PhD School on Agriculture, Environment and Bioenergy
(**http://sites.unimi.it/dottorato_aab/**)

(XXXVI cycle, 2020-22)

Project draft

1. **Field of interest**

*Indicare **il/i** settore/i scientifico disciplinari:*

AGR13: Bioeconomy, biomass recovery, circular economy.

2. Project title: Bio-electrorecycling of carbon dioxide into valuable products

3. Tutor Fabrizio Adani

- Eventually: co-tutor/s: Tommy Pepe Sciarria

4. Relevance of the topic and state of the art:

Nowadays the world is facing an environmental crisis because of the increase of CO2 and other greenhouse gas (GHG) emissions and the accumulation in the environment of non-biodegradable, fossil-fuel plastics. CO2 rise, due primarily to fossil fuel combustion (77% of the total emission), contributes to the global warning, that could lead to dramatic effects on earth's climate within the next decades.
The use of CO_2 as feedstock for chemicals and biobased products is gaining attention as demonstrated recently by the Bio-Based Industries Joint Undertaking (BBI JU), which aims to moving towards a biobased economy in Europe using domestic renewable raw materials to locally produce food, feed, chemicals, materials and fuels. Moreover, the use of CO_2 as feedstock is twice advantageous because it is greatly available and does not compete with food supply chain. Biological CO_2 sequestration methods using microorganisms as catalysts could lead to important advances compared to conventional CO_2 capture methods, which usually are considered expensive. In this sense, microbial electrosynthesis (MES) is based on the use of bio-electrochemical systems (BES), where the CO_2 is reduced into multi carbon organic compounds by microorganism, which are employed as biocatalysts in either the cathode or anode to achieve electricity-driven synthesis of a wide variety of compounds.

5. Layout of the project (draft):

Microbial Electrochemical Technologies (METs) connects microbiology and electrochemistry to take profit of microorganisms able to transfer electrons extracellularly (Schroder et al., 2015). This technological approach has allowed to investigate different derivatives applications such as electricity production, water electrolysis, nutrient recovery, electromethanogenesis, microbial electro-remediation, microbial electrosynthesis, which have the potential to improve conventional technologies in their fields (Pepè Sciarria et al., 2018; Blasco-Gómez et al., 2017; Pous et al., 2018). Microbial electrosynthesis (MES) from carbon dioxide (CO_2) is a promising approach to transform gaseous waste streams into commodity chemicals such as methane or volatile fatty acids by applying an external voltage to a bio.ectrochemical system where microorganism can work as biocatalysts in either the cathode or anode to achieve electricity-driven synthesis of a wide variety of compounds (Battle Vilanova et al., 2017). In this project, different Microbial electrosynthesis (MES) tests will be carried out in order to implement CO_2 recovery by the bio-electrocatalyst activity of Carboxydotrophic consortia. The project will be organized in order to achieve a CO_2 recovery platform where, at the beginning, the main objective will be the production of short chain carbon compounds such as CH_4 and Acetate. Once this first goal will be acquired, the project will be shift also, to the application of these carbon compounds into other biotechnology processes such as bioplastic production or single-cell protein (SCP) production.

5.1. Materials & Methods: ½ to 1 page

Carboxydotrophic mixed culture consortia will be selected starting from an anerobic sludge inoculum collected from a wastewater treatment plant. The first selection will be carried out in batch reactors fed with a gas mixture of H2 + CO2. At the end of the selection, the selected consortia will be inoculated into the bio-electrochemical system reactors. Single chamber and double chambers reactors (working volume 500ml) equipped with stainless steel and graphite brush electrodes will be used to perform the CO2 catalyst reduction. The CO2 will be purged into the systems according with the methodology reported in literature (Battle-Vilanova et al., 2017). In the same way, the external voltage will be applied according with the carbon compound production target of the experiment.

5.2. Schedule and major steps (3 years):

In particular, the project will be divided in 3 steps:

Step 1 (M1-M12): **Selection of bacteria consortia and production of short chain high value compounds**

During the first part of the project Hydrogenotrophic (H2 + CO_2), Methylotrophic (CH_4) consortia will be selected starting from an anerobic sludge inoculum. At the end of the selection, the selected consortia will be inoculated into the bio-electrochemical system reactors. The CO2 will be purged into cathode chamber and the BES will be operated with the biocathode poised at -800 mV vs SHE to drive the reduction of the CO_2 fraction into methane. The BES will be operated in batch mode to characterize methane production under continuous flow to demonstrate its viability at long-term

Step 2 (M13-24): **Production of short chain volatile fatty acids**

During the second part of the project the entire pool of reactors will be used in order to produce volatile fatty acids from the biocatalyst reduction of CO_2. In this case, the biocathode will poised at a potential of -0.8 V vs. SHE, using a potentiostat and chambers will be filled with mineral medium similar to ATCC1754 (Tanner et al., 1993), containing 2-bromoethanesulfonate to inhibit methanogenesis.

Step 3 (M25-36): **Application of high value compounds**

During this stage will study the implementation of a two-steps technological platform able to produce single-cell protein from resources harvested in waste streams. To this aim, a promising biotechnological platform, Microbial Electrochemical Technologies (METs), will be integrated into the protein production chain. Overall process will provide food by applying minimal energy (power-to-protein). CO_2, CH_4/H2 and NH^{4+} stream coming from operating BES will be recycled to feed Hydrogenotrophic (H_2 + CO_2), Methylotrophic (CH_4) consortia such as *Methylococcus*. However, this microbe is being cultivated with natural gas (Ritala et al., 2017), which is a limited resource with a strong demand from the energetic sector. In this case, the aim this step, carbon and nitrogen could be gathered from BES waste streams instead of syngas.

6. Available funds (source and amount): Fondi: Fabrizio Adani (> 100.000 €)

6. Literature: max 10 citazioni

Batlle-Vilanova P., R. Ganigue, S. Ramió-Pujol, L. Bañeras, G. Jiménez, M. Hidalgo, M. D. Balaguer, J. Colprim and S. Puig, Bioelectrochemistry, 2017, 117, 57.

Blasco-Gómez, R., Batlle-Vilanova, P., Villano, M., Balaguer, M.D., Colprim, J., Puig, S., 2017. On the Edge of Research and Technological Application: A Critical Review of Electromethanogenesis. Int. J. Mol. Sci. 1–32. DOI: 10.3390/ijms18040874.

Ganigue R., S. Puig, P. Batlle-Vilanova, M.D. Balaguer, J. Colprim, Microbial electrosynthesis of butyrate from carbon dioxide, Chem. Commun. 51 (2015) 3235–3238, http://dx.doi.org/10.1039/C4CC10121A.

Pepè Sciarria T., Batlle-Vilanova P., Colombo,B. Scaglia, B, Balaguer M. D., Colprim J.,. Puig * S and Adani F. Bio-electrorecycling of carbon dioxide into bioplastics Green Chem., 2018,20, 4058-4066

Pous, N., Balaguer, M.D., Colprim, J., Puig, S., 2018. Opportunities for groundwater microbial electro-remediation. Microb. Biotechnol. 11(1), 119-135. DOI: 10.1111/1751-7915.12866.

Ritala, A., Häkkinen, S.T., Toivari, M., Wiebe, M.G., 2017. Single cell protein-state-of-the-art, industrial landscape and patents 2001-2016. Front. Microbiol. 8(OCT), 2009. DOI: 10.3389/fmicb.2017.02009.

Schroder, U., Harnisch, F., Angenent, L.T., 2015. Microbial electrochemistry and technology: terminology and classification. Energy Environ. Sci. 8(2), 513-519. DOI:10.1039/C4EE03359K