High butanol-producing bacterial strain

Auburn University is seeking a licensee or development partner for a biobutanol-producing bacterial strain.

**Overview:** n-Butanol (or butanol) has multiple industrial uses as a solvent, intermediate/feedstock in various industrial processes, a renewable fuel with advantages over ethanol, and others. Chemical synthesis of butanol, however, can be costly and generates unwanted pollutants. Clostridial bacteria have long been studied as a potential source of butanol (biobutanol), but production levels sufficiently high to make them cost effective had not been reached. To address this, *Clostridium* was strategically engineered, increasing biobutanol production by 100% or more over previous strains. This technology has potential applications in the solvent/extractant industries as well as biofuels and white biotechnology sectors.

**Advantages:**
- **HIGH BUTANOL PRODUCTION** - twice that of the conventional strain, providing up to a predicted 33% in energy savings in downstream recovery processes.
- **LESS BYPRODUCTS** - No acetone, facilitating conversion to more valuable products.

**Description:** Butanol has been known to be produced by microorganisms, most notably by clostridial bacteria. Previous attempts to increase production topped out at 13 g/L. Through genetic modifications to a strain of *Clostridium tyrobutyricum* replacing the *cat1* gene with the alcohol dehydrogenase gene *adhE1/adhE2*, biobutanol production was boosted to 26.2 g/L - the highest level to date and a 100% increase over the conventional biobutanol-producing strain. In addition, coproduction of acetone was extremely low or undetectable. Culturing was done under standard anaerobic batch conditions. A CRISPR-Cas system capable of efficient single or multiplex modifications was developed for genome engineering in this strain. Additional engineering and methods to further boost production are being explored.

**Status:**
- A provisional patent application has been filed
- Lab scale demonstration of high *n*-butanol production under anaerobic batch conditions
- This technology is available for co-development and licensing

Fermentation profiles of engineered *C. tyrobutyricum* showing 26.2 g/L butanol production in a batch fermentation.