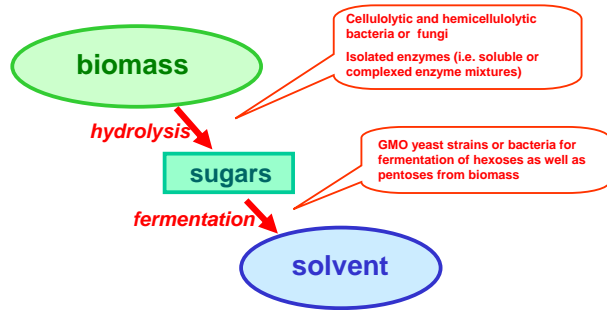


# Bacteria for our future biofuels

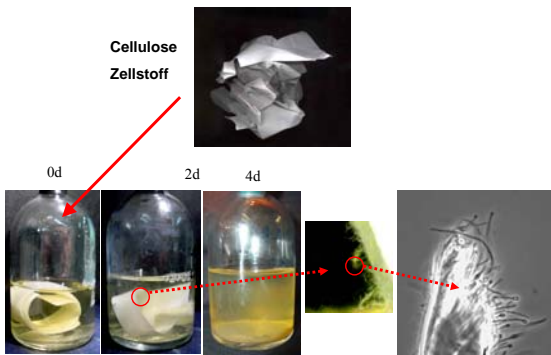
## Introduction

Bacteria are involved in at least two areas of biofuel production processes from renewable biomass (lignocellulosic biomass, LCB): (1) production of enzymes and abd hydrolysis of polysaccharides, and (2) fermentation of the sugar syrups to short chain fatty acids or alcohols. An example is butanol through the Weizmann process with *Clostridium acetobutylicum*. A crucial advantage of bacterial fermentations is the usage of pentoses (in addition to the hexoses) which are produced from hydrolysis of hemicellulose. Examples of such processes and new developments are shown here.



## *C. thermocellum* has a highly efficient cellulase system

Filter paper is degraded by a growing culture of *C. thermocellum* within 2 days at 55 to 68 °C. Fermentation products are ethanol, acetate, CO<sub>2</sub> and hydrogen. The bacterial cells adsorb tightly to the substrate surface and take up the hydrolysis products directly (Fig. 2). *C. thermocellum* forms small colonies on an anaerobically prepared agar plates. Colonies hydrolyze crystalline cellulose around the colonies on the plate surface (Fig. 3).



David Ramey: "Butanol is an alternative power grade alcohol that replaces gasoline in your car today without having to be highly modified."

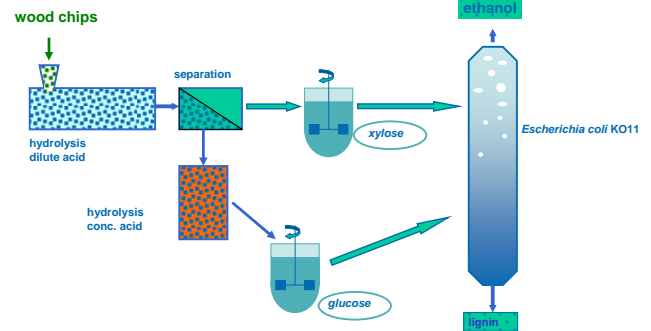
**Table 1:** Comparison of fuel characteristics: End products of the AB process and some basic data, including combustion energy, for the products. Ethanol from yeast fermentation of corn starch (industrial average) and some data on gasoline and diesel are shown for comparison.

product	kg <sup>a)</sup>	kJ/g	MJ/t substrate <sup>b)</sup>
butanol	207 ± 10	33.1	6852
acetone	100 ± 8	28.5	2850
ethanol	56 ± 15	26.7	1495
CO <sub>2</sub>	≈ 600	-	-
H <sub>2</sub>	≈ 19	119.8	2276
<b>Total AB</b>			<b>13273</b>
ethanol (yeast)	≈ 380	26.7	10146
CO <sub>2</sub> (yeast)	≈ 330	-	-
gasoline	-	43.4	-
Diesel fuel	-	42.7	-
<b>Total ethanol</b>			<b>10146</b>

a) kg product per ton of starch equivalent; b) kJ combustion energy (net calorific value) in end product per ton of starch equivalent.

## Bioethanol production from wood-chips

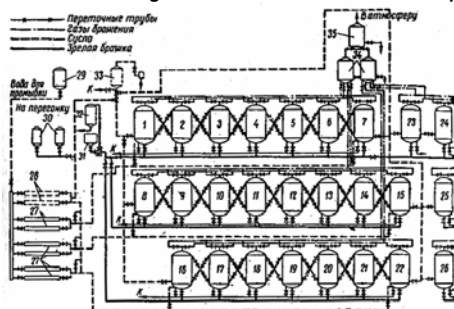
The bioethanol plant of BC Intl. Corp., Dedham (MA) hydrolyses wood-chips in a two step-acid hydrolysis producing a pentose syrup from hemicellulose (step 1) and glucose syrup from cellulose (step 2). The sugar syrups are fermented with the recombinant *E. coli* strain KO11 (the "Florida-strain") to ethanol and CO<sub>2</sub>.



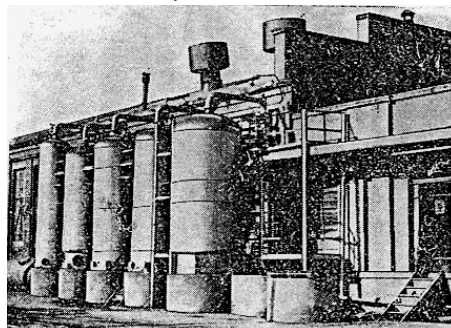
## An example butanol plant for LCB usage

Schematical presentation of the acetone-butanol production line: the substrate passes 3 parallel lines of 7 or more 250m<sup>3</sup> fermenters each (#1-26). The preculture fermenter is #33, the feeding fermenter is #30. #27/28 are substrate coolers. CO<sub>2</sub> is collected in #34/35. At the time of description in 1975 this plant has been run successfully for 10 years and produced 70.000 to/a solvents (ABE 4:6:1) and 28.000 to/a gases (CO<sub>2</sub>, H<sub>2</sub> 1,5:1). Eight such plants are known.

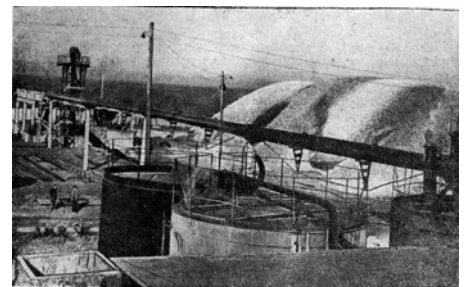
### Schematical drawing of the Dokshukinski fermentation plant



### The Dokshukino plant in 1969: fermentation



### Substrate storage and hydrolysis at Dokshukino plant (1963): corn stubs



It was reported that the *Dokshukinski* plant was run with a mixture of flour (heat treated) and molasses (30:70). About 7,5 % of an acid hydrolysate from corn stubbers and other agricultural waste was used (sulfuric and phosphoric acid 3:1 or 4:1).

**Efficiency:** 50 % of the substrate sugars were converted to CO<sub>2</sub>, and 33-39 % to solvents (60 % of that was butanol), leading to a yield of 9 tons solvents per 100 tons grain. By-products were CO<sub>2</sub>, vitamin B<sub>12</sub> and yeast protein for feed. The production of methane gas provided the energy for running the process.