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# Acclimation of continuous biomethanation process to extremely high ammonia levels

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## **Abstract**

Anaerobic digestion (AD) is an effective technology to recover energy (i.e. biogas) from biomass. However, when protein-rich bioproducts are used as feedstocks, could lead to low methane yields and even to complete process failure. The reason is that high ammonia levels from protein degradation could inhibit the microbes mediating the AD process. Acclimatized methanogenic communities to high ammonia levels can offer a solution to this problem. Nevertheless, the acclimation of ammonia tolerant methanogenic communities for continuous AD of protein-rich substrates still poses serious problems (e.g. washout effect). Thus, the present study investigates the acclimation of methanogenic communities during AD of protein-rich bioproducts. Microalgae *Chlorella vulgaris* (>50% protein in dry matter), was co-digested with cattle manure (80/20 VS microalgae/VS manure) in two identical continuous stirred tank reactors (CSTR) under mesophilic conditions ( $37\pm 1^\circ\text{C}$ ). The reactors had a hydraulic retention time of 23 days and organic loading rate of  $1.95\pm 0.1 \text{ g VS L}^{-1}\text{d}^{-1}$ . Acclimation strategy was followed a stepwise increasing ( $0.5 \text{ g NH}_4^+\text{-N L}^{-1}$  each step) the total ammonia (TAN) concentration starting from 4 up to  $10 \text{ g NH}_4^+\text{-N L}^{-1}$  using  $\text{NH}_4\text{Cl}$ . During the baseline period ( $4 \text{ g NH}_4^+\text{-N L}^{-1}$ ) the methane yield was  $335 \text{ NmL CH}_4 \text{ g}^{-1} \text{ VS}$ . Throughout the acclimation period, the methane yields of the CSTR reactors were stable with an average variation less than 10% compared to the yield at baseline period. Additionally, pH decreased from 8.4 to 7.8 during the ammonia acclimation process. Owing to the pH drop, the free ammonia (FAN) concentration was kept in high but stable levels ( $920\pm 150 \text{ mg NH}_3\text{-N L}^{-1}$ ), even though the TAN increased 1.5-fold. This indicates that only TAN increase was not enough to cause inhibition and that FAN plays the most crucial role in the ammonia inhibitory mechanism. The present study showed that it is possible to acclimatize methanogenic communities to extremely high TAN levels. Finally, the results derived from this study could offer a practical approach to digest protein-rich bioproducts.

**Keywords:**

Adaptation; Ammonia inhibition; Anaerobic digestion; CSTR.