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# Bacterial genetic diversity evaluation in anaerobic digested biomass, based on the 16S rDNA gene by denaturing gradient gel electrophoresis

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## INTRODUCTION

Anaerobic waste treatment technologies in (biogas plant) enable disposal of a wide variety of wastes, being substrates for this process. These include, inter alia, agricultural waste, or waste from the food industry, sewage sludge or municipal waste. In recent years there has been a significant increase in interest in the possibility of anaerobic organic waste degradation from various sources in order to obtain high-biogas production.

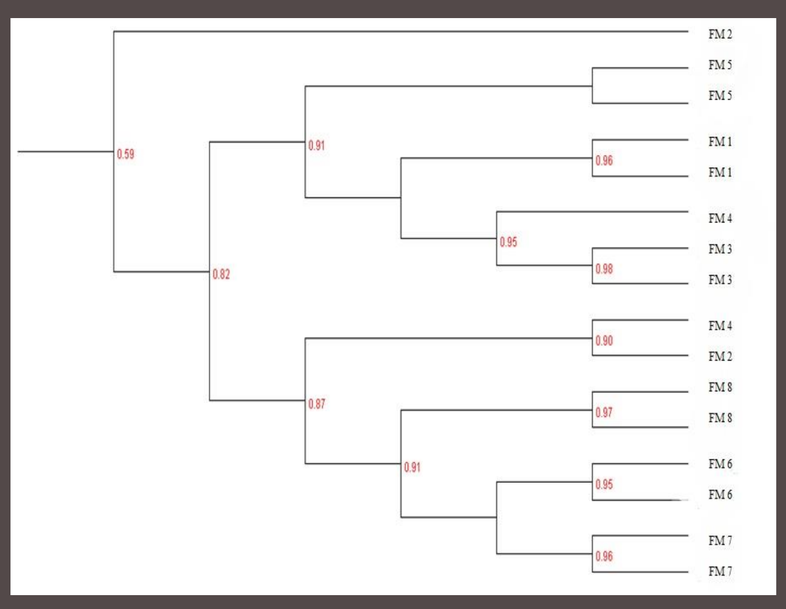
Transformation of complex organic compounds in organic wastes into $CH_4$ and $CO_2$, is possible due to the interaction of four different groups of microorganisms. These groups include the following: bacteria of primary fermentation, secondary fermentation bacteria (syntrophic and acetogenic bacteria).

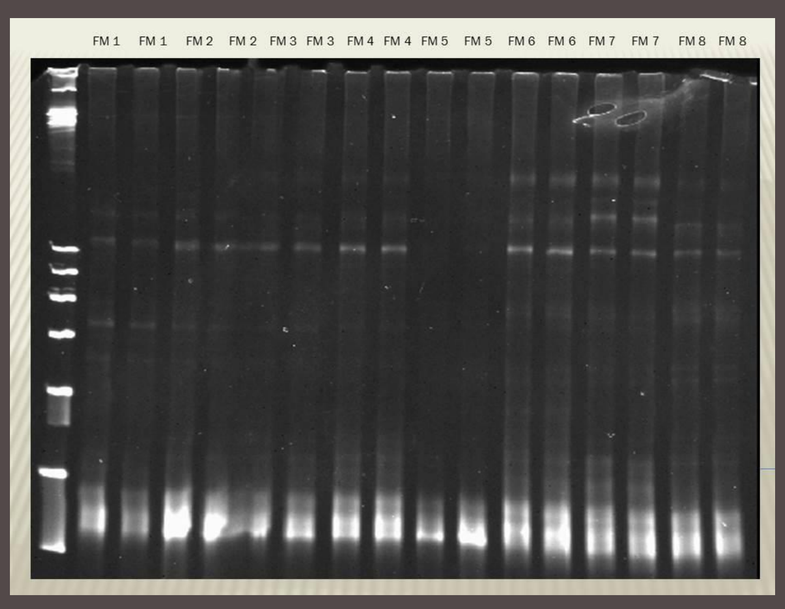
## OBJECTIVE

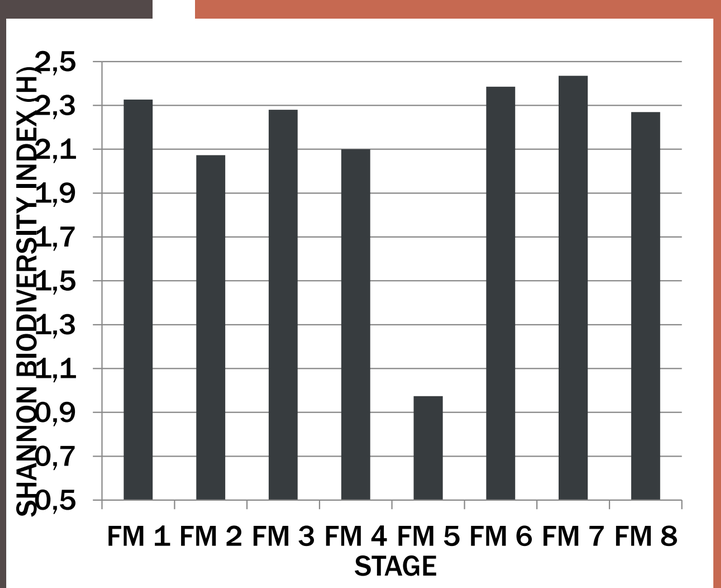
The aim of this study was to determine the changes in genetic diversity of bacterial communities inhabiting the fermentation biomass collected at 8 different stages of the anaerobic digestion process (FM1-FM8).

## MATERIAL AND METHODS

The analysis consisted of two steps: amplification of the 16S rDNA gene and a denaturing gradient gel electrophoresis (DGGE). For amplification the following primers were used: gc-968f (5'*CGCCCGGGGCGCGCCCCGGGCGGGGCGGGGGCAC GGGGGGAACGCGAAGAACCTTAC*3') and UNI1401r (5'*GCGTGTGTACAA GACCC*3'). The process of amplification consisted of the following cycle: 94°C – 90 s, 56°C – 30 s, 72°C – 45 s, (95°C – 20 s, 56°C – 30 s, 72°C – 45 s) x 27, 72°C – 7 min. In a next step the electrophoresis was performed in the denaturing gradient gel (DGGE) (conditions were as follows: 16 hours, 70 V, 60°C in TAE buffer 1x). The composition of the analysed biomass included: waste of fruit processing (25%), dairy sewage sludge (25%), corn silage (12%) and grain decoction (38%).




## RESULTS and KEY FINDINGS

The results showed a great variation between communities of bacteria depending on the stage of the process. This study highlighted changes in anaerobic bacterial diversity during 8 stages of anaerobic digestion process. Particular stages were characterized by succession of different anaerobic populations involved in this process, which are important for effective run of the process.