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"Bridging the gap between increasing knowledge and decreasing resources"

Effect of Nitrate Supplemented to Diets Differing in Concentrate to Forage Ratio on Methane Production *in vitro*

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Abstract

Dietary nitrate has been gaining attention due to its persistent methane mitigation and the objective of this study was to evaluate the effects of encapsulated nitrate as soybean meal replacer in diets contrasting in concentrate:forage ratio on ruminal fermentation and methane production by using the *in vitro* semi-automatic gas production technique. This study was conducted at Laboratory of Animal Nutrition, CENA/USP, in Piracicaba, São Paulo, Brazil. A randomised complete block design in a 2×4 factorial arrangement with two types of diet (20:80 and 80:20 concentrate:forage ratio) and three levels of encapsulated nitrate inclusion (0, 1.5, 3.0, and 4.5 % in dietary DM) was applied. There were four different inoculums, each inoculum considered as a block. Encapsulated nitrate replaced soybean meal to achieve isonitrogenous diets (13 % CP). After 24 h of incubation, there was a diet $\times$ nitrate interaction ($p < 0.05$) for methane production (mL/g OM degraded and mL/g NDF degraded). Diets with 20 % concentrate reduced ($p < 0.01$) the gas production and methane by 58 % and 62 %, respectively, when compared to the contrasting diets. There was a linear reduction in CH_4 production with addition of encapsulated nitrate to both diets, reaching a reduction of 60 % when comparing the contrasting diets with 4.5 % and 0 % of encapsulated nitrate. Encapsulated nitrate didn't affect degradability of organic matter, so that increasing the nitrate content did not affect the availability of the substrate for fermentation. The replacement of soybean by encapsulated nitrate in protein equivalent was able to reduce the CH_4 emission, particularly in diets with 20 % concentrate without affecting the degradability of organic matter.

Keywords: Gas production, greenhouse gas, methanogens, rumen fermentation

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