Shodex Rapid SUGAR Series: Fast Fermentation Monitoring and Sourdough Cultures

Alexander Schrum, Ronald Benson, Marlon Rettis

Showa Denko America, Inc.

Introduction

It was not until the 19th century that living organisms (yeast and bacteria) were identified as the agents that produced and spoiled common food items. Baking, especially of bread, was among the first industries to be transformed by the new understand of fermentation. The introduction of commercialized yeast in the early 20th century steadily reduced the time needed to bake a loaf of bread both at home and on the production line (1). Synthetic biology is pushing the field further, converting feedstock into a wide range of useful compounds. However, unaffected by recent advancements, traditional fermentation techniques, including sourdough breads, having resurged in popularity. Typical sourdough cultures are created from yeast and bacteria populations naturally present in the air and on many surfaces, providing sourdough loaves with their characteristic rise and "tangy" flavor by converting fermentable sugars into CO2 and flavor compounds like lactic acid. In this application note, a "homemade" sourdough starter culture was analyzed to determine how key fermentation products changed over the course of 24 hours with the new Rapid SUGAR SH1011 8C.

The Shodex Rapid SUGAR SH1011 8C contains a styrene divinylbenzene base material ligand exchange column and is designed for rapid fermentation monitoring, allowing for the analysis of saccharides, organic acids, and ethanol in less than 5 minutes. The method uses simple aqueous conditions and RID detection, ideal for large sample workflows and QC environments.

Experimental

Seven common fermentation compounds (Maltotriose, Maltose, Glucose, Lactic acid , Acetic acid, Glycerol, Ethanol) were used as standards.

The sourdough starter culture was created using the following procedure

Day 1: Into 500 ml beaker, 1 cup of King Arthur Bread flour + ½ cup of Di water and stirred. Covered and left at room temperature

Day 3: the mixture was stirred and approximately half was removed, 1 cup of flour + ½ cup of water was added and mixed

Day 4-14: Day 3 procedure was repeated daily

Note: By day 14, the sourdough started culture exhibited vigorous bubbling within approximately 2 hours of feeding

Sample preparation: adapted from Dominique (2)

Approximately 10 g of sample was homogenized with 90 mL distilled water (maximal speed, 30 s, Blender). Five milliliters of 1 mol/L HClO4 solution was added to a 10 mL aliquot of the homogenate. The mixture was centrifuged for 15 min at 4000 g at 15 1C, the supernatant was neutralized (pH 7.0) with 2 mol/L KOH and the volume was adjusted to 25 mL with distilled water. After 30 min precipitation on ice, the solution was filtered on 0.45 mm cellulose filter

Samples were taken starting on Day 15 at three time points

Sample 1: Immediately after mixing in flour and water, labelled time 0

Sample 2: 12 hours after time 0

Sample 3: Day 16, approximately 24 after time 0

Shodex™ SUGAR SH1011 8C (8.0 mm I.D. x 100 mm, 6 µm) was used with a Shodex RI-501. The eluent conditions were as follows: 1 mM H_2SO_4 aq. The column was kept at 75 ºC and the flow rate was 1.0 mL/min.

Results and Discussion

Fig. 1 shows the RI chromatograms of the standards. Peaks of the major by-products were fully resolved using the developed method.

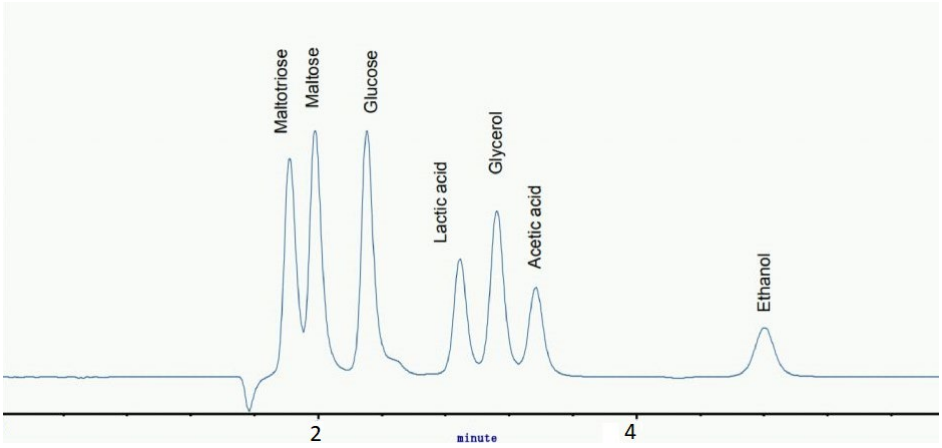

Figure 1: RI chromatogram of fermentation standards, 0.1% w/v each, 5 uL injection

This simple method using 1 mM Sulfuric Acid as the eluent demonstrated a successful simulates analysis of fermentation by-products in less than 5 minutes, including the column equilibration time.

For the analysis of the sourdough starter culture a guard column was used (Shodex™ SUGAR SH-G (6.0 x 50 mm id, 10 µm)). (Fig.2)

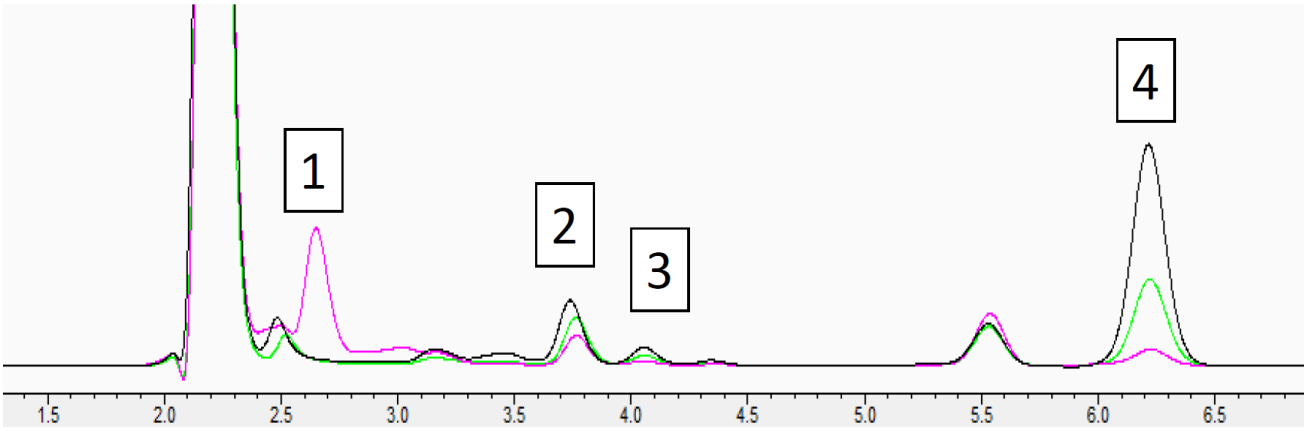

Figure 2: Sourdough timepoints – Pink is time 0, Green is 12 hours, Black is 24 hours. Peak 1 – Glucose and sample matrix. Peak 2 – Lactic Acid. Peak 3 – Glycerol. Peak 4 – Ethanol

A method for rapid fermentation monitoring was developed using the Shodex™ SUGAR SH1011 8C column. The eluent used was a simple dilute sulfuric acid solution. Fermentation sample measurement completes in 6 minutes. The column showed good robustness when used with complex sample matrixes like sourdough cultures. The results show that the SH1011 8C can be used to monitor the increase of fermentation by-products like organic acids and ethanol as feedstock is converted to finished products.

References

1) Marx, Jean & Litchfield, John H. (1989). A Revolution in biotechnology. Cambridge, UK: Cambridge University Press. p. 71.
2) Dominique, Lefebvre & Valerie, Gabriel & Vayssier, Y & Fontagné-Faucher, Catherine. (2002). Simultaneous HPLC Determination of Sugars, Organic Acids and Ethanol in Sourdough Process. LWT - Food Science and Technology. 35. 407-414..

Shodex™/Showa Denko America, Inc.

420 Lexington Avenue Suite 2335A, New York, NY, 10170

Phone: 212-370-0033x116

Email: Support@shodexhplc.com

Fax: 212-370-4566

www.shodexhplc.com