

## Stabilizing food waste anaerobic digestion

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The increasing production of food waste (FW) worldwide and the recent application of international regulations call for the development of novel strategies for the valorization of this biomass. Among the different options, anaerobic digestion (AD) is a promising alternative that offers an efficient waste treatment, together with its valorization in the form of biogas and digestate (Capson-Tojo et al., 2016). However, AD of FW is a complicated process, usually associated with accumulation of ammonia nitrogen (TAN) and volatile fatty acids (VFAs). These issues have frequently led to low methane yields and process kinetics, and even process failure (Banks et al., 2008; Zhang et al., 2015). Industrially, efficient AD is achieved by substrate dilution, a practice that has clear environmental and economic limitations. Therefore, it is critical to develop novel AD strategies to avoid the accumulation of VFAs, favoring their consumption and thus stabilizing the process.

In this study, three different strategies for stabilization of FW AD have been first compared using consecutive batch pilot reactors during about 5 months: (i) working at low temperatures (30 °C vs 37 °C in the other conditions, aiming to reduce the free ammonia nitrogen (FAN) proportions), (ii) co-digestion of FW with paper waste (PW) (to dilute the nitrogen input, providing at the same time buffering capacity) and (iii) supplementation of trace elements (TEs) (to favor VFA consumption by promoting the synthesis of enzymes). A control reactor simply fed with FW was also run. Since all these strategies conducted to a strong accumulation of VFAs and in particular propionic acid (HPr), batch essays were run to investigate the consumption of the accumulated VFA. Digestate from the co-digestion reactor were sampled and used to test different options: (i) addition of the TEs concentration defined previously for the pilot reactor (corresponding to 100 mg Fe l<sup>-1</sup>), (ii) a reactor with a 5-folded TEs concentration (corresponding to 500 mg Fe l<sup>-1</sup>), (iii) supplementation of granular activated carbon (GAC) to favor adsorption of inhibitors and direct interspecies electron transfer (DIET), avoiding the formation of electron shuttles (such as hydrogen or formate) and favoring acetic acid (HAc) consumption (Dang et al., 2016; Lee et al., 2016), (iv) ½ dilution to evaluate the effect on VFA consumption aiming to reduce thermodynamic inhibitions. A Control reactor was also defined to perform an un-biased evaluation of the effect of these different options. The reactors used were specifically designed to allow sampling of the digesting medium during the AD process without disturbing the gas in the headspace (Motte et al., 2015).

Considering first the three different strategies for stabilization, the obtained results proved that FW can be efficiently valorized via AD, obtaining high methane yields, of around 500 ml CH<sub>4</sub>·g VS<sup>-1</sup> (300 ml CH<sub>4</sub>·g VS<sup>-1</sup> in the co-digestion reactor due to the lower degradability of PW) and high volumetric productivities, of 16 l CH<sub>4</sub>·l reactor<sup>-1</sup>. However, all the conditions showed the same complication after two or three feeding cycles: propionic acid (HPr) accumulated progressively in the reactors, decreasing the kinetics of methane production, the pH values and eventually leading to reactor acidification at the moment of reloading, thus limiting the organic load applied. Nevertheless, although similar behaviors were observed in the four conditions tested, the methane production kinetics, as well as that of HPr accumulation, were totally different in the reactors. The reactor working at 30 °C showed much slower methane production kinetics when compared to the other reactors (*i.e.* batch duration of 40 days vs. 30-40 days in the control reactor) due to its lower temperature. In addition, although lower concentrations of FAN were observed, HPr still accumulated in the reactor, jeopardizing the AD process. Regarding the reactor co-digesting FW and PW, this substrate efficiently diluted the TAN concentrations in the reactor (*i.e.* 6-7 g TAN·l<sup>-1</sup> vs. 9-10 g TAN·l<sup>-1</sup> in the control reactor). However, this TAN reduction did not result in a less intense accumulation of HPr. On the contrary, this reactor showed the worst performance in terms of VFA accumulation (*i.e.* over 20 g·l<sup>-1</sup> of HPr at the end of the operational period vs. 8 g·l<sup>-1</sup> in the control reactor). This could be explained by the slower degradability of PW when compared to FW, which caused higher residual amounts of acetic acid present in the reactor, which resulted in thermodynamic inhibition of the oxidation of propionate. Finally, the supplementation of TEs clearly improved the AD kinetics, allowing higher substrate loads and leading to higher methane production rates. In addition, although TEs addition could not avoid HPr build-up, the extent of accumulation of this VFA was lower in the TEs-doped reactor when compared to the control system, suggesting that dosing TEs favored HPr oxidation. This experiment allowed producing methane efficiently in successive batch reactors, identifying at the same time a main issue occurring during FW AD: HPr accumulation. Moreover,

working at low temperatures and co-digestion with PW were discarded as effective options for process stabilization in large scale applications (lower productivities and HPr accumulation).

The influence of the addition of TEs and GAC on the VFAs consumption was tested using digestate from the co-digestion reactor (concentrations of HAc and HPr of 6.41 and 21.0 g l<sup>-1</sup>, respectively). Whatever the operational conditions, three successive phases could be identified. During the first phase, corresponding to about the first 28 days, methane production occurred because of the degradation of HAc and readily available residual substrates but HPr was not consumed in any condition. At the end of this phase, the HAc concentration was approximately of 0.4 g l<sup>-1</sup>. In the case of undiluted reactors, a “stable phase” occurred during which the concentrations of HPr and HAc did not vary and no significant production of methane was observed (2<sup>nd</sup> phase). After about 52 days, HPr started to be degraded in those conditions (3<sup>rd</sup> phase). Simultaneously, a slight increase in the HAc concentrations was observed since HPr was degraded into HAc and methane was also produced. During this last phase, even if the global behavior of the undiluted reactors (Control, addition of TES, addition of GAC) was similar, slight differences can be highlighted. The addition of TEs (5xTEs) improved slightly the kinetics of HPr degradation when compared to the Control reactor, probably by favoring the synthesis of formate dehydrogenase (Banks et al., 2012). Interestingly, syntrophic propionate oxidation (SPO) was clearly improved when adding GAC. The results indicate that the most plausible explanation is the occurrence of DIET in the reactors. Other than DIET, the steeper slope of HPr degradation in the GAC reactor also suggests that the growth of HPr oxidizing bacteria during the exponential phase was promoted, probably through biofilm formation onto the GAC surface, allowing syntrophic interactions to occur. Considering the diluted reactor (1/2 Dilution), SPO started earlier, right after the first phase. This can be explained by the lower concentration of electron shuttles (hydrogen and/or formate) due to the addition of water. Although this alternative is widely applied in industrial AD of solid waste for substrate pretreatment before AD, it leads to greater reactor volumes, and higher amounts of digestate. In these experiments, the total consumption of HPr took between 66 and 114 days, confirming that the degradation of this VFA is clearly a problem after its build-up.

Further research must be carried out to elucidate if the process performance can be improved by allowing the growth of HPr oxidizers, achieving eventually a stable HPr-degrading community. Promising options such as GAC addition and TEs optimization have the potential of improving greatly the performance of FW AD (*i.e.* improving the biogas productivities and reducing the retention times).

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