icfBulletin
Indagine scientifica ed informazione veterinaria
Results

Biofilms

Dr Sue Paterson MA VetMB DVD DipECVD FRCVS,
RCVS and European Specialist Veterinary Dermatology.
Virtual Vet Derms
virtualvetderms.com

icf Bulletin
Indagine scientifica ed informazione veterinaria
Results

Biofilms

Dr Sue Paterson MA VetMB DVD DipECVD FRCVS,
RCVS and European Specialist Veterinary Dermatology.
Virtual Vet Derms virtualvetderms.com

Introduction

Bacteria exist in two forms. The first is as solitary free floating "planktonic" organisms most commonly seen in acute infections. The second and most predominant form of microbial growth is in biofilms (Costerton et al., 1978). Biofilms can be defined as a microbially derived sessile community characterised by cells that are irreversibly attached to a surface or interface or to each other; are embedded in a matrix of extracellular polymeric substances that they have produced, and exhibit an altered phenotype with respect to growth rate and gene transcription (Donlan and Costerton, 2002). Biofilms are everywhere and can form on virtually any body surface. They have been recognised in man in chronic sinusitis, chronic otitis media, chronic tonsillitis, dental plaque, chronic laryngitis, endocarditis, lung infections, biliary and urinary tract infections, osteomyelitis and chronic wounds (Bjarnsholt, 2013). Biofilms are now recognised as being clinically important in veterinary medicine and should be suspected in any chronic disease process (Gardner, 2011).

Not all bacterial species produce biofilm and the level of biofilm production can alter between different species. Bacteria found in canine diseases that have been shown to have biofilm producing properties include *Staphylococcus pseudintermedius* (Singh et al., 2013, Osland et al., 2012, Casagrande Proietti et al., 2015, Walker et al., 2016), *Pseudomonas aeruginosa* (Pye et al., 2013) and *Escherichia coli* (Nam et al., 2013, Oliveira et al., 2014, Vijay et al., 2015, Shimizu and Harada, 2017). Work by Han (2015) has shown that more than 90% of the isolates of both meticillin sensitive and resistant *Staphylococcus pseudintermedius* from healthy dogs are capable of producing biofilms (Han et al., 2015). A study by Pye (2013) showed that 40% of the isolates of *Pseudomonas aeruginosa* isolated from clinical cases of canine otitis externa were found to be capable of producing biofilms. In this study the biofilm minimum inhibitory concentrations (MICs) of *Pseudomonas spp.* for polymyxin B, neomycin, gentamicin and enrofloxacin were significantly higher than for the planktonic form, demonstrating the increased resistance of biofilm organisms at sites where they cause clinical disease (Pye et al., 2013). *Malassezia pachydermatis* has been shown to be capable of producing biofilms (Figueredo et al., 2012) particularly from cases of canine seborrheic dermatitis (Bumroongthai et al., 2016).

Diagnosis of biofilm infections

In human medicine establishing that a biofilm infection is present can be challenging. A range of criteria can be useful these include, a history of a condition that predisposes to the development of biofilm formation such as an orthopaedic implant. The presence of cytological findings consistent with biofilm formation such as microbial aggregates. A culture revealing microbes known to be associated with biofilm formation; recurrence of infection despite appropriate antimicrobial therapy or therapeutic failure despite appropriate antibacterial therapy based on culture and susceptibility (Rowson and Townsend, 2016). Medics have access to more specialised diagnostic techniques including molecular methods such as polymerase chain reactions and

Both of these techniques have been shown to have a higher degree of sensitivity than culture (Hall- Stoodley et al., 2006) but are not widely available or economically viable for veterinary clinicians. Veterinarians though can use the other diagnostic criteria to help make a diagnosis.

1) A history of a condition that predisposes to the development of biofilm formation
2) Clinical signs consistent with a biofilm infection
3) The presence of cytological findings consistent with biofilm formation
4) Therapeutic failure either due to lack of response or recurrence of infection despite appropriate therapy

A history of a condition that predisposes to the development of biofilm formation

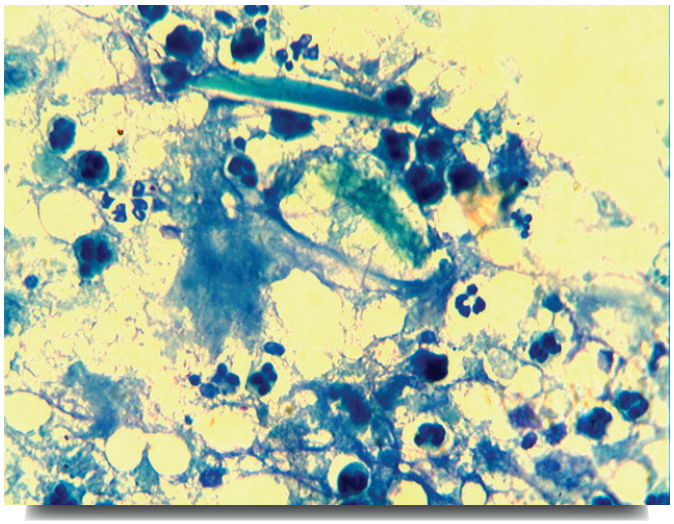
Almost any chronic disease process can lead to the formation of a biofilm infection. In dogs and cats biofilm infections have been associated with urine tract disease (Shimizu and Harada, 2017) , gastrointestinal disease (Silva et al., 2014, Reis et al., 2014), periodontal disease (Holcombe et al., 2014, Oliveira et al., 2016), otitis (Pye et al., 2013), dermatitis (Casagrande Proietti et al., 2015, Bumroongthai et al., 2016), chronic wounds (Swanson et al., 2014, Bayne, 2014) and implant infections (Thompson et al., 2011, Gallagher and Mertens, 2012, Savicky et al., 2013, Nicoll et al., 2014).

Clinical signs consistent with a biofilm infection

Where biofilm infection is present the micro-organisms within the biofilm are embedded in a self- produced matrix of extracellular polymeric substance (EPS). This appears as a thick mucoid discharge (see George otitis case below).

The presence of cytological findings consistent with biofilm formation

Cytology samples of discharge from an infection may be taken with a sterile swab. The swab should be gently rolled across a clean microscope slide, heat fixed and then stained with a Romanowsky stain such as Diff Quik. Microscopic examination will reveal signs of aggregates of infectious organisms often co-localised with inflammatory cells. This differs to the more distinct isolates found in cases where planktonic infection is present. Typically numbers of micro-organisms are sparse and found within the self-produced EPS matrix which usually takes up stain and appears as fine lacy background material (figure 1).



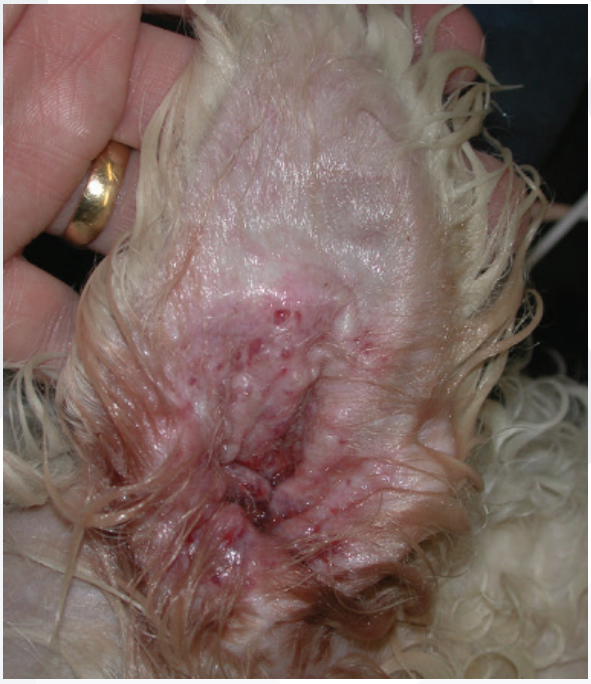
(figure 1)

Results

Therapeutic failure due to lack of response or recurrence of infection despite appropriate therapy

Typical cases will fail to respond to antibiotic therapy that has been based on appropriate culture and susceptibility testing. Often the same pathogen is identified on numerous occasions with an unchanged susceptibility pattern (see figure 2 below). In other cases the infection appears to have resolved on antibiotic therapy but relapses once drugs are discontinued.

George is a 6 year old Clumber spaniel with a chronic history of otitis externa. His condition has shown a partial response to topical antibiotic therapy but has relapsed on cessation of treatment on several occasions. On this most recent occasion he has had a topical ear drop prescribed which contains marbofloxacin. This has been prescribed on the basis that *Pseudomonas spp.* has been cultured with a good sensitivity to marbofloxacin.



After 3 weeks of treatment his owner returned to complain the dog is no better. Repeat cultures grow *Pseudomonas spp.* again with the susceptibility below.

Susceptibility	Antibiotic
Framycetin	R
Gentamicin	R
Polymyxin	S
Enrofloxacin	S
Marbofloxacin	S
Fusidic acid	R
Florfenicol	R

The lack of response to therapy, the repeat cultures revealing the same pathogen with the same susceptibility pattern is consistent with a biofilm infection

(figure 2)

Therapy of biofilm infections

Bacteria growing within a biofilm have the capacity to withstand and persist in the presence of higher levels of antibiotics than free living planktonic bacteria – this property is called recalcitrance (Rowson and Townsend, 2016). In clinical terms this means that biofilm bacteria can be from 100 to 1000 fold more resistant to antibiotics than their planktonic counterparts (Hoiby et al., 2010). Biofilm bacteria have several mechanisms that facilitate their resistance this includes restricted penetration of antibiotics; restricted growth at low oxygen tension; expression of biofilm specific genes and the presence of persisters (Ciofu et al., 2017). A range of different treatment options may need to be explored to effectively manage biofilm infections. Mechanical removal of the biofilm is the most effective method, however this is not always feasible to do. As well as the traditional use of antimicrobial therapy other more novel anti-biofilm strategies have been explored in both human and veterinary medicine. Often combinations of therapy that include antimicrobial drugs with other anti-biofilm agents prove the most successful. EDTA tris has long been recognised as a drug with benefits in treating chronic Gram negative infections. One of the newest drugs which has been found to have excellent activity against biofilms is N- acetyl cysteine.

EDTA tris

Ethylenediaminetetraacetic acid (EDTA) has been demonstrated to have antibacterial activity when combined with tromethamine (Tris). Studies have shown:

1) Damages the cell walls of planktonic bacteria
2) Synergistic with some antibiotics
3) Has low ototoxic potential
4) Use adjunct therapy for biofilm infection

EDTA tris has been shown to have the ability to **damage planktonic bacterial cell walls** to increase antimicrobial penetration (Wooley and Jones, 1983, Farca et al., 1997, Buckley et al., 2013). It is well tolerated and demonstrates **low ototoxicity** (Paterson, 2018). It has been shown to have **additive effects with a wide range of antibiotics** including gentamicin and fluoroquinolones (Buckley et al., 2013) as well as silver sulphadiazine and chlorhexidine (Guardabassi et al., 2010). More recently, *in vitro* work has shown that EDTA tris may be a **useful adjunctive treatment** for chronic cases of *Pseudomonas* otitis where biofilms have developed, if gentamicin or neomycin is to be used as a topical treatment (Pye et al., 2014). EDTA tris is probably best employed because of its ability to potentiate antibiotics, as the initial pre-treatment flush for acute (planktonic) Gram negative infections.

N-acetyl cysteine

N acetyl cysteine is a mucolytic drug that has been shown to have a wide range of additional properties. Studies have shown :

1) Antibacterial properties
2) Synergy with some antibiotics
3) Decreased biofilm formation
4) Reduced production of EPS
5) Promotion of mature biofilm disruption

N-acetyl cysteine (NAC) has **antibacterial properties** against a range of different pathogens. *Pseudomonas aeruginosa* appears to be particularly susceptible with *in vitro* antibacterial activity being recorded at minimum inhibitory concentrations (MIC) as low as 2- 20 ug/ml (Parry and Neu, 1977). A more recent study by Zhao (2010) has shown the MIC of NAC for *Pseudomonas aeruginosa* isolates from respiratory human respiratory infections to be 10 – 40mg/ml. Work by May (2016) has demonstrated that NAC has an MIC of 5 – 20mg/ml for common otic isolates *Staphylococcus pseudintermedius*, *Pseudomonas aeruginosa*, *Corynebacterium spp.* and *Beta haemolytic Streptococcus* (May et al., 2016). **Synergism** has been demonstrated with some of the third

Results

generations antibiotics such as carbenicillin and ticarcillin (Roberts and Cole, 1981) and although these antibiotics are restricted for veterinary use this early study demonstrated the potential for NAC's ability to enhance antibiotic activity. NAC has also been shown *in vitro* to be synergistic with ciprofloxacin (El-Feky et al., 2009) suggesting that a fluoroquinolone NAC combination may be useful in cases of *Pseudomonas spp.* infection. NAC **decreases biofilm formation** by a variety of bacteria notably *Staphylococcus epidermidis* (Perez-Giraldo et al., 1997); *E. coli* (Marchese et al., 2003) and *Pseudomonas aeruginosa* (Zhao and Liu, 2010). It appears to have the ability to reduce adherence and also reduce the **production of EPS**. The production of EPS by biofilm bacteria acts as a physical barrier to the penetrations of drugs which play a significant part in the increased resistance to antibiotic therapy. Work by Zhao (2010) has shown that EPS production by *Pseudomonas aeruginosa* is significantly decreased in the presence of NAC. EPS production was decreased *in vitro* by 27% and 45% in the presence of 0.5mg/ml and 1mg/ml NAC (Zhao and Liu, 2010). NAC also has a direct effect to **promotes the disruption** of mature biofilms and reduce sessile cell viability within the biofilm (Olofsson et al., 2003).

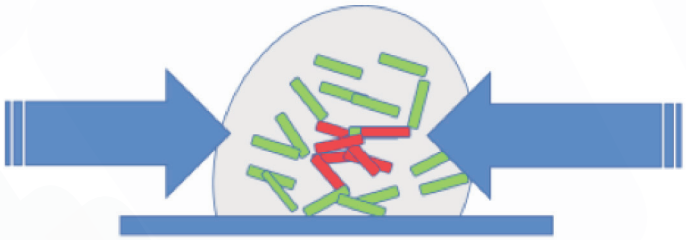
Human studies have shown that NAC is highly effective used with ciprofloxacin as a licensed otic (Ciprodex otic) to treat refractory otitis cases. In one *in vivo* study, Ciprodex otic was used with 0.5 or 2.0% NAC. Seven subjects with an average of 18.4 months of otorrhea despite therapy were included. Cessation of otorrhea was achieved in 6 of 7 subjects within 4 weeks of treatment. No subjects demonstrated ototoxicity via pre-treatment and post-treatment audiometry (Choe et al., 2007). A second *in vitro* study using the ciprofloxacin showed that where *P. aeroginosa* strains were resistant to ciprofloxacin, pre-treatment with NAC overcame the resistance leading to resolution of infection (Lea et al., 2014).

Veterinary use of NAC would seem based on human experiences to be suited to use with fluoroquinolones, selected on the basis of culture and sensitivity, in chronic otitis cases where first line antibiotics are ineffective. NAC also shows promise as an antibacterial agent for use in chronic infections in its own right (May et al., 2016) and may be useful with other topical drugs where biofilms are present, due to its vitro ability to reduce the production of EPS and reduce bacterial attachment, as a pre-treatment flush prior to antimicrobial therapy. A suitable protocol for use in *Pseudomonas spp.* otitis externa is outlined below.

POSSIBLE MODE OF ACTION OF BIOFILM BUSTING DRUGS

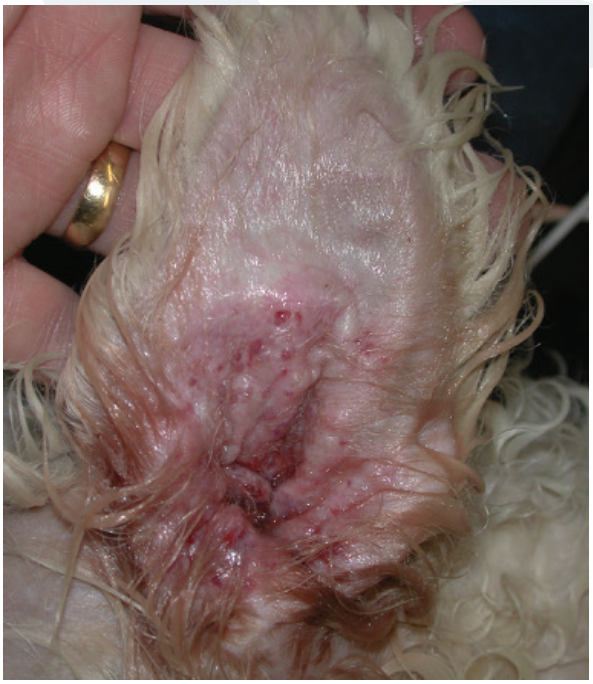
NAC

Antibacterial properties
Reduced production of EPS
Promotion of mature biofilm disruption



Tris EDTA

Direct damage to bacteria cell wall to increase microbial penetration
May affect bacterial efflux pump



Approach to George's otitis. George's clinical history, response to therapy and repeat culture results are consist with the presence of a biofilm infection. The combination of NAC as a pre-treatment flush is useful before application of antibiotics.

Step 1 Clean the ear thoroughly to remove as much of the discharge as possible. This is probably best achieved under heavy sedation or ideally under a full general anaesthetic. Appropriate analgesia is important to keep the dog comfortable during the procedure and post flush.

Step 2 Flood the ear canal with a solution of N-acetyl cysteine (Tris-NAC®). This should remain in situ for a minimum of 5 minutes.

Step 3 Remove as much of the excessive liquid as possible using suction or absorbing the fluid onto a cotton wool pad. Instil 0.5ml of the marbofloxacin based ear drop into the ear. The dog may be sent home with an ear cleaning solution (Otodine®) to clean the ear effectively before application of the NAC solution (Tris-NAC®). This should remain in place for 5 mins, before excessive fluid is removed using a cotton wool pad, before applying the antibiotic solution.

Conclusion

Bacterial biofilms are a major cause of infection in man and animals. They contribute to a wide range of veterinary diseases including urinary tract infections, periodontal disease, wound infection and otitis. Conventional antimicrobial therapy often fails to eliminate biofilm infection and new novel therapies to tackle these infections are currently being developed. N-acetyl cysteine appears to be a useful novel therapeutic agent to help in the management of biofilm infections.

References

BAYNE, D. 2014. Biofilm-infected wounds. J Am Vet Med Assoc, 244, 1126.

BJARNSHOLT, T. 2013. The role of bacterial biofilms in chronic infections. APMIS Suppl, 1-51.

BUCKLEY, L. M., MCEWAN, N. A. & NUTTALL, T. 2013. Tris-EDTA significantly enhances antibiotic efficacy against multidrug-resistant Pseudomonas aeruginosa in vitro. Vet Dermatol, 24, 519-e122.

BUMROONGTHAI, K., CHETANACHAN, P., NIYOMTHAM, W., YURAYART, C. & PRAPASARAKUL, N. 2016. Biofilm production and antifungal susceptibility of co-cultured Malassezia pachydermatis and Candida parapsilosis isolated from canine seborrheic dermatitis. Med Mycol, 54, 544-9.

CASAGRANDE PROIETTI, P., STEFANETTI, V., HYATT, D. R., MARENZONI, M. L., CAPOMACCIO, S., COLETTI, M., BIETTA, A., FRANCIOSINI, M. P. & PASSAMONTI, F. 2015. Phenotypic and genotypic characterization of canine pyoderma isolates of Staphylococcus pseudintermedius for biofilm formation. J Vet Med Sci, 77, 945-51.

CHOE, W. T., MURRAY, M. T., STIDHAM, K. R. & ROBERSON, J. B. 2007. N-Acetylcysteine as an adjunct for refractory ear infections. Otol Neurotol, 28, 1022-5.

CIOFU, O., ROJO-MOLINERO, E., MACIA, M. D. & OLIVER, A. 2017. Antibiotic treatment of biofilm infections. APMIS, 125, 304-319.

COSTERTON, J. W., GEESEY, G. G. & CHENG, G. K. 1978. How bacteria stick. Sci. Am. , 238, 86-95.

DONLAN, R. M. & COSTERTON, J. W. 2002. Biofilms: Survival mechanisms of clinically relevant microorganisms. Clin Microbiol Rev, 15, 167-193.

EL-FEKY, M. A., EL-REHEWY, M. S., HASSAN, M. A., ABOLELLA, H. A., ABD EL-BAKY, R. M. & GAD, G. F. 2009. Effect of ciprofloxacin and N-acetylcysteine on bacterial adherence and biofilm formation on ureteral stent surfaces. Pol J Microbiol, 58, 261-7.

FARCA, A. M., PIROMALLI, G., MAFFEI, F. & RE, G. 1997. Potentiating effect of EDTA-Tris on the activity of antibiotics against resistant bacteria associated with otitis, dermatitis and cystitis. J Small Anim Pract, 38, 243-5.

FIGUEREDO, L. A., CAFARCHIA, C., DESANTIS, S. & OTRANTO, D. 2012. Biofilm formation of Malassezia pachydermatis from dogs. Vet Microbiol, 160, 126-31.

GALLAGHER, A. D. & MERTENS, W. D. 2012. Implant removal rate from infection after tibial plateau leveling osteotomy in dogs. Vet Surg, 41, 705-11.

GARDNER, A. J. 2011. Biofilms and role to infection and disease in veterinary Medicine. In: PERCIVAL, S. L., KNOTTENBELT, D. C. & COCHRANE, C. A. (eds.) Biofilms and Veterinary Medicine Springer Berlin Heidelberg.

GUARDABASSI, L., GHIBAUDO, G. & DAMBORG, P. 2010. In vitro antimicrobial activity of a commercial ear antiseptic containing chlorhexidine and Tris-EDTA. Vet Dermatol, 21, 282-6.

HALL- STOODLEY, L., HU, F. Z. & GIESEKE, A. 2006. Direct detection of bacterial biofilms on the middle ear mucosa of children with chronic otitis media. JAMA, 296, 202-211.

HAN, J. I., YANG, C. H. & PARK, H. M. 2015. Emergence of biofilm-producing Staphylococcal pseudintermedius isolated from healthy dogs in South Korea. Vet Q, 35, 207-10.

HOIBY, N., BJARNSHOLT, T., GIVSKOV, M., MOLIN, S. & CIOFU, O. 2010. Antibiotic resistance of bacterial biofilms. Int J Antimicrob Agents, 35, 322-32.

HOLCOMBE, L. J., PATEL, N., COLYER, A., DEUSCH, O., O'FLYNN, C. & HARRIS, S. 2014. Early canine plaque biofilms: characterization of key bacterial interactions involved in initial colonization of enamel. PLoS One, 9, e113744.

LEA, J., CONLIN, A. E., SEKIROV, I., RESTELLI, V., AYAKAR, K. G., TURNBULL, L., DOYLE, P., NOBLE, M., RENNIE, R., SCHREIBER, W. E. & WESTERBERG, B. D. 2014. In vitro efficacy of N-acetylcysteine on bacteria associated with chronic suppurative otitis media. J Otolaryngol Head Neck Surg, 43, 20.

MARCHESE, A., BOZZOLASCO, M., GUALCO, L., DEBBIA, E. A., SCHITO, G. C. & SCHITO, A. M. 2003. Effect of fosfomycin alone and in combination with N-acetylcysteine on E. coli biofilms. Int J Antimicrob Agents, 22 Suppl 2, 95-100.

MAY, E. R., CONKLIN, K. A. & BEMIS, D. A. 2016. Antibacterial effect of N-acetylcysteine on common canine otitis externa isolates. Vet Dermatol, 27, 188-e47.

NAM, E. H., KO, S., CHAE, J. S. & HWANG, C. Y. 2013. Characterization and zoonotic potential of uropathogenic Escherichia coli isolated from dogs. J Microbiol Biotechnol, 23, 422-9.
NICOLL, C., SINGH, A. & WEESE, J. S. 2014. Economic impact of tibial plateau leveling osteotomy surgical site infection in dogs. Vet Surg, 43, 899-902.
OLIVEIRA, M., DIAS, F. R. & POMBA, C. 2014. Biofilm and fluoroquinolone resistance of canine Escherichia coli uropathogenic isolates. BMC Res Notes, 7, 499.
OLIVEIRA, M., TAVARES, M., GOMES, D., TOURET, T., SAO BRAZ, B., TAVARES, L. & SEMEDO-LEMSADDEK, T. 2016. Virulence traits and antibiotic resistance among enterococci isolated from dogs with periodontal disease. Comp Immunol Microbiol Infect Dis, 46, 27-31.
OLOFSSON, A. C., HERMANSSON, M. & ELWING, H. 2003. N-acetyl-L-cysteine affects growth, extracellular polysaccharide production, and bacterial biofilm formation on solid surfaces. Appl Environ Microbiol, 69, 4814-22.
OSLAND, A. M., VESTBY, L. K., FANUELSEN, H., SLETTEMEAS, J. S. & SUNDE, M. 2012. Clonal diversity and biofilm-forming ability of methicillin-resistant Staphylococcus pseudintermedius. J Antimicrob Chemother, 67, 841-8.
PARRY, M. F. & NEU, H. C. 1977. Effect of N-acetylcysteine on antibiotic activity and bacterial growth in vitro. J Clin Microbiol, 5, 58-61.
PATERSON, S. 2018. Brainstem auditory evoked responses in 37 dogs with otitis media before and after topical therapy. J Small Anim Pract, 59, 10-15.
PEREZ-GIRALDO, C., RODRIGUEZ-BENITO, A., MORAN, F. J., HURTADO, C., BLANCO, M. T. & GOMEZ-GARCIA, A. C. 1997. Influence of N-acetylcysteine on the formation of biofilm by Staphylococcus epidermidis. J Antimicrob Chemother, 39, 643-6.
PYE, C. C., SINGH, A. & WEESE, J. S. 2014. Evaluation of the impact of tromethamine edetate disodium dihydrate on antimicrobial susceptibility of Pseudomonas aeruginosa in biofilm in vitro. Vet Dermatol, 25, 120-3, e33-4.
PYE, C. C., YU, A. A. & WEESE, J. S. 2013. Evaluation of biofilm production by Pseudomonas aeruginosa from canine ears and the impact of biofilm on antimicrobial susceptibility in vitro. Vet Dermatol, 24, 446-9, e98-9.
REIS, A. C., SILVA, J. O., LARANJEIRA, B. J., PINHEIRO, A. Q. & CARVALHO, C. B. 2014. Virulence factors and biofilm production by isolates of Bacteroides fragilis recovered from dog intestinal tracts. Braz J Microbiol, 45, 647-50.
ROBERTS, D. & COLE, P. 1981. N-acetylcysteine potentiates the anti-pseudomonas activity of carbenicillin in vitro. J Infect, 3, 353-9.
ROWSON, C. & TOWNSEND, R. 2016. Biofilms: prevention and treatment. Bristish Journal of Hospital Medicine 77, 699-703.
SAVICKY, R., BEALE, B., MURTAUGH, R., SWIDERSKI-HAZLETT, J. & UNIS, M. 2013. Outcome following removal of TPLO implants with surgical site infection. Vet Comp Orthop Traumatol, 26, 260-5.
SHIMIZU, T. & HARADA, K. 2017. Determination of minimum biofilm eradication concentrations of orbifloxacin for canine bacterial uropathogens over different treatment periods. Microbiol Immunol, 61, 17-22.
SILVA, J. O., MARTINS REIS, A. C., QUESADA-GOMEZ, C., PINHEIRO, A. Q., FREIRE, R. S., ORIA, R. B. & DE CARVALHO, C. B. 2014. In vitro effect of antibiotics on biofilm formation by Bacteroides fragilis group strains isolated from intestinal microbiota of dogs and their antimicrobial susceptibility. Anaerobe, 28, 24-8.
SINGH, A., WALKER, M., ROUSSEAU, J. & WEESE, J. S. 2013. Characterization of the biofilm forming ability of Staphylococcus pseudintermedius from dogs. BMC Vet Res, 9, 93.
SWANSON, E. A., FREEMAN, L. J., SELEEM, M. N. & SNYDER, P. W. 2014. Biofilm-infected wounds in a dog. J Am Vet Med Assoc, 244, 699-707.
THOMPSON, A. M., BERGH, M. S., WANG, C. & WELLS, K. 2011. Tibial plateau levelling osteotomy implant removal: a retrospective analysis of 129 cases. Vet Comp Orthop Traumatol, 24, 450-6.
VIJAY, D., DHAKA, P., VERGIS, J., NEGI, M., MOHAN, V., KUMAR, M., DOIJAD, S., POHARKAR, K., KUMAR, A., MALIK, S. S., BARBUDDHE, S. B. & RAWOOL, D. B. 2015. Characterization and biofilm forming ability of diarrhoeagenic enteroaggregative Escherichia coli isolates recovered from human infants and young animals. Comp Immunol Microbiol Infect Dis, 38, 21-31.
WALKER, M., SINGH, A., NAZARALI, A., GIBSON, T. W., ROUSSEAU, J. & WEESE, J. S. 2016. Evaluation of the Impact of Methicillin-Resistant Staphylococcus pseudintermedius Biofilm Formation on Antimicrobial Susceptibility. Vet Surg, 45, 968-971.
WOOLEY, R. E. & JONES, M. S. 1983. Action of EDTA-Tris and antimicrobial agent combinations on selected pathogenic bacteria. Vet Microbiol, 8, 271-80.
ZHAO, T. & LIU, Y. 2010. N-acetylcysteine inhibit biofilms produced by Pseudomonas aeruginosa. BMC Microbiol, 10, 140.

Tris-NAC®


Tris-NAC®
Soluzione sterile
Sterile solution
TAMPONE TRIS-EDTA
+ N-Acetil Cisteina (NAC)
ad elevata forza ionica
per cani e gatti
TRIS-EDTA BUFFER
+ N-Acetylcysteine (NAC)
with high ionic strength
for dogs and cats
120 ml
Evolution of Knowledge

Notes:

DPE0131

Evolution of Knowledge

EFFICACY • QUALITY • SAFETY
One Two Passions

THE LONG – TERM PARTNER of ESVD/ECVD

www.icfpet.com • customercare@icfsrl.it