Dextran and Fructose production using *Leuconostoc mesenteroides* NRRL.B512(F) with sucrose as substrate

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Keywords:dextran, fructose, dextransucrase, fermentation

Dextran and fructose have many industrial applications. Dextran is widely used as a blood volume expander, in food industry and as a chromatographic media. Fructose is a low caloric sugar.

The strain *Leuconostoc mesenteroides* NRRL.B512(F) grows in sucrose rich media which induces the production of an extracelular enzyme: dextransucrase. This enzyme also uses sucrose as a substrate to produce dextran and fructose as follows:

$$n\ C_{12}H_{22}O_{11} \xrightarrow{\text{enzyme}} (C_6H_{10}O_5)n + n\ C_6H_{12}O_6$$

Sucrose Dextran Fructose

In this work a 2 l bioreactor (Setric Genie SET02, Incheltec, France) with control units for temperature, pH and agitation was used. Strain growth and products concentration for various operating conditions was studied.

Batch fermentations were carried out for sucrose concentration in the range 10 to 120g/l, temperatures from 20 to 40ºC, pH of 6.9 (optimum pH for strain growth) and 5.5 (for mimizing loss of enzyme activity) and aeration rate of 0.05 vvm. Fed-batch fermenations were carried out with different start-up times and feed flowrates.

Cell growth was significantly higher for T=35ºC and fermentation took 4 hours less than at 20ºC. Experiences at 25ºC and controlled pH at 6.7 resulted in higher cell growth than at pH 5.5. At lower pH values (5.5) dextransucrase production was faster in the earlier hours of fermentation and the decrease in acctivity was slower. Cell growth and product formation were not affected by aeration. Results with fed-batch operation were similar to those obtained with batch operation.

Studies of metabolic engineering of the strain were made. The evolution of the Carbon/Nitrogen ratio throughout the fermentation was studied. A fermentation was carried out correcting the nitrogen level around the 4th and 7th hour. Results in enzyme productivity were improved.

No growth of the bacteria was observed in the presence of other carbon sources than sucrose, such as maltose, lactose and galactose.

The enzyme production in the presence of these sugars together with sucrose was analysed. None of the three sugars was consumed by the bacteria and the cellular growth and enzyme activity were smaller comparing with the results of a fermentation using only sucrose.

The kinetics of enzymatic conversion of sucrose in the presence of dextransucrase follow Michaelis-Menten equation. When maltose is used together with sucrose reaction rate decreased.