



In vitro lytic efficiency of *Staphylococcus aureus* bacteriophages in bacteria from bovine mastitis: a meta-analysis

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ABSTRACT: Bacteriophages have been investigated as alternative to the treatment of bacterial infections, including bovine mastitis, in production animals. In this meta-analysis, we evaluated *in vitro* efficiency of phages of *Staphylococcus aureus* against *S. aureus*, which is involved in the etiology of bovine mastitis. Seventeen studies were included and the bacterial lytic activity was extracted using proportion analysis. The lytic efficiency of phages was obtained in this meta-analysis using a random-effects model [significant difference ($P < 0.05$)]. Forest plots were used to graphically represent the efficiency of phages on bacterial isolates. Most phages (e.g., CSI, DW2, ΦSA011, ΦSA012, ΦSA022, ΦSA023, ΦSA024, ΦSA025, ΦSA037, ΦSA038, ΦSA039, ΦSA041, ΦSA042, ΦSA043, ΦSA044, MSA6, Ufv-aur2 to Ufv-aur11, SAH-1, SPW, vB_SauM_JS25, SaPh1 to SaPh6, SA, SANF, SA2, ΦSA012, ΦSA039, phi11, phiPLA88, phiPLA35, phiPLA-RODI, phiPLA-C1C, SAJK-IND, vBSP-A1, vBSP-A2, STA1.ST29, EB1.ST11, EB1.ST27, Remus, and ISP) were efficiently lytic or infected most *S. aureus* isolates, demonstrating 80% ($P < 0.05$) lytic efficiency. The phages SA, SANF and SA2, also demonstrated lytic activity or infected the non-*Staphylococcus aureus* and *Micrococcus caseolyticus* isolates. In this meta-analysis, we compared and demonstrated the *in vitro* efficiency and host range of *S. aureus* phages. Additionally, the phages represent an alternative to be researched to treat bovine mastitis in dairy cattle caused by the prevalent microorganism, *S. aureus*.

Key words: bacteriophages, bovine mastitis, phages, *Staphylococcus aureus*.

Eficiência lítica *in vitro* de bacteriófagos de *Staphylococcus aureus* em bactérias de mastites bovina: uma meta-análise

RESUMO: Os bacteriófagos têm sido investigados como alternativa ao tratamento de infecções bacterianas em animais de produção, incluindo a mastite bovina. Nesta meta-análise, avaliamos a eficiência *in vitro* de fagos de *Staphylococcus aureus* contra *S. aureus* envolvidas na etiologia da mastite bovina. Dezesete estudos foram incluídos e a atividade lítica bacteriana foi extraída usando análise de proporção. A eficiência lítica dos fagos foi obtida nesta meta-análise, usando um modelo de efeitos aleatórios (diferença significativa ($P < 0,05$)). Os gráficos de Forest plots foram usados para representar graficamente a eficiência dos fagos em isolados bacterianos. Os fagos avaliados, na sua grande maioria, (por exemplo, CSI, DW2, ΦSA011, ΦSA012, ΦSA022, ΦSA023, ΦSA024, ΦSA025, ΦSA037, ΦSA038, ΦSA039, ΦSA041, ΦSA042, ΦSA043, ΦSAfe Uof, ua04 SPW, vB_SauM_JS25, SaPh1 a SaPh6, SA, SANF, SA2, ΦSA012, ΦSA039, phi11, phiPLA88, phiPLA35, phiPLA-RODI, phiPLA-C1C, SAJK-IND, vBSP-A1, vBSP-A2, STA1.ST29, EB1.ST11, EB1.ST27, Remus, e ISP) foram eficientemente líticos ou infectaram a maioria dos isolados de *S. aureus*, demonstrando 80% ($P < 0,05$) de eficiência lítica. Os fagos SA, SANF e SA2 também demonstraram atividade lítica ou infectaram os isolados *Staphylococcus* não-*aureus* e *Micrococcus caseolyticus*. Nesta meta-análise, comparamos e demonstramos a eficiência *in vitro* e gama de hospedeiros de fagos de *S. aureus*. Adicionalmente, os fagos representam uma alternativa a ser pesquisada para o tratamento da mastite bovina em gado leiteiro causada pelo microrganismo prevalente, ou seja *S. aureus*.

Palavras-chave: bacteriófagos, mastite bovina, fagos, *Staphylococcus aureus*.

INTRODUCTION

Bovine mastitis is defined as an inflammation of the mammary gland and the use of antimicrobials to treat of this disease in dairy cows

has increased (GRAVE et al., 1999; MITCHELL et al., 1998; SHARMA; SINGH; BHADWAL, 2011). Among the etiological agents involved in the occurrence of mastitis of bacterial origin, *S. aureus* has been reported as the most prevalent microorganism

globally (BARKEMA; et al., 2006; SOUZA, et al., 2016; SEYOUM et al., 2017; SULEIMAN et al., 2018; MESQUITA, et al., 2019). Furthermore, an increase in penicillin resistance has been reported, as well as methicillin-resistant *S. aureus* (MRSA) in bovine milk (AQIB et al., 2017; SILVA et al., 2020). Once, the penicillin resistance is related to indiscriminate use of antibiotics with the absence of laboratory diagnosis and the antimicrobials sensitivity test to in the bovine mastitis, mainly caused by *S. aureus* (BEURON et al., 2014).

Therefore, bacterial resistance to available antimicrobials has increased, including in the veterinary field (PALMA et al., 2020). Thus, the discovery of alternatives to control bovine mammary gland infections is critical and therapeutic alternatives to treat bacterial infections, including the use of bacteriophages or phages, have been studied globally (BASDEW & LAING, 2011; ROHDE et al., 2018).

Phages infect bacteria, causing cell lysis; consequently, they are considered potential therapeutic agents for more than a century (D'HERELLE, 1931). These viruses have received attention from many research groups (SULAKVELIDZE, 2004; MONK et al., 2010; SILLANKORVA & AZEREDO, 2014), owing to their characteristics, including host specificity, lytic activity in pathogenic bacteria, absence of harm to humans and animals, and rapid application to combat the emergence of new bacterial threats (SULAKVELIDZE & ZEMPHIRA; JR, 2001).

The mechanism of action of phages is completely different from those of antimicrobials and they are effective against multi-drug-resistant bacteria. Moreover, owing to their environmental presence, it is easier to find new phages than to develop new drugs or immunobiologicals (VEIGA-CRESPO; BARROS-VELÁZQUEZ; VILLA, 2010). Therefore, phage therapy is considered a promising alternative compared to the use of antimicrobials (PELFRENE et al., 2016), making this study remarkably relevant.

In vitro studies have investigated the use of phages in the biological control of bovine mastitis; however, variable results have been reported (SON et al., 2010; BASDEW & LAING, 2011; IWANO et al., 2018). Therefore, these studies were selected for this meta-analysis, as well as standardized by the eligibility criteria to homogenize the samples. In addition, the studies with *in vivo* tests were not included because they did not meet the eligibility criteria. Thus, in this study, we performed a meta-analysis of *in vitro* efficiency of *S. aureus* phages against isolates *S. aureus* involved in bovine mammary-gland infections.

MATERIALS AND METHODS

Search strategies

A systematic review of the literature was performed, identifying studies that tested the lytic activity of phages of *S. aureus* isolates from bovine mastitis. The investigation was developed by pairs, focusing on studies that determine the host range of the phages. This review was conducted in the following four stages: identification, selection, eligibility assessment, and inclusion, according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (MOHER et al., 2009).

Selection of studies

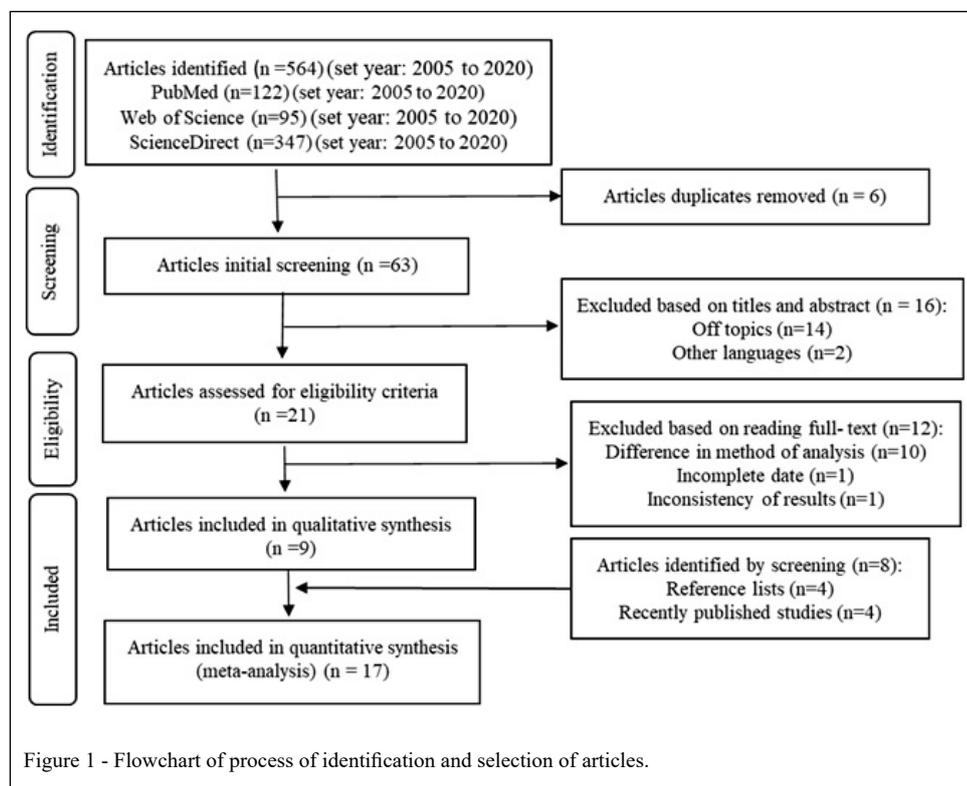
Bibliographic research was performed using data from scientific journals indexed in the following databases: PubMed, Web of Science and ScienceDirect, using the search terms: phage OR phages OR bacteriophage OR bacteriophages AND mastitis NOT humans. Only publications in the English language were considered for this analysis (PURBA et al., 2020; Figure 1).

During the selection phase, duplicate publications were removed. Subsequently, the studies were evaluated based on the title and abstract. All studies related to tests using the lytic enzymes were excluded, as well as literature reviews.

The articles included in this meta-analysis were comprehensively assessed through the complete reading of the selected texts. The selected papers fulfilled the following eligibility criteria: *S. aureus* isolated from bovine mastitis, host-specific phages of *S. aureus* of bovine milk, determination of the range hosts, methodology that employed the use of a single phage, no association with other phages, and the performance of *in vitro* analyses. Additionally, the plaque assay, spot test and double-layered agar methods were included.

Notably, we excluded studies that reported the concomitant use of other molecules with antimicrobial potential, as well as those that included intracellular bacteria or other analyses that did not allow the visualization of lysis. These exclusions were necessary to ensure the homogeneity of samples to compare studies that were included in this meta-analysis.

The references cited in the articles were also analyzed and included in the present study based on the eligibility criteria described above. Thus, the information from the articles was systematized in a spreadsheet (Microsoft Excel®). The bacterial lytic activity was extracted using proportion analysis.



Statistical analysis

Initially, a logit transformation of the proportion data was performed based on the total number of bacterial isolates (total) *versus* the number of lysed bacterial isolates (events). Additionally, this procedure was used for each phage isolated and tested in each study. The lytic efficiency of *S. aureus* phages was obtained using random-effects model [significant difference ($P < 0.05$)]. Forest plots were used to graphically represent the meta-analysis result. In particular, the meta-analysis was conducted using the “Metaprop” package of R v3.4.2 software®.

RESULTS

Selection of studies

A total of 564 articles were found, including 122 studies from PubMed, 95 from Web of Science and 347 from ScienceDirect. Next, the articles were screened, and 21 were chosen and read completely, after which the eligibility criteria were evaluated. Consequently, nine articles met all eligibility criteria for this meta-analysis. The bibliographic references of the selected studies were reviewed and two other scientific publications were included. We performed

a general search using the same indexers and other websites for studies that were not reviewed, as well as recently published studies to update the references. Thus, eight studies were included, resulting in a total of 17 articles that were analyzed in this current study (Figure 1). The period of publication of the selected articles ranged from June 2005 to September 2020. Information regarding the following aspects was obtained from each article: phage isolates, phage family, bacteria isolated from bovine milk, the presence of antimicrobial resistance or resistance genes, and techniques for analyzing the occurrence of bacterial lysis (Table 1 and Table 2).

Occurrence of lytic activity and host range

The total number of bacterial isolates analyzed in the studies was 603, of which 92.37% (557/603) were *S. aureus*, which included 47.21% (263/557) of isolates that were resistant and 52.78% (294/557) of isolates that were non-resistant to antimicrobials. Based on the analysis of the Forest plots (Figure 2 and Figure 3), it was evident that the phages lysed the bacteria isolated from bovine milk samples. However, the lytic activity presented was variable in relation to the different phages and bacterial

Table 1 - Main characteristics of the 17 studies that were selected starting systematic review and included in the meta-analysis database.

Author	Country	Phages family	Phages isolates	Analysis method	Bacterial isolated	N ^o **	Antimicrobial resistance
O'FLAHERTY et al., (2005)	Ireland	<i>Siphoviridae</i>	CS1 DW2	Spot test	<i>S. aureus</i>	3	N/A
SYNNOTT et al., (2009)	Japan	<i>Myoviridae</i>	ΦSA003 ΦSA004 ΦSA011 ΦSA012 ΦSA022 to ΦSA026 ΦSA037 to ΦSA039 ΦSA041 to ΦSA044	Double-layered agar	<i>S. aureus</i>	16	N/A
GARCÍA et al., (2009)	Spain	N/A	ΦA72 ΦH5 ΦL7 ΦL13 ΦA8 ΦG7	Double-layered agar	<i>S. aureus</i>	14	N/A
SON et al., (2010)	Korea	<i>Myoviridae</i>	SAP-1 SAP-3	Double-layered agar	<i>S. aureus</i> ; <i>S. epidermidis</i> ; <i>S. agalactiae</i> ; <i>S. uberis</i> ; <i>E. faecalis</i> ; <i>E. coli</i> ; <i>L. plantarum</i> ; <i>C. koseri</i> ; <i>S. haemolyticus</i>	20	Nal, Str, Gen, Ami, Ct, Cfl, Nor, Eri, Oxa
KWIATEK et al., (2012)	Poland	<i>Myoviridae</i>	MSA6	Double-layered agar	<i>S. aureus</i> ; <i>S. epidermidis</i> ; <i>S. saprophyticus</i>	35	MRSA, VRSA
DIAS et al., (2013)	Brazil	<i>Myoviridae</i>	Ufv-aur2 to Ufv-aur11	Double-layered agar	<i>S. aureus</i>	20	Amp, Gen, Pen, Tet, Eri, Rif, Ami, Clo, Cro, Oxa, Cfl, Cli, Cip, Van, Sut
HAN et al. (2013)	Korea	<i>Myoviridae</i>	SAH-1	Double-layered agar	<i>S. aureus</i> ; <i>E. faecalis</i>	47	MRSA
LI & ZHANG (2014)	China	<i>Myoviridae</i>	SPW	Spot test	<i>S. aureus</i> ; <i>E. coli</i>	05	MRSA

MRSA: methicillin-resistant *S. aureus*. VRSA: vancomycin-resistant *S. aureus*. Nal: nalidixic acid. Str: streptomycin. Ct: colistin. Gen: gentamycin. Ami: ampicillin. Cfl: cephalothin. Nor: norfloxacin. Eri: erythromycin. Oxa: oxacillin. Amp: ampicillin. Cfo: cefoxitin. Van: vancomycin. Dc: dicloxacillin. Pef: pefloxacin. Cxm: cefuroxime. Ctx: cefotaxime. Sxt: sulfamethoxazole-trimethoprim. Caz: ceftazidime. Met: methicillin. Pen: penicillin. Pen G: penicilina G. A/F: amoxicillin/ fucloxacillin. Ob: oxytetracycline. Tr: trimethopim. Tet: tetracycline. Rif: rifampicin. Clo: chloramphenicol. Cli: clindamycin. Cip: ciprofloxacin. Sut: sulfazotrim. **Number of isolates bacterial from each study. N/A: Not available.

isolates tested. Therefore, most *S. aureus* phages (e.g. CS1, DW2, ΦSA011, ΦSA012, ΦSA022, ΦSA023, ΦSA024, ΦSA025, ΦSA037, ΦSA038, ΦSA039, ΦSA041, ΦSA042, ΦSA043, ΦSA044, MSA6, Ufv-aur2 to Ufv-aur11, SAH-1, SPW, vB_SauM_JS25, SaPh1 to SaPh6, SA, SANF, SA2, ΦSA012, ΦSA039, phi11, phiIPLA88, phiIPLA35, phiIPLA-RODI, phiIPLA-C1C, SAJK-IND, vBSP-A1, vBSP-A2, STA1.ST29, EB1.ST11, EB1.ST27, Remus, and ISP) were efficiently lytics or infected most of *S. aureus* isolates tested, except phages ΦA72, ΦH5, ΦL7, ΦL13, ΦA8, ΦG7, ΦSA003, ΦSA004, ΦSA026,

SAP-1, SAP-3, MSP, Romulus, and DSM105264, which presented low lytic efficiency or infected a low number of *S. aureus* isolates. The phages demonstrated 80% ($P < 0.05$) lytic efficiency against *S. aureus* isolates (Figure 2). Additionally, the lytic activity of phages is species-specific, that is, these viruses are only able to destroy their bacterial host. Conversely, the phages SANF, SA2 and SA also demonstrated lytic activity for isolates of non-*S. aureus* bacteria (e.g. *Staphylococcus chromogenes*, *Staphylococcus saprophyticus*, *Staphylococcus xylosus*, *Staphylococcus sciuri*, and *Staphylococcus*

Table 2 - Main characteristics of the 17 studies that were selected starting systematic review and included in the meta-analysis database (continued).

Author	Country	Phages family	Phages isolates	Analysis method	Bacterial isolated	N ^{o**}	Antimicrobial resistance
ZHANG et al. (2014)	China	<i>Myoviridae</i>	vB_SauM_JS25	Spot test	<i>S. aureus</i>	52	N/A
BASDEW & LAING (2015)	South Africa	<i>Myoviridae</i>	SaPh1 to SaPh6	Spot test	<i>S. aureus</i>	4	Amp, Pen G, Met, A/F, Eri, Str, Tet, Ob, Van, Tr
TAHIR et al. (2017)	Pakistan	N/A	SA SANF SA2	Spot test	<i>S. aureus</i> ; <i>S. xylosus</i> ; <i>M. caseolyticus</i> ; <i>S. saprophyticus</i> ; <i>S. succinus</i> ; <i>S. sciuri</i>	11	N/A
IWANO et al. (2018)	Japan	<i>Myoviridae</i>	ΦSA012 ΦSA039	Spot test	<i>S. aureus</i>	93	N/A
VARELA-ORTIZ et al. (2018)	México	<i>Siphoviridae</i> <i>Myoviridae</i>	phi11 phiIPLA88 phiIPLA35 phiIPLA-RODI phiIPLA-C1C	Spot test	<i>S. aureus</i>	27	Pen, Dc, Amp, Tet, Ctx, Cfl, Pef, Gen, Eri, Stx, Caz, Cxm
GANAIIE et al. (2018)	India	<i>Myoviridae</i> <i>Podoviridae</i>	SAJK-IND MSP	Spot test	<i>S. aureus</i> ; <i>E. coli</i> ; <i>S. agalactiae</i> ; <i>K. pneumoniae</i> ; <i>P. aureuginosa</i>	125	MRSA
GENG et al. (2019)	China	<i>Myoviridae</i> <i>Podoviridae</i>	vBSM-A1 vBSP-A2	Double-layered agar	<i>S. aureus</i> ; <i>S. chromogenes</i> ; <i>K. pneumoniae</i> ; <i>S. parasanguinis</i> <i>A. pyogenes</i> ; <i>S. agalactiae</i> ; <i>E. coli</i>	29	N/A
TITZE et al. (2020)	Germany	<i>Myoviridae</i> <i>Podoviridae</i>	STA1.ST29 EB1.ST11 EB1.ST27	Spot test	<i>S. aureus</i>	92	N/A
NGASSAM-TCHAMBA et al. (2020)	Belgium	N/A	Romulus Remus ISP DSM105264	Spot test	<i>S. aureus</i>	10	Cfo, Amp, Tet, Amo, Cip, Cli, Ct, Eri, Ff, Gen, Lzd, Pen, Tr, Sulfatrim, Sulfamycin,

MRSA: methicillin-resistant *S. aureus*. VRSA: vancomycin-resistant *S. aureus*. Nal: nalidixic acid. Str: streptomycin. Ct: colistin. Gen: gentamycin. Ami: ampicillin. Cfl: cephalothin. Nor: norfloxacin. Eri: erythromycin. Oxa: oxacillin. Amp: ampicillin. Cfo: ceftazidime. Van: vancomycin. Dc: dicloxacillin. Pef: pefloxacin. Cxm: cefuroxime. Ctx: cefotaxime. Sxt: sulfamethoxazole-trimethoprim. Caz: ceftazidime. Met: methicillin. Pen: penicillin. Pen G: penicilina G. A/F: amoxicillin/ fucloxacillin. Ob: oxytetracycline. Tr: trimethopim. Tet: tetracycline. Rif: rifampicin. Clo: chloramphenicol. Cli: clindamycin. Cip: ciprofloxacin. Sut: sulfazotrim. Ff: Florfenicol. Lzd: Lizenoid. **Number of isolates bacterial from each study. N/A: Not available.

succinus) and other species (e.g. *Macrocooccus caseolyticus*) (Figure 3). Therefore, the lytic efficiency was 30% for non-*S. aureus* bacteria and 16% for other bacterial species (e.g. *Streptococcus agalactiae*, *Streptococcus uberis*, *Streptococcus parasanguinis*, *Arcanobacterium pyogenes*, *Kocuria rosea*, *Enterococcus faecalis*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Lactobacillus plantarum*, *Citrobacter koseri*, and *Macrocooccus caseolyticus*) ($P < 0.05$) (Figure 3).

DISCUSSION

Data analysis and review revealed that most phages of *S. aureus*, specifically in the family *Myoviridae*, presented prominent and specific activity against 603 bacterial isolates from bovine mastitis. These findings demonstrate that these phages may be studied for the development of therapeutic alternatives for bovine mastitis and could be used to target infections involving *S. aureus*.

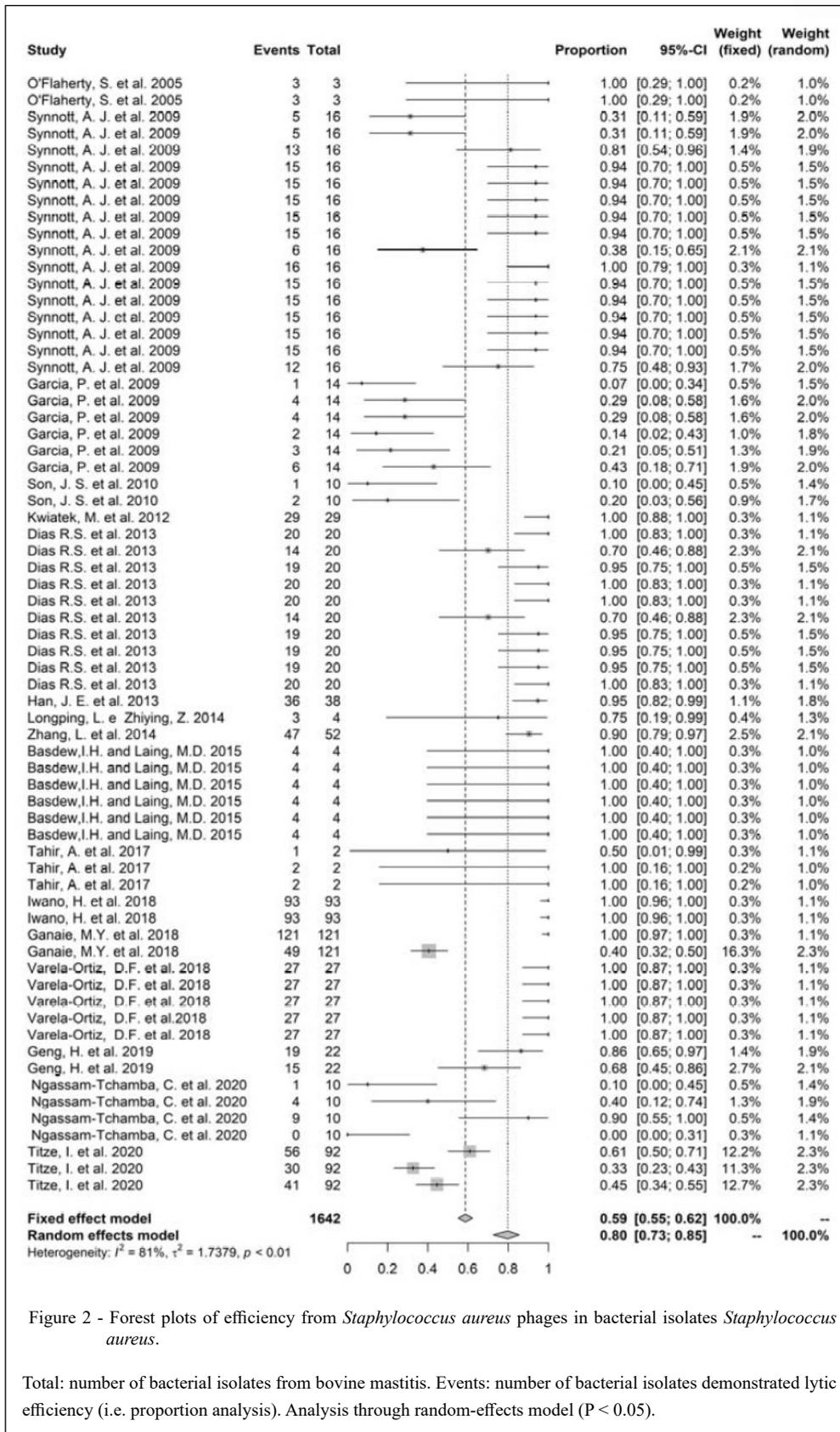


Figure 2 - Forest plots of efficiency from *Staphylococcus aureus* phages in bacterial isolates *Staphylococcus aureus*.

Total: number of bacterial isolates from bovine mastitis. Events: number of bacterial isolates demonstrated lytic efficiency (i.e. proportion analysis). Analysis through random-effects model ($P < 0.05$).

Additionally, the efficiency of the phages against several bacterial isolates permitted us to verify their probable infectivity and lethality in bacterial isolates responsible for bovine mastitis, but principally those against *S. aureus*. As shown in Figure 2, the lytic efficiency against *S. aureus* isolates was 80%. The results of this meta-analysis also verified the possibility of using the analyzed phages to infect other bacterial species involved in bovine mastitis.

It was observed that phages SA, SA2 and SANF demonstrated lytic activity against *S. saprophyticus*, *S. xylosus*, *S. sciuri*, *S. succinus*, and *M. caseolyticus*. Although the main characteristic of phages is their host specificity, some phages can infect different bacterial genera and species (BOHANNAN & LENSKI, 1997). Consequently, these phages should be tested in more isolates to confirm their possible host amplitude. Once, if there

is no resistance displayed by a specific bacterium, the phage does not need to find another host cell to infect, but if resistance is exhibited, the phage seeks diversification of the species host to continue its infectious cycle

The high efficiency of cell lysis by phages is based on the specificity of bacterial receptors and cell disintegration after a short viral life cycle, leading to the rapid elimination of the target cell (LECLERC et al., 2000; MATSUZAKI et al., 2005). In gram-positive bacteria, such as *S. aureus*, the receptors are located in the peptidoglycans (or mureins) and teichoic acids in the bacterial cell wall (SHAW & CHATTERJEE, 1971; KANEKO et al., 2009; XIA et al., 2011). Phages are bound to these compounds present in the cell wall to be adsorbed into the cell.

In this study, some phages (SA, SANF and SA2) were lytic in other *Staphylococcus* species and

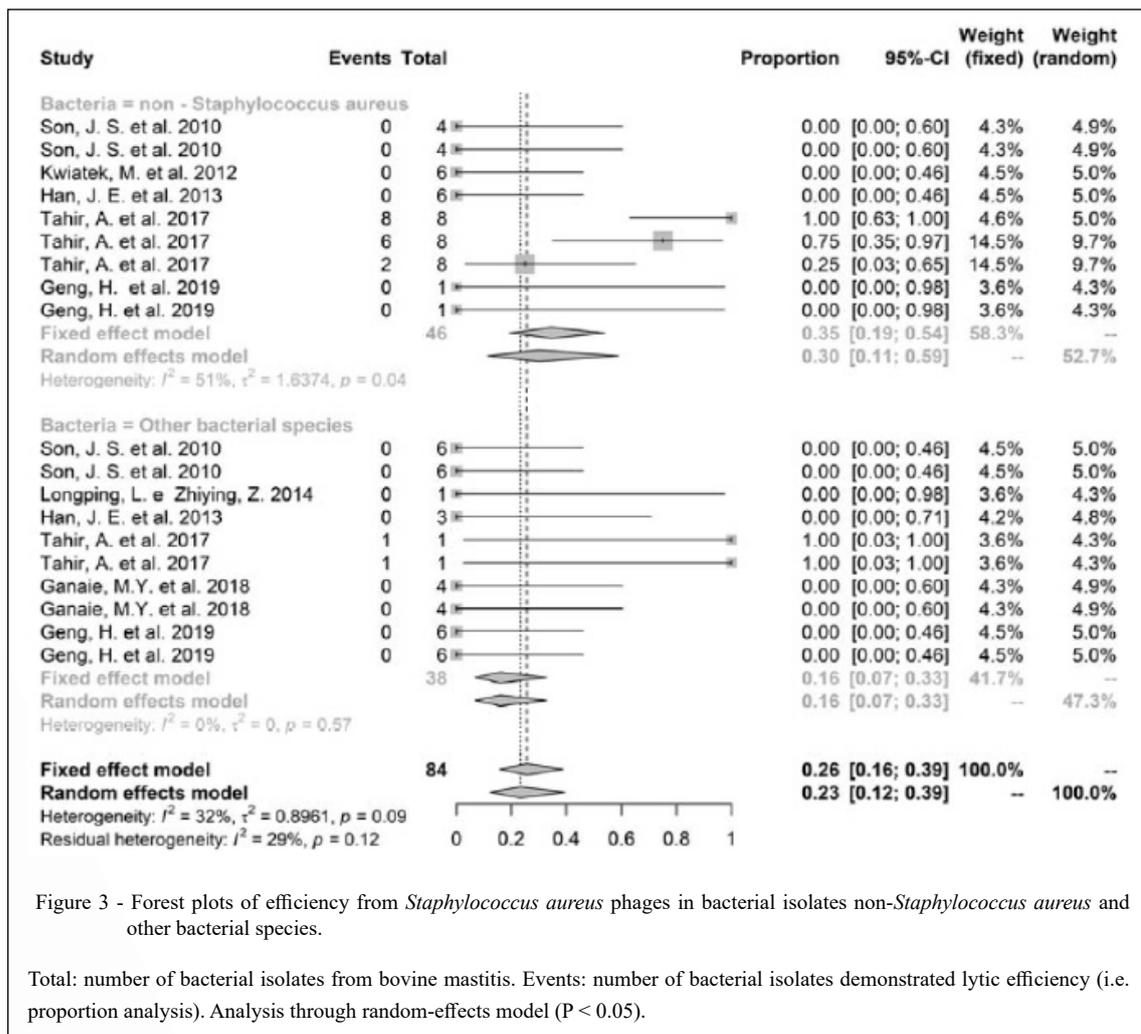


Figure 3 - Forest plots of efficiency from *Staphylococcus aureus* phages in bacterial isolates non-*Staphylococcus aureus* and other bacterial species.

Total: number of bacterial isolates from bovine mastitis. Events: number of bacterial isolates demonstrated lytic efficiency (i.e. proportion analysis). Analysis through random-effects model ($P < 0.05$).

Macrococcus caseolyticus. Importantly, the genus *Macrococcus* belongs to the same family as *S. aureus* and is also a Gram-positive bacterium. This suggests that these microorganisms share similar receptors for the phages.

The methodology used allowed greater precision in establishing the effects of the treatments, regulating data heterogeneity, and facilitating the verification of a greater number of results (LOVATTO et al., 2007). *In vitro* lytic activity tests of studies presented variability; thus, the eligibility criteria conferred the best homogeneity of samples, allowing comparisons and analyses of data.

Phage SA (HAMZA et al., 2016) was excluded from this meta-analysis because of a contradiction in the results of the phage lytic activity against *K. pneumoniae* isolates. In addition, studies using bacterial isolates from humans and other animals were also excluded, since the aim of the present study was to focus on phages used for the prevention and treatment of bovine mastitis.

Notably, there is a scarcity of studies evaluating *in vivo* lytic activity of *S. aureus* phages in cows (LERONDELLE & POUTREL, 1980; GILL et al., 2006a) and mice (ALDOORI et al., 2015; BREYNE et al., 2017; IWANO et al. et al., 2018; GENG et al., 2019). However, we opted to exclude these studies to homogenize the data for this meta-analysis.

Phage therapy against mastitis presents many important challenges. Some of the problems encountered are phage stability, inhibitory effects on the cow's immune system and certain thermolabile proteins are present in raw milk that affect phage-cell interaction (O'FLAHERTY et al., 2005; GILL et al., 2006b; TANJI et al., 2015; BARI et al., 2017).

Another issue is the possibility of bacteria developing phage resistance. However, the use of a cocktail containing multiple phages could be indicated to circumvent this phenomenon, since the bacterium must develop simultaneous resistance mechanisms against multiple phages. This phenomenon is less likely to occur when compared to a single phage used to lysis a particular bacterium (OECHSLIN, 2018). A limitation for the use of phages for mastitis therapy would be the need to perform laboratory diagnosis and testing the susceptibility of the agent to phages. However, this fact is also a current limitation for antimicrobial use.

The increase in multi-resistant bacteria, including *S. aureus*, the principal microorganism involved in the etiology of bovine mastitis, has necessitated the search for alternative treatments for this disease (FERNÁNDEZ et al., 2018). Thus, the use of phages may be recommended in cases where

mastitis is caused by antimicrobial-resistant bacteria (SULAKVELIDZE; ZEMPHIRA; JR, 2001), since phages infect and eliminate bacteria through mechanisms distinct from those of antimicrobials (MONK et al., 2010).

CONCLUSION

This meta-analysis describes the *in vitro* infectivity of *S. aureus* phages against bacterial cells, demonstrating the lytic potential of these viruses. Thus, this study evaluates the efficiency of *S. aureus* phages in the control of bovine mastitis owing to their lytic activity, particularly against *S. aureus* isolates, including MRSA.

In addition, we suggest that among the evaluated phages, those with high lytic efficiency could be selected to produce a cocktail to be tested *in vivo* for the treatment and/or prevention of mastitis caused by *S. aureus*. Other investigations using phages as an alternative treatment method, should be performed to ensure the health of the dairy cattle using environmentally sustainable technologies, thereby preserving the efficacy of antimicrobials.

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DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflict of interest. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results.

AUTHORS' CONTRIBUTIONS

All authors contributed equally for the conception and writing of the manuscript. All authors critically revised the manuscript and approved of the final version.

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