In Brazil, the annual production of ethanol is estimated in 25 billion liters. For each liter of ethanol produced, approximately 12-15 L of vinasse is generated. Currently, vinasse is used as a fertilizer and potassium source in the cultivation of sugarcane. However, the long-term use of vinasse in productive lands can cause desertification and land salinization, resulting in productivity decrease. As such, environmental agencies are limiting the amount of vinasse added per hectare of land due to its high pollutant potential. Several studies have been conducted with the objective of developing new treatment forms and disposal of vinasse. The use of this residue for biogas production and other compounds of economic interest have been investigated. The present study evaluates the production of hydrogen and volatile fatty acids by a mixed bacterial culture using different concentrations of sugarcane vinasse (7-20g/L COD) as substrate, at 37°C and with initial pH 6.0 in anaerobic conditions. The inoculum was obtained from a granulated sludge of an UASB reactor used for vegetable oil wastewater treatment. The biomass was macerated and preheated at 90º for 10 min. Higher hydrogen production (5.50 and 10.84 mmolH₂) was observed with the two highest concentrations of vinasse used (15 and 20g/L COD) respectively, in a period of 24 h. Acetic, propionic and butyric acid production also was higher as vinasse concentration was increased. With the highest concentration of vinasse (20 g/L COD) a final consumption of 68% of total carbohydrate was determined. Microbial growth and COD reduction were observed in all concentrations tested. The microscopic analysis showed the presence of the gram-positive bacillus bacteria. Molecular biology and DGGE analysis of the microbial consortium revealed the presence of bacteria belonging to the genus *Clostridium* in all vinasse concentrations evaluated. This genus is probably involved in the biologic production of H₂. These results showed that vinasse has the potential to be used as a substrate for the
production of compounds with economic interest, which may alternatively reduce the inappropriate disposal of this residue in the environment.

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