



## Comparison of mixed-acid fermentations inoculated with six different mixed cultures

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### HIGHLIGHTS

- ▶ Fermentations performed under same operating parameters with six inoculum sources.
- ▶ 16S rRNA sequencing identified the bacterial communities in the fermentations.
- ▶ Yue–Clayton similarity calculations revealed that they were extremely different.
- ▶ All fermentations had similar conversion and end-product formation performance.
- ▶ The operating parameters determined the fermentation end-products.

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### ABSTRACT

The MixAlco™ process biologically converts biomass to carboxylate salts that may be converted to a variety of chemicals and fuels. This study examines the fermentation performance of six different mixed cultures, and how the performance was affected by the bacterial composition of each community. All six countercurrent fermentations had very similar performance, but were dissimilar in microbial community composition. The acid concentrations varied by only 12% between fermentation trains and the conversions varied only by 6%. The microbial communities were profiled using 16S rRNA tag-pyrosequencing, which revealed the presence of dynamic communities that were dominated by bacteria resembling Clostridia, but they shared few taxa in common. Yue–Clayton similarity calculations of the communities revealed that they were extremely different. The presence of different but functionally similar microbial communities in this study suggests that it is the operating parameters that determine the fermentation end-products.

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### 1. Introduction

As energy demand increases and the availability of fossil fuels decreases, the need for alternative energy sources grows. Anaerobic fermentation of waste lignocellulosic biomass has the potential to meet this need (Chan and Holtzapple, 2003; Holtzapple et al., 1999). Over a billion tons of agricultural, municipal, and industrial wastes are generated annually in the United States that could potentially be used in biofuel production (Perlack et al., 2005).

The most common method to convert lignocellulosic biomass to products is simultaneous saccharification and fermentation (SSF), which enzymatically converts lignocelluloses into sugars that are

then fermented into alcohols (Lin and Tanaka, 2006). Unfortunately, the commercial cellulases for this process are expensive (Rubin, 2008). The process also requires sterility which is difficult to maintain with lignocellulosic biomass because of its variable consistency and porosity. A lesser known alternative is the carboxylate platform, also known as the MixAlco™ process (Holtzapple et al., 1999).

The MixAlco™ process is a flexible and cost-effective means of converting a variety of lignocellulosic feedstocks (e.g., agricultural residues, municipal solid waste, and biosolids) into chemicals and liquid fuels. A mixed culture of naturally occurring microorganisms ferments the biomass into carboxylate salts, which can be converted into a wide array of chemicals, including alcohols, jet fuel, and gasoline (Aiello-Mazzarri et al., 2006; Granda et al., 2009). The product spectrum from this process is temperature dependent and can be varied in response to market demand (Chan and Holtzapple, 2003). Additionally, this process has no sterility

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