Effect of salinity on the production of biogas from the macroalgae *Caulerpa prolifera*

R. García-Montero[1]; M. Cortijo-Ruiz[1]; C. García-Delgado[2]; M. Rodríguez-Rastrero[3]; S. López-Rayo[1]; N. Carreras[3]

[1]Department of Agricultural Chemistry and Food Science, Faculty of Sciences, University Autónoma of Madrid, 28049-Madrid, Spain
[2]Department of Geology and Geochemistry, Faculty of Sciences, University Autónoma of Madrid, 28049-Madrid, Spain
[3]Soil Conservation and Remediation Unit. Department of Environment. CIEMAT, Avenida Complutense 40, 28040-Madrid, Spain
Presenting author email: carlos.garciadelgado@uam.es

Keywords: Mar Menor lagoon; beach-cast seaweeds, biomethane; anaerobic digestion.

The Mar Menor lagoon, located on the Mediterranean coast of the Region of Murcia (SE Spain), is an extensive coastal saltwater lagoon (170 km^2 with 73 km coastline) with shallow depth (3.6 m average and 7 m maximum), artificially connected with the Mediterranean Sea at several points (IEO, 2020). Spanish Law 19/2022 grants the Mar Menor and its basin rights to protection, conservation, maintenance and, where appropriate, restoration, by governments and riverside inhabitants. This includes waters, biological communities, as well as terrestrial and aquatic subsystems that are part of the lagoon and its basin. However, since the 1980s, the Mar Menor has received large polluting flows from agricultural activity in the Campo de Cartagena area (IEO, 2020). The resulting eutrophication (Rybarczyk, 1996) is associated with an overpopulation of micro and macroalgae, among which the green macroalga *Caulerpa prolifera* stands out. This species is favoured from the 70s by the strong imbalance in the lagoon ecosystem generated after the dredging and expansion of the Estacio channel, which connects the Mar Menor with the Mediterranean Sea. *Caulerpa prolifera* currently occupies the largest surface area of the Mar Menor seabed, causing large biomasses of beach-cast seaweeds.

Faced with the environmental challenge of minimizing and valorising these biomasses, the Algarikon project "Valuation of the algae accumulated on the coasts of the Mar Menor as a consequence of its eutrophication" was proposed. Algarikon Project is funded by the European Union within the Recovery, Transformation and Resilience Plan of the Spanish Ministry of Science and Innovation. In this sense, algae are an important research topic regarding their biofunctional and energetical properties as antioxidants, antibacterials, antifungals, biofuels, as well as biogas potential (Pradhan et al, 2022). Specifically, this work aims to determine the methanogenic potential of beach-cast seaweed biomasses, consisting, for the most part, of *Caulerpa prolifera*. A widely used AMPTS II® device based on a batch-type anaerobic digestion system will be used for this purpose. AMPTS is a universal platform for anaerobic biodegradability, biochemical methane potential (BMP) and specific methanogenic activity (SMA) testing protocols, allowing standardization of measurement procedures, data interpretation and reports. The VDI 4630 standard was used as a reference.

The conditions for collecting beach-cast seaweeds imply very high salinity, which requires considering the potentially inhibitory role of this factor in anaerobic digestion (Feijoo et al., 1995; Lefebvre et al., 2007; Zhang et al., 2017b; Pang et al., 2020). In this work, three salinity levels were tested for beach-cast seaweed substrates, using different washing intensities: a) *EC100*: seaweed in suspension with deionized water, until reaching an electrical conductivity in the washing water of approximately 100 µS cm^{-1}, b) *EC1000*: seaweed in tap water (about 80 µS cm^{-1}) until obtaining about 1,000 µS cm^{-1}, and c) EC25000: seaweed without cleaning treatment, whose average electrical conductivity was about 25,000 µS cm^{-1}. Previously, a study was carried out to evaluate the influence of mechanical pretreatment on the anaerobic digestion of *Caulerpa prolifera*, in order to obtain substrates with different active surfaces. Specifically, it involved crushing and subsequent sieving to sizes of <1, 1-2, and 2-4 mm. The test confirmed the absence of significant differences among particle sizes, so the least demanding preparation, 2-4 mm, was chosen for the BMP tests carried out.

The tests were carried out for 27 days, at 37ºC (mesophilic) and in triplicate. The inoculum used in the tests came from the digesters of a WWTP. Blanks (inoculum only) were set up to determine the production of endogenous biomethane. In order to determine the biomethane production by degraded SV, total solids (TS) and volatile solids (SV) were analysed both initially and at the end of the BMP tests. In addition to biogas production, pH, EC, TCOD, SCOD, NTK and NTA were analysed to assess the digestion process.

Figure 1 shows the daily Nml CH_4 production of the blank and the three selected salinity levels. The biogas kinetics for the three EC levels indicated biogas production from the first day of incubation, that is, without lag. EC100 produced the highest average biogas (341.4 ml CH_4), followed by EC1000 (317.8 ml CH_4) and EC25000 (275.2 ml CH_4). After the first 4 days of rapid increase in biogas production, the gradual depletion of biodegradable organic matter convertible into biogas led to a decrease in production per time unit. The test was terminated when the daily methane production was consistently less than 1% of the accumulated production.

Observed differences among treatments have not been considered statistically significant. The high variability in intra-treatment production must be also highlighted.

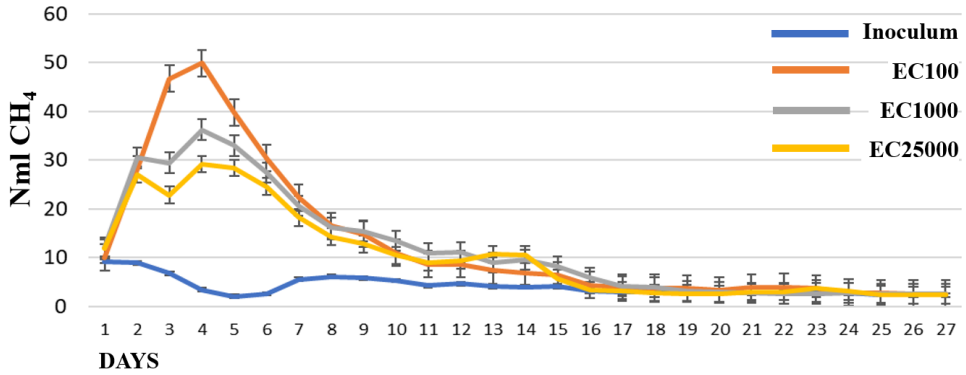

Figure 1: Daily methane production (Nml)

It should be noted that the specific methane production, that is, the production relative to the SV consumed, gave different results (Figure 2), corresponding to EC25000, algae without any previous treatment, the highest value (210.2 Nm^3 CH_4 gVS^{-1}).

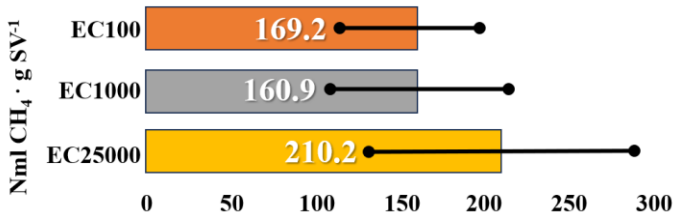

Figure 2: Specific methane production per treatments

This work constitutes one of the first assessments of the widely distributed and frequently invasive *Caulerpa prolifera*, as a substrate for biomethane production. The first results of this research suggest that beach-cast of *Caulerpa prolifera* is a suitable substrate for biogas production through anaerobic digestion, even when this substrate is not subjected to prior washing treatment. This fact can favour, in a context of large-scale extraction of this waste on the coastline of the Mar Menor, a significant saving of fresh water resources. The use of these biomasses to produce renewable energy can be integrated into the global solution to the serious problem of eutrophication of the Mar Menor and other similar scenarios.

References

- Feijoo, G.; Soto, M.; Méndez, R.; Lema, J.M. (1995). Sodium inhibition in the anaerobic digestion process: Antagonism and adaptation phenomena. *Enzyme and Microbial Technology* 17:18&188, 1995. DOI: 10.1016/0141-0229(94)00011-F
- IEO. *Informe de evolución y Estado actual del Mar Menor en relación al proceso de eutrofización y sus causas. Instituto Español de Oceanografía. Ministerio de Ciencia e Innovación. Gobierno de España*. (2020). Available in: https://www.miteco.gob.es/content/dam/miteco/es/prensa/informemarmenorjulio2020_tcm30-510566.pdf
- Lefebvre, O., Quentin, S., Torrijos, M., Godon, J. J., Delgenes, J. P., & Moletta, R. (2007). Impact of increasing NaCl concentrations on the performance and community composition of two anaerobic reactors. Applied microbiology and biotechnology, 75(1), 61-69. DOI: 10.1007/s00253-006-0799-2.
- Pang, H., Xin, X., He, J., Cui, B., Guo, D., Liu, S., ... & Nan, J. (2020). Effect of NaCl concentration on microbiological properties in NaCl assistant anaerobic fermentation: hydrolase activity and microbial community distribution. Frontiers in Microbiology, 2449. DOI: 10.3389/fmicb.2020.589222.
- Pradhan, B.; Bhuyan, P.P.; Patra, S.; Nayak, R.; Behera, P.K.; Behera, C.; Behera, A.K.; Ki, J.S.; Jena, M. Beneficial effects of seaweeds and seaweed-derived bioactive compounds: Current evidence and future prospective, Biocatalysis and Agricultural Biotechnology, Volume 39, 2022, 102242, DOI: 10.1016/j.bcab.2021.102242.
- Rybarczyk, H.; Elkaim, B.G.; Wilson, J.; Loquet, N. *L'eutrophisation en Baie de Somme: mortalités des peuplements benthiques par anoxie*, *Oceanol. Acta* 19. (1996) 131–140.
- Zhang, Y., Li, L., Kong, X., Zhen, F., Wang, Z., Sun, Y., ... & Lv, P. (2017). Inhibition effect of sodium concentrations on the anaerobic digestion performance of Sargassum species. Energy & Fuels, 31(7), 7101-7109. DOI: 10.1021/acs.energyfuels.7b00557.