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Evaluation of Lacticin 3147 to reduce in vitro ruminal methane production (Untersuchungen zur Methanreduktion von Lacticin 3147 in vitro)

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Bacteriocins are potent additives for manipulating ruminal fermentation into desirable ways, including reducing enteric methane emissions from ruminant animals. In this study lacticin 3147, a bacteriocin produced by *Lactococcus lactis* subsp. *lactis* DPC3147, a strain isolated from an Irish kefir grain (1), was evaluated for its potential to reduce methane production from an *in vitro* rumen fermentation system.

Methods: Lacticin 3147 was obtained from Dairy Products Research Centre, Teagasc, Moorepark, Republic of Ireland. A factorial completely randomized design was used to examine the effect of different levels of lacticin 3147 (0, 10, 25 and 50 µM) added to hay and hay:concentrate (1:1, w/w) substrates on gas and methane productions after 24 h incubation period. Approximately 380 mg of the substrates were incubated with 30 ml buffered medium containing rumen liquor in the *in vitro* Hohenheim gas production method (2). The lacticin was prepared by solubilizing in sodium phosphate buffer pH 6.8, and was injected (≤1 ml) into 100 ml calibrated syringe from the syringe nozzle before dispensing buffered rumen liquor. Data obtained were analysed using factorial analysis of variance (ANOVA), followed by Tukey's (HSD, honest significant difference) test to compare different treatments. All statistical analyses were performed using STATISTICA software version 6.0.

Results: Lacticin 3147, up to 50 µM, did not decrease net methane production per unit net gas production when added to hay or hay:concentrate diets. Similarly absolute gas and methane productions (in ml) did not decrease. Substitution of hay by concentrate (50% on dry matter basis) increased significantly ($P<0.05$) gas and methane production, both as net methane and ml methane per 100 ml net gas.

Other bacteriocins such as nisin and bovicin HC5 have been found to be effective in reducing methane production (3, 4). Nisin decreased methane production by 36% when added at 10 µM in mixed ruminal bacteria *in vitro*. Bovicin HC5 inhibited methane production by approx. 35 % and 50% when added at 128 activity units (AU)/ml (8.4 µM) and 500 activity units (AU)/ml (32.8 µM). Lacticin 3147 was not effective at 50 µM suggesting that lacticin is not as effective as nisin and bovicin HC5. Fast degradation of lacticin by rumen proteases could be one of the reasons for this.

Table 1. Effect of lacticin 3147 addition on *in vitro* gas and methane productions

Substrate	Hay				Hay:concentrate			
Lacticin (µM)	0	10	25	50	0	10	25	50
Gas (ml)	77.2^{a}	77.3^{a}	75.0^{a}	77.8^{a}	90.8^{c}	90.0^{bc}	91.8^{c}	87.5^{b}
CH_4 (ml)	13.2^{ab}	13.2^{ab}	12.7^{a}	13.5^{b}	16.9^{d}	16.7^{cd}	17.0^{d}	16.2^{c}
CH_4 (ml/100ml)	17.1^{a}	17.1^{a}	17.0^{a}	17.3^{a}	18.6^{b}	18.5^{b}	18.5^{b}	18.5^{b}

Conclusions: Lacticin 3147 is not effective in reducing methane production up to a concentration of 50 µM.

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