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(1:64–1:256), MRSP 1:128 (MICs 1:64–1:256; MBCs 1:64–1:128), EC 1:128 (1:64–1:128), ESBL-EC 1:128 (MICs 1:64–1:256; MBCs 1:32–1:256), PA 1:64 (1:32–1:512), and MP 1:64 (MICs 1:64–1:256; MBCs 1:32–1:64). There was no association of MIC or MBC with resistance to antimicrobial classes (Spearman's rank correlation $P = 0.95$ and $P = 0.37$) or individual drugs (multiple logistic regression $P = 0.83$ and $P = 0.74$). Confluent growth was seen with all the hair samples. 0.011% hypochlorous acid shows *in vitro* bactericidal efficacy, although some PA, ESBL-EC and MP isolates are less susceptible. There was no residual activity on hair.

Source of funding: Self-funded.
Conflict of interest: None declared.

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Evaluation of the *in vitro* synergistic activity of chlorhexidine and trizEDTA

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This study assessed the *in vitro* synergistic activity of chlorhexidine and trizEDTA against meticillin-susceptible *S. pseudintermedius* (MSSP), meticillin-resistant *S. pseudintermedius* (MRSP), *Escherichia coli* (EC), extended spectrum beta-lactamase *E. coli* (ESBL-EC) and *Pseudomonas aeruginosa* (PA). Ten isolates of each organism were incubated for 10 min with doubling dilutions of chlorhexidine (10–0.02 μg/mL) and trizEDTA (from 35.6/9.4 to 0.28/0.07 mg/mL; PA 71.2/18.8 to 0.56/0.14 mg/mL) in 96-well plates in a chequerboard design. The isolates were then incubated in tryptone soy broth overnight. For each plate the fractional inhibition concentrations (FIC) were calculated for each well along the growth/no growth interface: FIC = (MIC chlorhexidine/MIC chlorhexidine in combination) + (MIC trizEDTA alone/MIC trizEDTA in combination); and FIC index (FICi) for each isolate = mean of the FICs. These values were used to calculate the mean FICi for each organism. The results were evaluated using the convention: FICi <1.0 = synergy; 1.0–4.0 = additive; and >4.0 = antagonism. The highest concentration of the trizEDTA reduced the MIC of the chlorhexidine by 3–6 dilutions, but with lower concentrations the MICs and MBCs reverted to those seen with chlorhexidine alone for most isolates. The mean (SD) FICi were: MSSP 1.2 (0.1), MRSP 1.2 (0.1), EC 1.0 (0.2), ESBL-EC 1.1 (0.4) and PA 0.9 (0.2). Chlorhexidine and trizEDTA show additive behaviour against MSSP, MRSP, EC and ESBL-EC, with weak synergy against PA. TrizEDTA therefore appears to potentiate the activity of chlorhexidine against these organisms, but only at high concentrations and true synergy is unlikely.

Source of funding: Self-funded.
Conflict of interest: None declared.

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Overexpression of TLR-2 and TLR-4 mRNA in feline polymorphonuclear neutrophils exposed to *Microsporum canis*

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Feline polymorphonuclear neutrophils (PMN) were recently shown to produce pro-inflammatory cytokines upon exposure to *Microsporum canis*. The recognition of fungal structures (pathogen associated molecular patterns, PAMP) by pattern recognition receptors (PRR) is the starting point of PMN activation. The toll-like receptors (TLR) and the C-type lectin receptors (CLR) are the two main PRR in phagocytic cells that recognize fungal components. Their role in PMN has never been investigated in dermatophytoses but can be assumed since TLR-2, TLR-4 and dectin-1 of PMN are especially involved in the recognition of fungal PAMP in many mycoses. The aim of this study was to evaluate the expression of TLR-2, TLR-4 and dectin-1 mRNA in feline PMN exposed to different components from *M. canis*. PMN isolated from healthy cats were stimulated for 2 h with either live arthrospores, structural (heat-killed arthrospores) or secreted components from *M. canis*. Lipopolysaccharide (LPS) and PBS were used as positive and negative controls respectively. The levels of PRR mRNA in stimulated PMN were quantified by qRT-PCR and expressed relative to that in non-stimulated PMN (negative controls) in three independent experiments. A significant increase of TLR-2 and TLR-4 mRNA levels in feline PMN stimulated with live and heat-killed arthrospores was observed, and was comparable to that obtained with the LPS. No significant variation in dectin-1 mRNA expression was observed in PMN exposed to the tested fungal components. The results suggest that TLR-2 and TLR-4 in PMN are involved in the feline immune response against *M. canis* through the recognition of fungal PAMP on arthrospores.

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