

# Adverse Impact of Additive Carbon Material on Microorganisms

Subjects: Engineering, Environmental

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Biochar could be a brilliant additive supporting the anaerobic fermentation process. However, it should be taken into account that in some cases it could also be harmful to microorganisms responsible for biogas production. The negative impact of carbon materials could be a result of an overdose of biochar, high biochar pH, increased arsenic mobility in the methane fermentation solution caused by the carbon material, and low porosity of some carbon materials for microorganisms.

Keywords: biochar ; hydrochar ; biogas ; anaerobic digestion ; carbon material

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## 1. Influence of Biochar on Changes in the Microbiocenosis Habitat and Colony Growth

The efficiency of adding biochar to anaerobic digestion microorganisms is highly dependent on the substrate used to make the biochar. For example, experiments conducted by <sup>[1][2]</sup>, and <sup>[3]</sup> have proven that the use of different substrates of biochar highly changes the composition of microorganism species and even the phylum. It is worth adding that even the mineral particle ratio in biochar structures could be highly affected by the species composition during anaerobic digestion <sup>[4]</sup>. The influence of biochar on microbiomes was proven by <sup>[5]</sup>; moreover, <sup>[6]</sup> proved that biochar and carbon felt could increase the microbiome ratio of *Methanosaetaceae*.

The high sorption features of biochar could be positive because of pollutants absorbed during anaerobic digestion <sup>[7]</sup>, but this feature could also be problematic. For example, when a methane fermentation solution has a low nitrogen ratio, an excessively large amount of biochar additives could lead to a deficiency in nitrogen and make it inaccessible to microorganisms <sup>[8][9]</sup>; moreover, a high pH of biochar could also be harmful for microbiocenosis <sup>[8]</sup>. Needless to say, the highly porous structure has some advantages, but, on the other hand, the small pores could trap some nutrients or elements inside, and because of their dimensions, prevent microorganisms from adsorbing those kinds of crucial elements <sup>[10]</sup>. In this case, the golden mean should be kept when designing the conditions for creating biochar to ensure the appropriate pore size for adsorbing pollutants and such that microorganisms have possible access to any nutrients that may be present in the pores. However, this could be a very difficult task, and perhaps it will be easier to control the content of the nutrients taking into account the possible "losses" of the nutrients in the pores.

The high pH of biochar could influence the pH level of the methane fermentation solution, and hence, the alkaline solution could start converting  $\text{NH}_4$  ions into toxic  $\text{NH}_3$ , which could pose a potential threat to microbial communities <sup>[11]</sup>. Interestingly, some studies have found no connection between the influence of the physical properties (like electrical conductivity and surface area) of the biochar and an increase in the biogas yield, although it could increase the rate of anaerobic digestion <sup>[12]</sup>.

That change could have a beneficial effect on biogas production because it could allow for the development of microorganism's abundant species, but it could also disturb a microorganism balance in the methane fermentation solution habitat; for example, it could decrease beneficial microorganisms in the microbiome mix and/or increase the amount of unnecessary microorganisms. Moreover, changing the habitat features could force beneficial microorganisms to use some amount of energy to adapt to new environmental conditions <sup>[13]</sup>. Solutions with low concentrations of nickel generally promote two types in microbial communities, *Methanosarcina* and *Methanosaeta* <sup>[14]</sup>. Biochar itself could also have more negative effects on eubacteria (like *Firmicutes* and *Proteobacteria*) than archons <sup>[15]</sup>.

Pyrochar produced in high-temperature pyrolysis (like 700 °C) could decrease the bacterial community from *Methanosaeta* species <sup>[16]</sup>. Biochar could also present a harmful influence on microorganisms by releasing toxic elements directly into an anaerobic digestion solution; for example, biochar modified by  $\text{KH}_2\text{PO}_4$  could increase arsenic mobility in

swine manure used as a biogas substrate <sup>[17]</sup>. This property of biochar, despite the quite good stabilization of heavy metals, such as Cr, Cu, Pb, and Zn, should be taken into account when trying to use waste containing high concentrations of heavy metals for energy purposes <sup>[18]</sup>.

It is worth noting that the very important aspects during the use of biochar-like additives in biogas production increase, as is commonly known “only the dose makes the poison”, and this sentence could also be accurate in this case. The overdose of biochar in an anaerobic digestion solution could negatively affect methanogenic efficiency and extend the lag phase <sup>[19]</sup>.

## **2. Hydrochar Influence of Microbiocenosis during Methane Production**

Hydrochar is a type of carbon material that could be produced from wet material, like fruit pomace <sup>[20]</sup>, kitchen waste <sup>[21]</sup>, or sewage sediment <sup>[22]</sup>, with the use of a hydrothermal carbonization process. Needless to say, that kind of process could help to utilize high-moisture waste and transform it into useful fuel. This kind of material could be used similarly to biochar (sometimes even with better results <sup>[19]</sup>, and also as an additive for improving the biogas yield <sup>[22][23]</sup>.

In general, hydrochar additives could improve biogas yields by promoting DIET and selecting microbiocenosis into a more productive mix with an increase in the *Methanobacterium* percentage <sup>[23]</sup>. Another noteworthy study <sup>[24]</sup> explores the use of hydrochar as an adsorbent for ammonia, a compound with potential biogas production benefits, in anaerobic digestion. However, the results indicate that the adsorption of ammonia by hydrochar may not significantly enhance biogas production. Moreover, hydrochar could enrich some microorganism species, like *Methanobacterium*, *Methanosaeta*, *Clostridium*, and *Methanosarcina* <sup>[25]</sup>. Unfortunately, hydrochar additives, of course, could enrich species, like *Methanosaeta* or *Syntrophomonas*, but also, at the same time, it could be harmful to the population of acidogenic and hydrolytic groups of microbiocenosis, for example, *Acinetobacter* <sup>[26]</sup>.

But, adding hydrochar to an anaerobic digestion solution does not always improve biogas production <sup>[24][27]</sup>; the properly chosen ratio of hydrochar promotion is crucial for improving biogas production and supporting microbiomes. Another important factor during the use of hydrochar in improving biogas production yields could be the temperature during the HTC process, as in an experiment provided by Choe et al. <sup>[24]</sup>. Hydrochar usually improves the biogas yield, except in the case when a tofu residue was caused by hydrothermal pretreatment at a temperature of more than 140 °C when the biogas yield starts to decrease linearly <sup>[24]</sup>. Needless to say, the negative influence of hydrochar on microbiocenosis requires further research.

## **3. Influence of Activated Carbon on Microbiocenosis during Methane Production**

Activated carbon is a carbon material characterized by strong porosity, due to the large surface area <sup>[28]</sup>. That area is crucial for the most important feature; it could be a great chemical adsorbent. Using activated carbon during the anaerobic digestion process could increase biomethane production, very similar to adding biochar and hydrochar, which were described in previous acts. Moreover, adding activated carbon to a methane fermentation solution could increase a population of similar groups for example; *Methanosaeta* <sup>[29]</sup> and *Methanosarcina* <sup>[30]</sup>. Moreover, activated carbon could be used as an adsorbent to remove H<sub>2</sub>S from the biogas mix <sup>[31]</sup>.

Activated carbon could reduce pathogenic microorganisms even by 18%, but on the other hand, this additive could harm microorganism biodiversity in the methane fermentation solution habitat <sup>[32]</sup>. It could be worth taking a closer look at the influence of decreasing biodiversity and its effect on the biogas production ratio. Quality and safety of the fertilizer from the biogas production process with activated carbon additives could be also interesting for further research.

## **4. Impact of Nanoparticles**

Nanoparticles are particles that do not exceed 100 nanometers but are larger than 1 nanometer <sup>[33]</sup>. Despite their small size, their influence on microorganisms is very significant; thus, metal nanomaterials from biochar have high levels of reactivity, a widespread surface area, and strong surface energy. Moreover, it is possible to modify the surface properties of biochar by using nanometal materials <sup>[34]</sup>, that feature could be helpful in the adsorption of pollutants from a biogas tank solution. Needless to say, not even nanoparticles, like a part of the biochar component, could influence microbiocenosis, and even changing the size of biochar could change its properties. For example, the features of macro-size biochars are different from nano biochars <sup>[35]</sup>.

Silver nanoparticles are strongly harmful to microorganisms [36], so it is very important to alleviate that impact. This kind of nanoparticle could be adsorbed by biochar [37] and allow microorganism colonies to grow in the easiest habitat.

It is worth saying that not only non-organic nanoparticles could affect the biogas yield. Nanographen could also have a harmful effect on microbiocenosis (for example, *Methanosaeta*, *Lactococcus*, and *Anaerolinea*) during long-term exposition in 120 mg/L concentration of nanographene in a methane fermentation solution [38]. That graphene additive, but on a macro-scale, could also decrease the population of *Methanosaeta* [39]. Moreover, too high of a concentration of graphene in the methane fermentation solution could be harmful to anaerobic digestion [39].

The summary of some examples of the negative impacts of carbon material additives on microbiocenosis in an anaerobic digestion habitat is presented in **Table 1**.

**Table 1.** Summary of some of the negative impacts of individual carbon material additives on microbiocenosis in an anaerobic digestion habitat.

Type of Carbon Material	Physicochemical Properties of Carbon Material	Potential Problem	Type of Negative Influence	References	Solution Proposal
Biochar	Strong porosity structure and adsorption capacity	Overdose of biochar	Scarcity of nitrogen supply for microbiocenosis when the ammonia nitrogen concentration is low	[8][19]	Care should be taken to choose the right dose for the biogas plant and remember that the dose should always be adjusted to the substrate used
Biochar	High biochar pH	High biochar pH that could promote the transformation of $\text{NH}_4^+$ into $\text{NH}_3$	A high pH of biochar could promote the conversion of $\text{NH}_4^+$ to $\text{NH}_3$ , which could be harmful to the microbiocenosis during anaerobic digestion because $\text{NH}_3$ is more toxic than $\text{NH}_4^+$	[8]	Monitor the pH level of the solution on an ongoing basis before and after adding biochar and correct the pH if necessary, depending on the possibilities
Biochar	Strong porosity structure	Pores of carbon materials are too narrow for microorganisms, preventing uptake by microorganisms	Too narrow pores could prevent absorbing nutrients and crucial chemical compounds for microbiocenosis	[10]	Preventing the formation of micropores that are inaccessible to microorganisms could be a difficult challenge. In this case, it is proposed to adjust the amount of medium, taking into account that some will be retained in the pores

Type of Carbon Material	Physicochemical Properties of Carbon Material	Potential Problem	Type of Negative Influence	References	Solution Proposal
Biochar/Hydrochar/Activated carbon/Graphene	Strong porosity structure and adsorption capacity, high biochar pH, a chemical component of the carbon material, and the content of heavy metals in the biochar. And any other properties that can affect the habitat of AD microbiocenosis.	Reducing the biodiversity of microorganisms in methane fermentation solution	Changing the properties of the anaerobic digestion solution that is a habitat for microbiocenosis may cause some groups of microorganisms to tolerate environmental change worse than others, which may disturb the original species composition.	[15][26]	Actions should depend on which bacterial species do not tolerate biochar additions. If these species do not participate directly or indirectly in the production of methane and the installation is industrial, this problem can probably be omitted or try to select biochar with other properties. Just remember to take the risk into account when modifying natural ecosystems with biocarbon additives
Biochar/probably most Carbon Material	Strong porosity structure and adsorption capacity, high biochar pH, a chemical component of the carbon material, and the content of heavy metals in the biochar. And any physicochemical property that may influence the change of the AD solution that is the habitat of the microbiocenosis	Necessity to invest energy by microorganisms to adapt to a new habitat enriched with biochar	Habitat changes may force microbiocenosis, part of energy expenditure, to adapt	[13]	The risk of whether biochar with given properties will be beneficial for microorganisms should be determined
Biochar	Adsorption properties of biochar	Could increase arsenic mobility in methane fermentation solution	Arsenic as a heavy metal could be harmful to methanogenic microorganisms	[26]	Choose a substrate with as few heavy metals as possible or try to use biochar with other properties. In addition, it is worth paying attention to the feature of feedstock from which it was used to create the biochar

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