective of the study is to provide a baseline of MRSA prevalence for cats and dogs in Germany, to evaluate the effect of new regulations and to interpret future AMR trends.

Methods

Ethics

This research was approved by the Central Ethics Committee of Freie Universität Berlin under Approval No. 2021-018.

Samples

All results of bacterial diagnostic samples from veterinary practices sent to Laboklin (accredited specialist laboratory for veterinary diagnostics) during 2019–2021 were analysed. The samples originated from dogs and cats presented at veterinary practices (total of 3491 practices and clinics) throughout Germany during this 3-year period. Sampling materials were blood, urine, different tissues and swabs of different origin. Depending on the anatomical origin, the samples were grouped into four categories:

(i) Skin and soft tissue (e.g. ear swabs)
(ii) Wounds (e.g. swabs from surgery wounds and abscesses)
(iii) Respiratory tract (e.g. nasal swabs and bronchoalveolar lavage)
(iv) Other (e.g. unknown or urinary samples)

Bacterial identification and antimicrobial susceptibility testing (AST) of *S. aureus*

Bacterial species were identified using MALDI-TOF-MS (Bruker Daltonics, Bremen, Germany) following standard protocols. For antimicrobial susceptibility testing (AST), the minimum inhibitory concentration (MIC) was determined by using the Micronaut system Merlin (Merlin GmbH, Bornheim-Hersel, Germany), performing broth microdilution assays and automated photometric evaluation of the individual customized microtiter plates. For feline and canine *S. aureus* isolates, the evaluation within the framework of the presented study focused on the following substances: cefoxitin, gentamicin, enrofloxacin, doxycycline, chloramphenicol, rifampicin, clindamycin and sulfamethoxazole + trimethoprim.

AST classification

The interpretation (S–I–R) of the MIC measured was performed according to the standardized procedures of the CLSI using the specifications of CLSI documents Vet01S-Ed6 and M100Ed33.[17,18] MRSA identification is based on a cefoxitin MIC >4 mg/L by the EUCAST v.13.0 and the CLSI M100Ed33. Dog-specific breakpoints from the CLSI document Vet01S-Ed6 were applied to enrofloxacin and clindamycin for all canine isolates, but also for feline isolates for clindamycin, as no feline-specific breakpoints were available. Human-specific clinical breakpoints from CLSI document M100 were applied for gentamicin, doxycycline, chloramphenicol, rifampicin and sulfamethoxazole + trimethoprim.

Human MRSA data

On the website of the Robert Koch-Institut, the data of the resistance statistics by ARS for the years 2019–2021 of all material groups, regions and specialties for the pathogen *S. aureus* in outpatients were queried (https://ars.rki.de/). ARS collects results of pathogen identification and resistance testing of the participating laboratories covering both inpatient hospital care and the outpatient care sector. Depending on the laboratory, either the guidelines of the CLSI or EUCAST were applied to evaluate the results. The MRSA prevalence was determined using the active substance cefoxitin.[8]

Statistical analysis

All statistical analyses were performed in R version 4.2.2 (R Foundation Vienna). Isolates and antimicrobial substances were identified using the R package AMR.[19] AST classification was performed in R using the previously described breakpoints. All results are reported using 95% Wilson confidence intervals (95% CI). Non-overlapping confidence intervals can be considered significantly different. The density map for Germany was created using coordinates based on first two digits of the postal codes of the submitting veterinary practice. A two-dimensional tensor spline was fitted with a Poisson regression using the number of resistant samples as outcome and the number of overall samples as offset

Table 1. Total number and percentages of samples/isolates per host species and year; total number and percentages of *S. aureus* overall and with regard to the assigned organ system per host species and year

	Overall	Dog	Cat
Samples	175 171	122 831	52 340
Samples with *S. aureus* isolated	**5526**	**2569**	**2957**
for which a FOX MIC is available (%)	5430 (98.3)	2516 (97.9)	2914 (98.5)
Year (%)	**5526 (100)**		
2019	2097 (37.9)	991 (38.6)	1106 (37.4)
2020	1698 (30.7)	787 (30.6)	911 (30.8)
2021	1731 (31.3)	791 (30.8)	940 (31.8)
Sample type (%)	**5526 (100)**		
respiratory tract	951 (17.2)	392 (15.3)	559 (18.9)
skin/soft tissue	1743 (31.5)	759 (29.5)	984 (33.3)
wound	977 (17.7)	503 (19.6)	474 (16.0)
other	1855 (33.6)	915 (35.6)	940 (31.8)

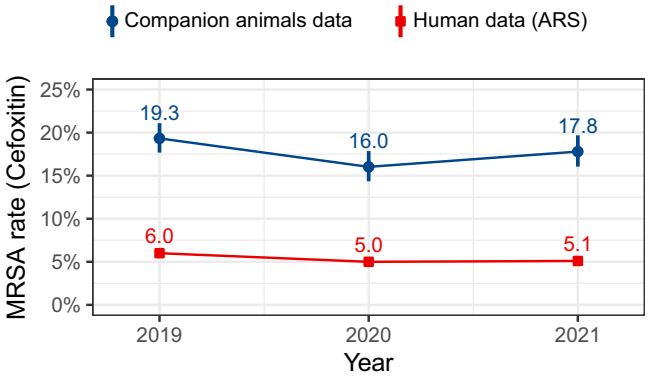

Figure 1. Proportion (%) of MRSA (defined by cefoxitin resistance) among *S. aureus* infections ($n = 5430$) in dogs and cats per year compared to outpatient human data (ARS RKI).[8] Error bars represent 95% confidence intervals. This figure appears in colour in the online version of *JAC* and in black and white in the print version of *JAC*.

(R package mgcv version 1.8-42). Our assumptions about the data generating process of routine or surveillance AMR data were visualized as a directed acyclic graph (DAG).

Results

In total, 175 171 results of a bacteriological examination of feline and canine specimens were available. A total of 27 917 samples (19 154 canine, 8763 feline) did not yield any growth of specific pathogenic bacterial species.

Overall, *S. aureus* was identified in 5526 samples. Each sample only contained exactly one *S. aureus* isolate. *S. aureus* was isolated in about 3.2% of all available samples, with varying occurrence rates between cats (5.6%) and dogs (2.0%). The pathogen was isolated in 6.0% of the 16 111 wound samples, 4.4% of the 21 398 respiratory tract samples, 2.6% of the 67 293 skin/soft tissue samples, and 2.6% of the 70 370 other samples. 96 *S. aureus* samples lacked a valid MIC for cefoxitin. The respective numbers of samples in which *S. aureus* was isolated and to which organ system they were assigned are listed in Table 1.

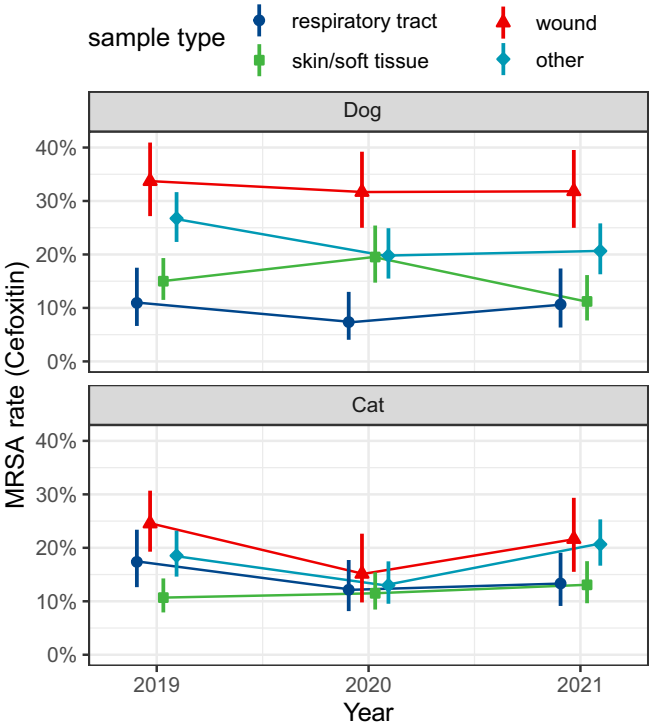

Figure 2. Proportion (%) of MRSA (defined by cefoxitin resistance) among *S. aureus* infections of four different sample origins (skin and soft tissue, wound, respiratory tract and other) of dogs and cats per year. Error bars represent 95% confidence intervals. This figure appears in colour in the online version of *JAC* and in black and white in the print version of *JAC*.

Overall, 17.8% (95% CI 16.8–18.9, $n = 968$) of the investigated *S. aureus* strains exhibited phenotypic methicillin resistance. When comparing host species, *S. aureus* isolated from dogs presented a resistance rate of 20.4% (95% CI 18.9–22.0, $n = 513$), which surpassed the 15.6% (95% CI 14.3–17.0, $n = 455$) resistance rate in *S. aureus* isolated from cats. The MRSA prevalence in our clinical samples was stable between 2019 and 2021 as seen in Figure 1.

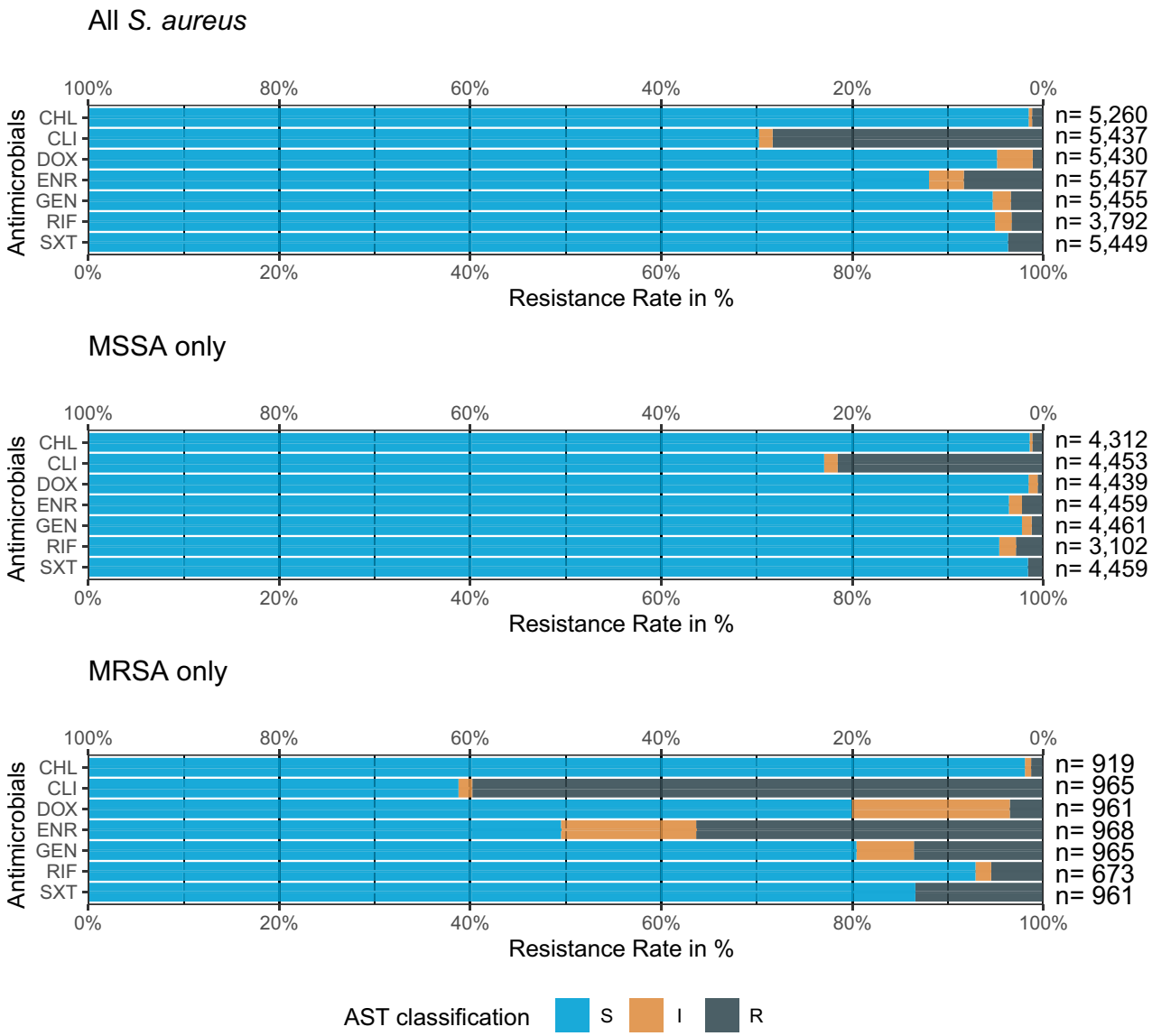

Figure 3. Percentage of antimicrobial resistance in all canine and feline *S. aureus* isolates ($n = 5526$) compared to resistance only in MSSA ($n = 4462$) and MRSA isolates ($n = 968$). Abbreviations: S, susceptible; I, intermediate; R, resistant. Antimicrobials: GEN, gentamicin; ENR, enrofloxacin; DOX, doxycycline; CHL, chloramphenicol; RIF, rifampicin; CLI, clindamycin; SXT, sulfamethoxazole + trimethoprim. This figure appears in colour in the online version of *JAC* and in black and white in the print version of *JAC*.

MRSA resistance rates in cats showed nearly no difference when comparing sample types. In dogs, especially *S. aureus* originating from wound samples showed a high resistance rate of over 30%. Samples originating from the respiratory tract in dogs on the other hand had a lower MRSA prevalence of around 10% (Figure 2). There were only 63 urine samples containing *S. aureus* overall, which is why we included them in 'other' instead of analysing them separately.

MRSA were generally more resistant to the tested non-beta-lactam antibiotics than methicillin-susceptible *S. aureus*. Co-resistance to clindamycin (59.8%, 95% CI 56.7–62.8) and enrofloxacin (36.4%, 95% CI 33.4–39.4) was particularly high. Of the MRSA samples, 13%–14% were resistant to sulfamethoxazole + trimethoprim and gentamicin, while resistance in chloramphenicol, doxycycline and rifampicin occurred in less than 6% of the samples (Figure 3). The underlying data are available as Supplementary Data (at *JAC* Online) (File S1).

Figure 4 shows that MRSA was detected in all German federal states. High occurrence rates of more than 25% were found in the northern part of Bavaria, in the southern part of Thuringia and in west Lower-Saxony. Cold spots with 15% MRSA prevalence were detected in the south of Germany as well as central-northern Germany and southwestern Germany.

Discussion

Our study provides the Germany-wide MRSA prevalence in different samples of clinically diseased cats and dogs over a 3-year period. To our knowledge, this is the largest study of AMR in

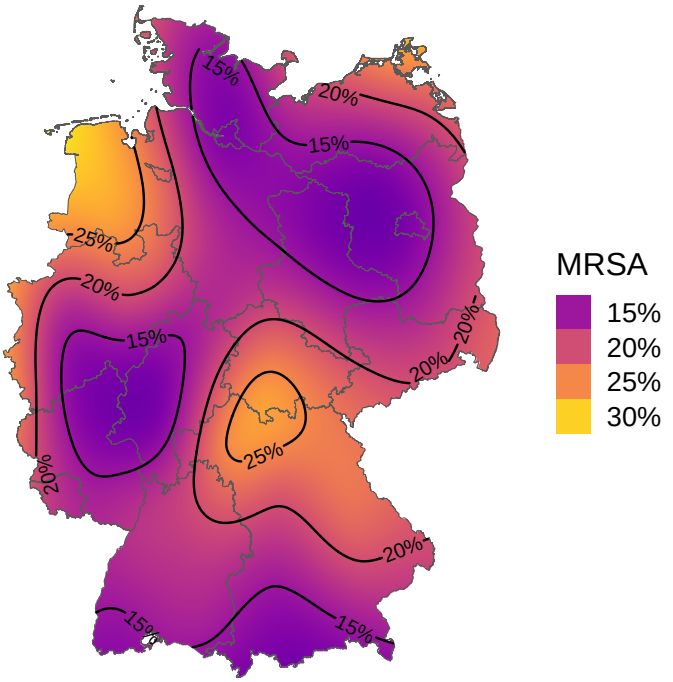

Figure 4. Map of Germany showing the proportion (%) of MRSA among *S. aureus* infections in dogs and cats based on 5430 *S. aureus* isolates. The borders of the German federal states are provided by GADM for academic use (GADM license). This figure appears in colour in the online version of *JAC* and in black and white in the print version of *JAC*.

clinical samples from companion animals to date, encompassing 175 171 samples. Furthermore, it is the first study to provide comprehensive data on a national scale, covering 3491 (33.1%) of the 10 558 registered practices and clinics in Germany in 2021.[20] This is comparable to the 31.7%–37.6% coverage of the human AMR surveillance system ARS in Germany.[8]

Overall, our MRSA prevalence for Germany between 2019 and 2021 was 17.8% which is similar to the MRSA prevalence reported by the German veterinary AMR surveillance system GERM-Vet of 11.1%–19.3% (6/31 in 2019, 5/45 in 2020) for dogs and cats.[9] AMR surveillance systems of other countries reported an MRSA prevalence in dogs and cats of 7% for Finland ($n=94$ *S. aureus* isolates, 2021)[15] and 10%–20% for France (*n* not reported, 2021).[14] Sweden, Denmark, Norway, Switzerland[21] and China[16] do not currently monitor MRSA in companion animals. However, the small sample sizes in most official systems introduces a high degree of uncertainty, which limits comparability and makes it difficult to analyse annual trends.

The data for food-producing animals published via GERM-Vet shows that the prevalence of MRSA in pigs was 48% in 2020 (11 of 23 isolates) and 65% in 2018 to 2019 (17 of 26 isolates), and therefore more than double the prevalence of MRSA in companion animals.[9] However, it is important to consider that companion animals have rarely contact with food-producing animals in Germany, as these are mainly kept in intensive farming systems. Instead, companion animals and humans share lifestyle factors and living environments, resulting in close physical interactions, with hugs, kisses and shared sleeping spaces.[22]

Hence, the comparison with human AMR data is more meaningful. We used human outpatient data of the German AMR surveillance system ARS for *S. aureus* in the years 2019–2021. MRSA rates for isolates of human origin were on average 5.4% and therefore notably lower than the 17.8% rate in *S. aureus* of cats and dogs.[8]

Transmission of MRSA between companion animals and humans presents a risk to both. While previous studies identified several risk factors for MRSA infections in dogs and cats—including multiple courses of antimicrobials, extended veterinary admissions, recent surgical implants, ongoing infections, contact with recently hospitalized humans, surgical site infections and veterinary practices with more than 10 employees[23,24]—Hackmann *et al.* conducted a meta-analysis showing that dog owners have a 2.3 times higher risk of MRSA carriage compared to non-pet owners.[10] This risk increase for pet owners has been commonly attributed to the pet being a vector for MRSA.[10,25] In previous studies it has further been shown that MRSA isolates from companion animals are closely genetically related to local human isolates.[3,26,27] Thus, humans and companion animals act as mutual MRSA reservoirs or have common sources of infection. Therefore, reducing MRSA rates is beneficial for both. This is especially important considering that pet ownership is common in the EU, with approximately 25% of households (90 million) owning at least one pet animal, resulting in a total population of 72 million dogs and 83 million cats.[11] Of course, the risk of transmission also depends on the closeness of the pet owner's contact with his pet. To reduce AMR rates, a One Health approach is needed, as exemplified by the WHO's global action plan.[7]

Among animals, *S. aureus* samples from dogs exhibited a higher MRSA prevalence of 20.4% compared to cats at 15.6%. At over 30%, the MRSA prevalence in wound samples from dogs is particularly high. High MRSA rates in samples from canine wound samples have been repeatedly described before.[27,28] In veterinary medicine nosocomial surgical site infections have a higher MRSA rate than non-nosocomial infections.[24] Surgical site infections being classified as wound samples could explain these high MRSA rates. However, MRSA rates in respiratory tract and skin/soft tissue samples were similar between dogs and cats. In cats the difference between sample types was negligible (see Figure 2).

One limitation of our study is, that data collection and analysis is not ongoing or organized as a surveillance programme. Continuous resistance monitoring is needed to evaluate the effectiveness of antibiotic use regulations and AMR control programmes. In addition, knowledge of AMR rates can increase awareness among veterinarians. Antimicrobial stewardship needs AMR surveillance for understanding local susceptibilities (see Figure 4) and their evolving trends.[29] This can improve the prudent use of antibiotics by guiding empirical therapy. Right now, only France has, with AST results from 25 139 isolates in 2021, a comprehensive surveillance of AMR in companion animals.[14] For the EU a large-scale AMR surveillance system with relevant coverage for companion animals is needed, but little progress has been made in the last 10 years.[3]

Evaluating regulations on AMU in veterinary medicine is crucial, as limiting antibiotics for companion animals leads to reduced animal welfare if infections stay untreated. Therefore, limiting access to treatment should not be the primary choice[1] and less severe methods to reduce AMR rates should be considered first. This evidence for effective AMR control strategies should be collected through AMR surveillance.[7]

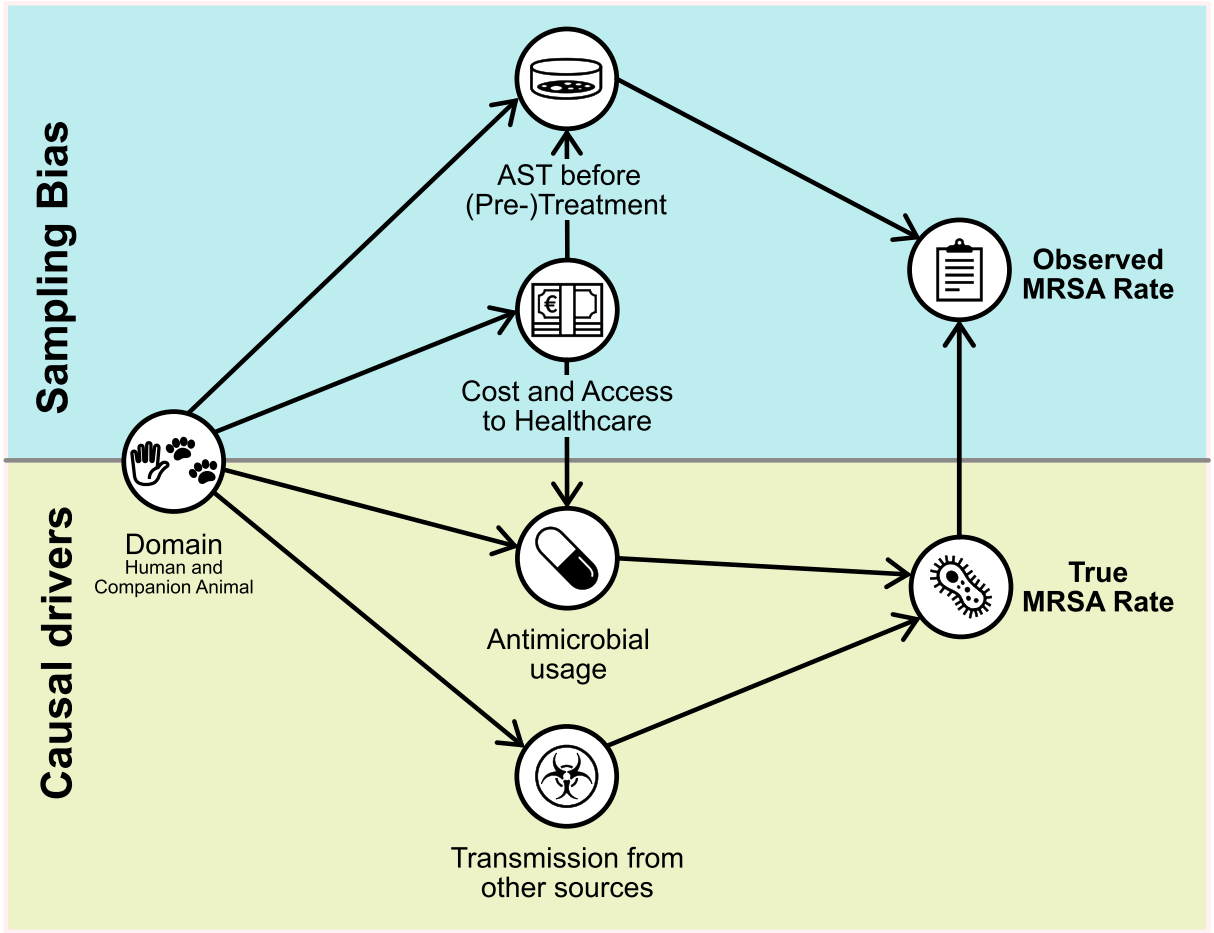

Figure 5. Causal DAG for the data generating process of surveillance scale AMR data in companion animals. The causal drivers of AMR (green, true MRSA prevalence) are mainly transmission and selection (antimicrobial usage) of bacteria with a resistant phenotype.[5] But the observed MRSA prevalence for large-scale AMR data is also affected by sampling bias (blue). Here the biggest difference between human and companion animal medicine is the cost and access to healthcare. In veterinary medicine, cost often limits the number of ASTs, and ASTs are often only conducted if treatment failed.[30] This results in a higher rate of pre-treated AST samples and pretreatment increases the observed resistance rates. This figure appears in colour in the online version of *JAC* and in black and white in the print version of *JAC*.

The observed difference in MRSA prevalence between companion animal and human laboratory AMR data can be due to causal and biasing effects. As shown in the causal graph (Figure 5), there are two outcomes: the true MRSA prevalence we want to know, and the observed MRSA prevalence that we actually measure with laboratory AMR surveillance. The true MRSA prevalence is affected by causal effects that select for bacteria with a resistant phenotype, such as antimicrobial usage, or transmission of resistant bacteria from other sources (human beings, animals, environment).[5] By contrast, the observed MRSA prevalence is a product of the true prevalence modified by sampling bias. In both companion animals and humans, not every antibiotic treatment is preceded by an antibiogram (i.e. AST). This introduces a sampling bias due to missing AST data from infections that are successfully treated without prior testing. Antibiograms are often performed only after initial treatments fail.[30] This leads to an overrepresentation of pre-treated cases in routine AMR data, since isolates from pre-treated samples are generally more resistant.[24] This bias increases the observed resistance rates. As a result, expanding the use of AST would probably reduce the observed resistance rate, even if the true rate remains unchanged. This sampling bias in routine AST data limits the comparability of pet and human resistance rates due to differences in cost and access to health care. Most visits to a veterinarian are usually paid out of pocket, with additional costs for AST.[30] In human medicine medical insurance is common and therefore cost for the patient is not an access barrier for AST. While both human and small animal medicine experience an upward bias of the observed AMR rates due to sampling bias, this bias probably impacts the small animal AMR rates more, but whether this is the case is difficult to assess. Differences in AST between veterinary and human medicine are also influenced by veterinary antimicrobial use regulations[31] and missing guidelines, including specific therapy recommendations, especially in veterinary medicine in Germany.

In our study, we employ a passive monitoring approach, by collecting routine clinical AMR results data. Since in-depth analyses such as genome sequencing are not routinely performed in clinical practice, this information is unavailable for the isolates in our dataset. Therefore, passive surveillance should be

complemented by active surveillance. Active surveillance provides in-depth analysis for a subset of isolates and can therefore provide information about the genetic factors driving the change in resistance patterns.[32] This active approach is already established in Germany within GERM-Vet.[9]

In our study information about repeated/duplicated samples was not available. Repeated samples could result in an upward bias in the observed resistance rates. According to expert opinion of our data provider the amount of duplication was considered low at <1% of samples, since most data was collected from outpatient visits.

From 2029 onwards it will be mandatory for every veterinary practice to report AMU for dogs and cats as required by the EU Regulation 2019/6.[12] This AMU surveillance will affect all 10558 registered practices and clinics in Germany.[20] In comparison, establishing a comprehensive AMR surveillance is reliant on only a few laboratory providers. As shown by this study, future integration of laboratory AMR data could therefore complement national veterinary AMR surveillance systems, like GERM-Vet in Germany.

Conclusion

In conclusion, our study provides comprehensive data on the prevalence of MRSA in clinically diseased companion animals across Germany over a 3-year period, with a coverage of 33.1% of the registered veterinary practices. The observed MRSA prevalence in companion animals is higher than in humans, but since this comparison is biased, as described in this paper, further research is needed. The need to reduce MRSA rates should be viewed through the One Health lens, especially given the high prevalence of pet ownership. The potential detriment to animal welfare due to restricted antibiotic use necessitates that new and existing measures are evaluated using AMR surveillance. Moreover, awareness of AMR rates among veterinarians can be improved this way, thereby fostering prudent antibiotic usage. Thus, there is an urgent need for large-scale passive AMR surveillance for companion animals. This enhances the existing active surveillance which is needed to get in-depth results for a subset of isolates. Our study is a first step showing continuous AMR monitoring is feasible with significant coverage in a practical way.

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Transparency declarations

The authors declare that there is no conflict of interest regarding the publication of this article. A.Ba., L.F. and R.M. conceptualized the study. R.M. and W.B. provided supervision, administrative support and acquired funding. B.K. provided the data. L.F., A.Ba., K.F., A.Be., A.L.B. and R.M. accessed and verified the data. A.Ba. and L.F. performed data analysis, and visualization. A.Ba., L.F., R.L. and B.K. performed data interpretation. L.F. and A.Ba. wrote the original draft. All authors reviewed and edited the final draft. All authors had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Data availability

The data used for the analysis are provided as a .csv file in the Supplementary Data (File S1). The file contains information on species, year, sample type and AST classifications. Geographical information is not made available due to privacy reasons.

Supplementary data

File S1 is available as Supplementary data at the *Journal of Antimicrobial Chemotherapy* Online.

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