

# Discrimination of the Olfactive Fraction of Different Renewable Organic Sources and their By-Products. A New Generation of MOX Sensor Tailor Made Device to Classify the Volatile Fingerprint

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Biogas is becoming one of the most used and profitable renewable sources. It is obtained through many different processes that frequently involve different green renewable sources. During the different steps of the process, it generates several by-products that could be reused as organic fertilizers. The aim of this work was to study the volatile fraction and determine the volatile fingerprint of 6 different organic samples involved in biogas production by the innovative Small Sensor System (S3) based on Semiconductor Metal Oxide (MOX) gas sensors. Obtained results show the volatile profile of each sample that at the same time support the sensor's device results. S3 result shows a perfect discrimination of the volatile fraction of the different studied matrices based on the different composition of their volatile set. matrix and shows how the sensors are able, in real time, to cluster and discriminate the fingerprint of these renewable sources. In the end, S3 results are very promising to enhance the traceability and the origin of the sources in the biogas industry at each specific stage of production, focusing on the possible release of off-flavors in the environment from different types of organic biomass and the reuse of their by-products supporting circular economy.

The aim of this work was to find and identify the VOCs set that characterizes different types of organic renewable sources through the use of the tailor made Small Sensor Systems S3 to distinguish between different odor sources based on their qualitative and quantitative differences in VOCs profile, in order to be able in the future to produce a user friendly fast, economic and with auto learning capabilities to support the aforementioned green industrial processes.

## 1. Introduction

Volatile organic compounds are organic chemicals that have a high vapor pressure at room temperature. High vapor pressure correlates with a low boiling point, a trait known as volatility. They play a remarkable and relevant role in agro-industrial processes, and food science and technology (Biasoli et al, 2011). VOCs are therefore able to uniquely characterize a matrix and its origin, thus becoming a true, unique and specific volatile fingerprint. Biogas is currently one of the biochemical transformation technologies available to produce electricity, heat and fuels. It is also the main bioenergy carrier that can be mainly produced with biomass from short-chain national biomass and that does not require import biomass (Molino et al, 2013). Anaerobic Digestion (AD) is a biological process that takes place naturally when bacteria break down organic matter in environments without oxygen. Generally, any type of organic material can be treated with anaerobic digestion. Some of them can be wastepaper, cardboard, grass clippings, leftover food, industrial effluents, sewage and animal waste (Long et al, 2019).

The main products of this AD process are classified in two major categories ([www.nextville.it/index/559](http://www.nextville.it/index/559). Accessed 23/08/2021): Biogas, the principal product which is formed by CH<sub>4</sub> (60%) and CO<sub>2</sub> (40%). In other words, it is formed by C, H, O. These are all elements that come from agri-cultural crops. This one can be purified in order to obtain biomethane (CH<sub>4</sub> 95%).

Digestate, a homogeneous material with a less humidity value due to the bacterial degradation of the dry matter and capable to hold climate-altering gas. The resultant organic substance is more stable, and it contains fertility elements (N, P, K) that can be really useful to the ground improving the future productivity. Biomethane is already in third place in Europe for energy powered by renewable sources with a total share of 17.41% of the total energy used at national level (Comotti et Bertagna, 2012). One of the most used sources of green and renewable sources for this kind of production is sewage. It is a liquid manure derived from solid and liquid dejections. Indeed, the sewage used for biogas production has a limit correlated with the emission of off flavor ([www.rinnovabili.it/mobilita/biometano-agricolo-filiera/](http://www.rinnovabili.it/mobilita/biometano-agricolo-filiera/)).

Among the energy crops utilized to the production of biogas, the ones most used are maize, sorghum, triticale. These are all plants belonging to the family of grasses, characterized by good crop yields, fast growth, adaptability to different types of soil and a high percentage of dry matter (Kobayashi et al, 2017). Starting from the limits imposed by the traditional technologies, and the use of these matrices, this study was developed in order to offer promising support to this sector for the protection of the territory thanks to the research and development of technological solutions for the reduction of the environmental impact (off-flavors), resulting from the use of traditional chemical fertilizers. In the last few years, this sector has experimented a significant growth since it combines tradition with technological innovation (Urriza-Arsuaga et al, 2019). To overcome some typical limits, such as time consumption and trained lab staff, approaches based on nanowire (Comini et al, 2002) and reotaxial growth thermal oxidation (RGTO) (Comini et al, 2009) (gas sensor technology could be engaged). Nanowire and RTGO gas sensors base their functional principle on the analysis and characterization of the volatile (VOCs) fingerprint of a determinate sample (Núñez Carmona et al, 2017). This kind of approach has already been applied successfully in many different fields such as human microbiota monitoring and environmental monitoring (Sberveglieri et al, 2015). This technology is based on semiconductor metal oxide gas sensors (MOX) [10-11], used with considerable success in several sectors, ranging from food safety (Concina et al, 2012), quality control (Sberveglieri et al, 2015) environmental monitoring and human health, thanks to its high sensitivity, fast responses, and low costs (Abbatangelo et al, 2020).

## 2. Materials and Methods

All the samples treated in this work came from organic renewable sources which involve different steps of the production chain of biomethane and its by-products. For all samples the sampling method was the same. For the sample preparations and for the headspace VOC's sampling method the Italian regulation UNI EN 13725:2004 (Standard number: UNI EN 13725: 2004) was used as reference, using collection sampling bags (Figure 1).



Figure 1. Example of the sampling collection bags with the gas samples used in this study.

A total number of 6 samples have been studied and are described as follows:

- Carbonate AGR: is obtained through the sludge purification. The obtained carbonate is a fertilizer and it is registered in the list of the fertilizers of the Italian legislation legislative decree D.Lgs. Governo n° 75 del 29/04/2010. The result is hygienic and free of pathogens such as *E. coli* and *Salmonella* spp. because of the chemical process of obtaining it.
- Digestate FI: is the digestate obtained through a thermophilic anaerobic digestion process that has as inlet waste deriving from the food production and consumption cycle, including purification sludge.
- Agricultural Digestate: it is a zootechnical digestate that is derived from an an-aerobic digestion plant of livestock slurry.
- Sewage AeS: is animal row manure. It is an untreated (i.e., not digested) livestock slurry deriving from a pig farm.
- Untreated Sludge: is composed by the untreated sewage sludge.
- Control AGR: the gas samples in this case are collected sampling the headspace of the sampling hood with no sample present on it. The main role of this sample was to create a sort of base line to differentiate with the VOCs present in the samples and those present in the sampling facilities.

## 2.1. Sampling points

### 2.1.1. Small Sensor System S3

The Small Sensor System (S3) is a device that has amply demonstrated the advantages of the application of this technology in recent years. The S3 instrument was developed by Nano Sensor System (NASYS) S.r.l. ([www.nasys.it](http://www.nasys.it)), an innovative start-up and spin-off of the University of Brescia. The innovative S3 device consists of an array, made of different semiconductor metal oxide gas sensors, flow, temperature and humidity sensors. In the system there is also a pump that transports inside the device volatile compounds from samples. For the management and control of signals there is a system that allows to store and analyze the data acquired in the cloud making S3 an IoT device (Abbatangelo et al, 2020). The mechanism of operation of metal oxide sensors is based on the variation of the electrical conductance of the sensing material, caused by interaction with the surrounding environment. The reaction between the oxygen species adsorbed on the surface of the sensitive element and the target molecules present in the gas samples causes a release of electrons which in turn modulate electrical properties, including electrical conductance (Núñez Carmona et al, 2009). S3 is composed of three essential parts:

1. Sensors chamber: The six MOX sensors are positioned into a steel chamber separated from the external environment, except for an inlet and an outlet path for the passage of volatile compounds. In addition to the MOX sensors, there are also a temperature, humidity, and a flow sensor which are fundamental to consider the number of features during the process. The dimensions of the chamber are 11 cm × 6.5 cm × 1.3 cm.
2. Fluid dynamic circuit for the distribution of volatile compounds: The fluid dynamic circuit is formed by a pump (Knf, model: NMP05B), polyurethane pipes, a solenoid valve, and a metal cylinder where there is an activated carbon for filtering any type of odors present outside of the instrument. The pump flow is set by a needle valve positioned at the chamber inlet.
3. Electronics control system: The electronic boards allow to register the resistance variations of the sensors, their correct heating, their operating temperature, and permit as well to send the registered data in real time to the dedicated Web App through an internet connection. In addition, it allows communication and synchronization with an autosampler.

A total number of 10 replicates of one minute each for all samples (Carbonate AGR, Control AGR, Digestate FI, Agricultural Digestate, Sewage AeS, Untreated Sludge) for a total of 60 replicas were carried out. The analysis was conducted inserting a “pipe” into the collection sample bags using the S3 pump operated at 100 sccm to transport the sample to the sensor chamber.

## 2.2. Data analysis

### 2.2.1. Small Sensor System S3

The data obtained from the S3 device were developed using the principal component analysis (PCA). This technique consists of clustering the sample variables, through linear combinations, that describe the link between one sample to the others, obtaining the principal components (PC), which are far fewer than the original variables (Granato et al, 2018). PCA allows to reveal all possible clusters of samples united by similar characteristics within the main components considered in the hyperplane.

Data analysis was performed using MATLAB® R2015a software (MathWorks, Natick, MA, USA). First, sensor responses in terms of resistance ( $\Omega$ ) were normalized when compared to the first value of the acquisition ( $R_0$ ). For all the sensors, the difference between the first value and the minimum value during the analysis time was calculated; hence,  $\Delta R/R_0$  has been extracted as featured. Principal Component Analysis (PCA) was applied to this data matrix to evaluate the ability of the system to clusterize the samples based on the different sensor responses to the different VOC's set of the different samples (Sato et al, 2018).

### 3. Results and Discussion

#### 3.1 Small Sensor System S3

The PCA in Figure 2 describes how the headspace of the 6 samples from organic sources are distributed well separated on the hyperplane. The first two Principal Components (PC) were used for a total explained variance (EV) equal to 99.2% (84.17% in PC1 and 15.03% in PC2). The sample distribution into the hyperplane explains how all the samples are well separated according PC1, which is the main axis and the one with the more explained variance (84,17%). In particular, two of them (Control and Carbonate) are equidistantly positioned and, as a consequence, completely different according sensor analysis. It is interesting since for the Carbonate sample is the one chemically treated with no microorganisms present and on the opposite site the control sample with just clean air and are effectively the most different samples of all studied samples regarding the volatile set compounds. As a consequence, the samples were clearly grouped according to their origin.

On the other hand, at the center of the hyperplane it is possible to observe that the other 4 samples are placed near the axis crossing point. Regarding PC1 axis the samples are in couples "Digestate FI" and "Sewage" on the negative part and "Sludge" and "Digestate AGR" on the positive part in an equidistant zone from the axis origin. It is possible to say that the presence of an active microflora in these samples has generated as indicated in the table present in Appendix 1 many different compounds coming from the transformation of the organic matter by the microorganisms (Sberveglieri et al, 2015)

Furthermore, PC2 provide an interesting insight since thanks to this axis it is possible to separate the non-treated samples as "Sludge" and "Sewage" to the others that have undergone chemical or microbial transformations.

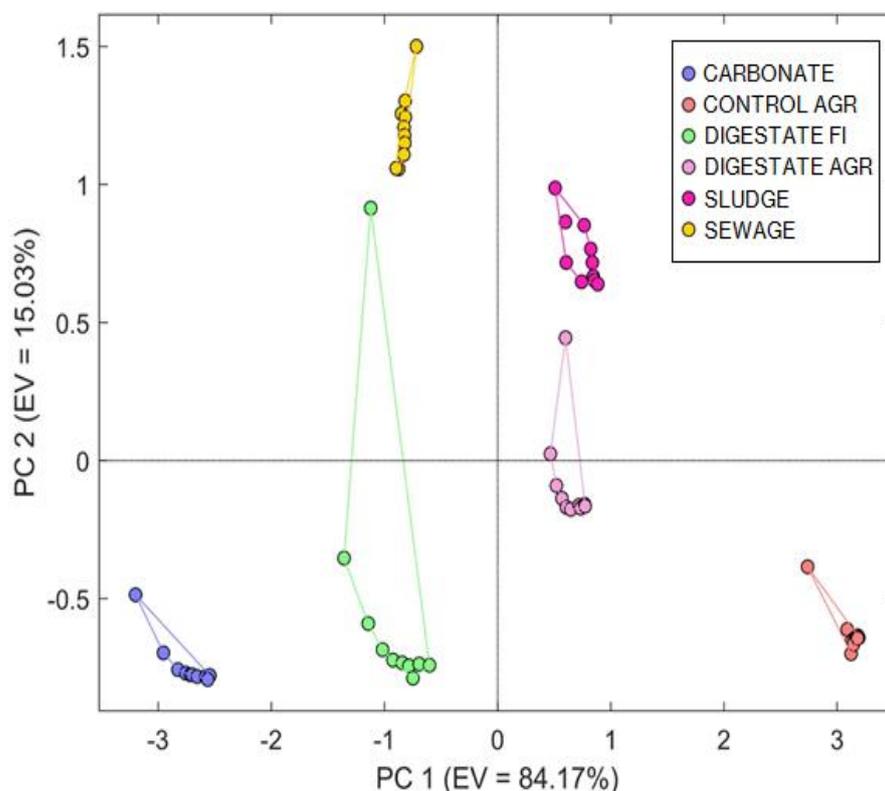


Figure 2. Principal component analysis plot depicting the 6 sample clusters. Each color represents a group sample to which the compounds belong.

#### 4. Conclusion

As a conclusion, it was possible to obtain a volatile fingerprint of the VOCs present on the 6 typologies of samples with respect to Small Sensor System (S3) device. It can be concluded that the volatile compounds of each sample have an important impact during the passage from organic source to biomethane and organic fertilizers. This work paves the way to the adoption of an array of chemical sensors to detect and recognize olfactory harassment related to biomethane production. As a matter of fact, biomethane will have in the future an important role in the energy field, being a renewable and sustainable energy source that can give a huge contribute to favor of the decarbonization. This new innovative technology demonstrated our hypothesis, since the possible differences between the different organic matrices involved in the different steps of biomethane production and green fertilizers obtained from different organic sources were proved. The success of this study can lay the foundation to deepen the research in this field to use S3 as a tool for distributed generation and management of green energy. Furthermore, it could have an important impact in the creation of an integrated and multifunctional HUB for the development of the waste treatment and green energy sector, combining the need for innovation in the waste-management chain through the synergy between renewable energy and green chemistry.

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