Towards understanding the role of product inhibition on acidogenic fermentation yield and profile

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Introduction

The application of circular economy is gaining attention due to population growth, increase in waste production and the depletion of natural sources. A transition in waste management schemes is needed where waste is conceived as a source of energy, nutrients and chemical products (Puyol et al., 2017). Fermentation stands as a key biotechnology in future resource recovery facilities (RRF). In fermentation, organic waste is converted into easily assimilable organic compounds like volatile fatty acids (VFA), alcohols and lactic acid. Subsequently, these compounds can be transformed into high value-added products.

In most acidogenic fermentation publications working at pH near 5-6, a stable concentration of the final products has been observed (Fernández-Domínguez et al., 2020; Garcia-Aguirre et al., 2019; Vidal-Antich et al., 2021). The product concentration and profile remained relatively constant at around 5 $gCOD_{acetic}/L$, 6 $gCOD_{propionic}/L$ and 9 $gCOD_{butyric}/L$ (Fernández-Domínguez et al., 2020; Vidal-Antich et al., 2021). These results indicate that product inhibition may control the acidogenic fermentation yield and profile.

The objective of this study was to investigate the product inhibition in acidogenic co-fermentation using waste activated sludge (WAS) as the main substrate and dog food (DF) and cellulose (CEL) as co-substrates. To assess product inhibition, three co-fermentation tests were carried out with different initial concentration of acetic, propionic, and butyric acid.

Material and methods

WAS was collected from a wastewater treatment plant from the Barcelona metropolitan area (Spain). Microcrystalline CEL or DF were used as co-substrate.

Anaerobic co-fermentation batch assays were performed in 250 mL serum bottles at 35 ºC. No inoculum was added. Co-fermentation batch was monitored by withdrawing a sample (4 mL) using a needle and a syringe. VFA, alcohols, pH, and soluble chemical oxygen demand (sCOD) were measured at each sampling event. VFA concentrations were rescaled (min-max normalization range [0, 1]) for further data analysis.

Three set of co-fermentation experiments were performed in this research, always using the same proportion between substrates 70:30 in %VS (WAS-CEL or WAS-DF). Experiment 1 aimed to understand the impact of product inhibition in VFA profile and concentration. Different initial acetic, propionic, and butyric acid concentrations were spiked to the co-fermentation assays. Experiment 2 was done to determine the gradual effect of increasing the concentration of each acid. Different concentrations of each acid (acetic, propionic, and butyric) were added based on the 50, 75 and 100% of the maximum concentration determined in Experiment 1. Experiment 3 was carried out to validate the response observed in Experiment 1 and 2. In Experiment 3, only the effect of adding acetic and propionic acid were tested. All experiments included a positive control (i.e., co-fermentation assay without acid addition).

Results and discussion

The results presented in Fig. 1 are from Experiment 1 using DF as co-substrate. Fig. 1A shows the control, whereas Fig. 1B and 1C show the results from an initial acetic acid concentration of 5 gCOD/L (ADF), while Fig. 1D and 1E show the results from an initial butyric acid concentration of 9 gCOD/L (BDF). Fig.1C and 1E represent the linear regression between the rescaled acid concentration of the condition with the rescaled acid concentration of the positive control, where a slope greater than 1 represents no-inhibition.

Fig. 1B shows that an initial concentration of acetic acid favoured butyric acid production. Fig. 1C shows that there was no inhibition of acetic acid on valeric and caproic acids. However, it slightly inhibited propionic acid, as shown by the minor slope decrease in Fig. 1C and the slight decrease in the final propionic acid concentration compared to the positive control (2.4 gCOD/L vs 2.7 gCOD/L, respectively). In contrast, acetic acid favoured the accumulation of butyric acid, as shown by the slope > 1 in Fig. 1C, as well as the higher butyric acid concentration compared to the control (7.1 g COD/L vs 5.5 gCOD/L, respectively). Fig. 1D and 1E show that butyric acid inhibited acetic and propionic acid production. The slope of these acids in Fig. 1E were lower than 1, corresponding

to lower concentrations (3.5 and 2.3 gCOD/L, acetic and propionic acid, respectively) when compared to the positive control (4.8 and 2.7 gCOD/L, acetic and propionic acid respectively). In the butyric acid experiment, the concentrations of valeric and caproic acid increased (3.8 and 4.7 gCOD/L, respectively) compared to the positive control (3.4 and 2.2 gCOD/L, respectively).

Experiment 2 and 3, allowed to evaluate the gradual effect of the acetic, propionic, and butyric concentration. The results obtained were reproducible with Experiment 1. The addition of acetic acid showed that propionic acid production was inhibited, while butyric acid concentration increased compared to the control. Propionic acid addition inhibited the production of acetic acid and butyric acid while promoting valeric acid accumulation. Finally, butyric acid addition in Experiment 2, showed that the inhibition of acetic and propionic acid and the increase in valeric and caproic acids was proportional to the initial butyric acid concentration.

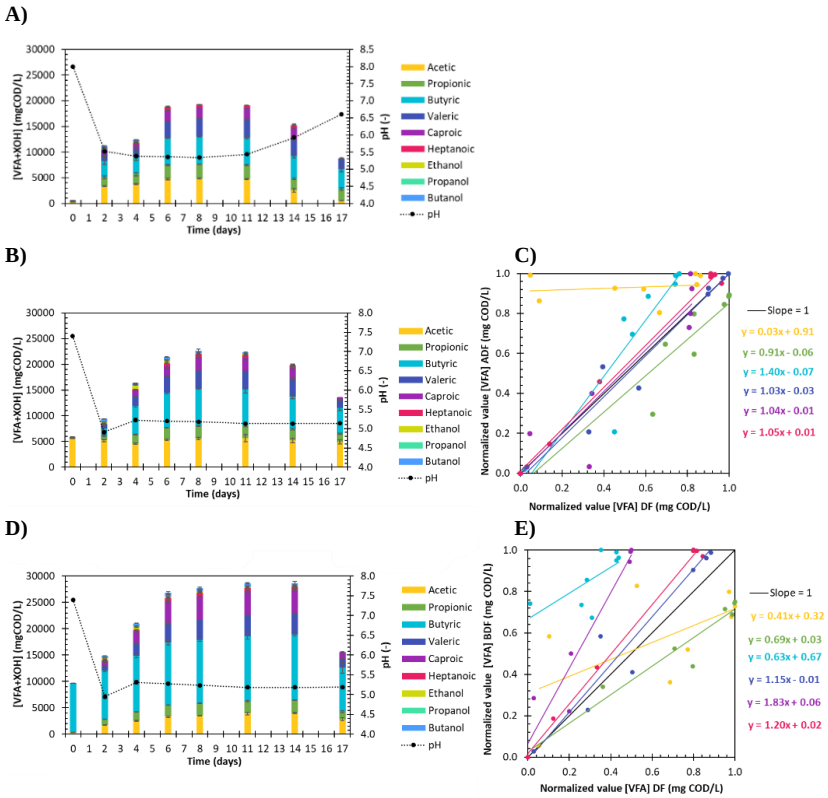

Fig. 1. A) Evolution of VFA+XOH in control in the experiment 1 A) Evolution of VFA+XOH of the condition ADF in the experiment 1B) Comparative concentrations of ADF respect to the control C) Evolution of VFA+XOH of the condition of BDF in the experiment 1. D) Comparative BDF concentrations respect to the control

Conclusion

Results revealed that the production of volatile fatty acids with a lower number of carbons than the added acids were inhibited (e.g., butyric acid inhibited the production of acetic and propionic acids), while the longer chain acids were favoured. Acetic acid spike inhibited the production of propionic acid and lead to an increase in the butyric acid yield. Propionic acid spike inhibited the production of acetic acid, whereas the valeric acid was favoured. As for butyric acid spike, although the acetic and propionic acids productions were inhibited, the caproic and valeric production were favoured.

Reference

Fernández-Domínguez, D., Astals, S., Peces, M., Frison, N., Bolzonella, D., Mata-Alvarez, J., & Dosta, J. (2020). Volatile fatty acids production from biowaste at mechanical-biological treatment plants: Focusing on fermentation temperature. *Bioresource Technology*, *314*, 123729.

Garcia-Aguirre, J., Esteban-Gutiérrez, M., Irizar, I., González-Mtnez de Goñi, J., & Aymerich, E. (2019). Continuous acidogenic fermentation: Narrowing the gap between laboratory testing and industrial application. *Bioresource Technology*, *282*, 407–416.

Puyol, D., Batstone, D. J., Hülsen, T., Astals, S., Peces, M., & Krömer, J. O. (2017). Resource Recovery from Wastewater by Biological Technologies: Opportunities, Challenges, and Prospects. *Frontiers in Microbiology*, *7*, 2106.

Vidal-Antich, C., Perez-Esteban, N., Astals, S., Peces, M., Mata-Alvarez, J., & Dosta, J. (2021). Assessing the potential of waste activated sludge and food waste co-fermentation for carboxylic acids production. *Science of The Total Environment*, *757*, 143763.