MEP instruments
The right chemistry.

MEP Application-0573-052015

Determination of organic anions in bread dough.

Branch

7. Food, stimulants, beverages, flavors

8. Biochemistry, biology

Keywords

IC / 930 / 858 / Compact IC / Metrosep Organic Acids– 250 / Formate / Acatate / Lactate / Conductivity / Chemical suppression / branch 7- 8 / 2.930.2360 / 2.858.0020 / dialysis

Background

A drop in the acidity of sourdough during the fermentation process has an important impact on the biochemical changes in the carbohydrate and protein structures.

The drop in pH value caused by organic acids doesn't only affects the taste and smell but also to physical properties such as the viscoelastic behavior of the dough.

In order to get a good understanding about the acidification process of the sourdough and partial acidification of the bread dough, it's important to quantify the acids that are responsible for the pH drop.

Certain acids will contribute stronger to the acidification than others due to the differences in the pKa values. (Refer to table below).

Organic acid	pKa
Lactic	$1.4x10^{-4}$
Formic	$1.8x10^{-4}$
Acetic	$1.8x10^{-5}$
Carbonic	$4.2x10^{-7}$

Summary

The organic acids were extracted by using a mixture of acetonitrile and water. The acetonitrile was added to stop the biological processes in the fermentation samples.

The separation of the anions was done via ion exclusion on the Metrosep Organic Acids column.

Samples

The fermentation samples were kindly provided by Allied Mills Australia.

Instruments

930 Compact IC Flex Oven/ChS /PP/Deg	2.930.2360
858 Professional Sample Processor	2.858.0020

Columns

Metrosep Organic Acids – 250 / 7,6	6.1005.000
Metrosep RP2 Guard	6.1031.030

Reagents

- Ultrapure water, resistivity >18 MΩ·cm (25 °C), type I grade (ASTM D1193)
- Acetonitrile LC-MS CHROMASOLV® / 4967 FLUKA
- Perchloric acid concentrate :0.01 M HClO4 in water (0.01N), eluent concentrate for IC / 50439-Fluka
- Lithium chloride : Powder, ≥99.99% / 203637 - Aldrich

Solutions

Eluent:	2.0 mmol/L $HClO_4$
Suppressor regenerant solution	0.1 M LiCl in water

Standard solutions

All standards were made up in ultra-pure water [mg/L]

	Std. 1	Std. 2	Std. 3
β(Lactate)	100	250	500
β(Formate)	10	25	50
β(Acetate)	10	25	50

Sample preparation

1.00 g of dough was weighed into a small beaker and almost immediately suspended in 10 ml of acetonitrile. The suspension was then transferred in a volumetric flask and further diluted to 100 ml with ultra-pure water. An aliquot of the sample is transferred to the IC-vial by using an ultrafiltration syringe filter. It is strongly advised to apply the Metrohm in-line dialysis instead of the ultrafiltration. Samples containing high concentrations of microorganisms are very hard to filter as they clog up the ultra-filtration membrane easily.

Analysis

The standards were injected via the 858 Professional Sample Processor.

Parameters

Flow	0.5 mL/min
Temperature	30 ^{O}C
Standard loop	20 µL
Data acquisition time	30 min

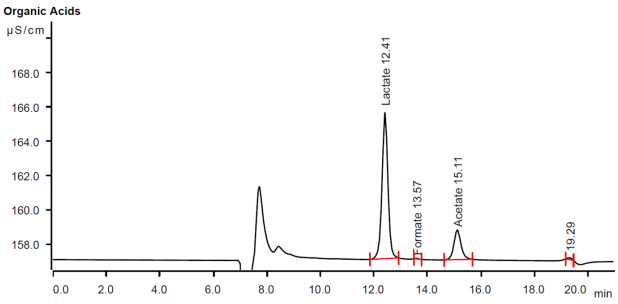
Calculation:

Automatic integration with MagIC Net 3.1 software using peak area for all analytes.

Results

Sample	β Lactate	β Formate	β Acetate
	mg/kg	mg/kg	mg/kg
1	9437	8	1129
2	9121	34	1076
3	9197	46	1070
4	8364	30	794
5	9236	9	861
6	9538	7	1121
7	11555	57	1178
8	11344	33	1223
9	12653	60	1338

Example determination



References

Influence of fermentation time on characteristics of sourdough bread / Brazilian Journal of Pharmaceutical Sciences vol. 49, n. 2, apr./jun., 2013 / Krischina Singer Aplevicz* , Paulo José Ogliari, Ernani Sebastião Sant'Anna

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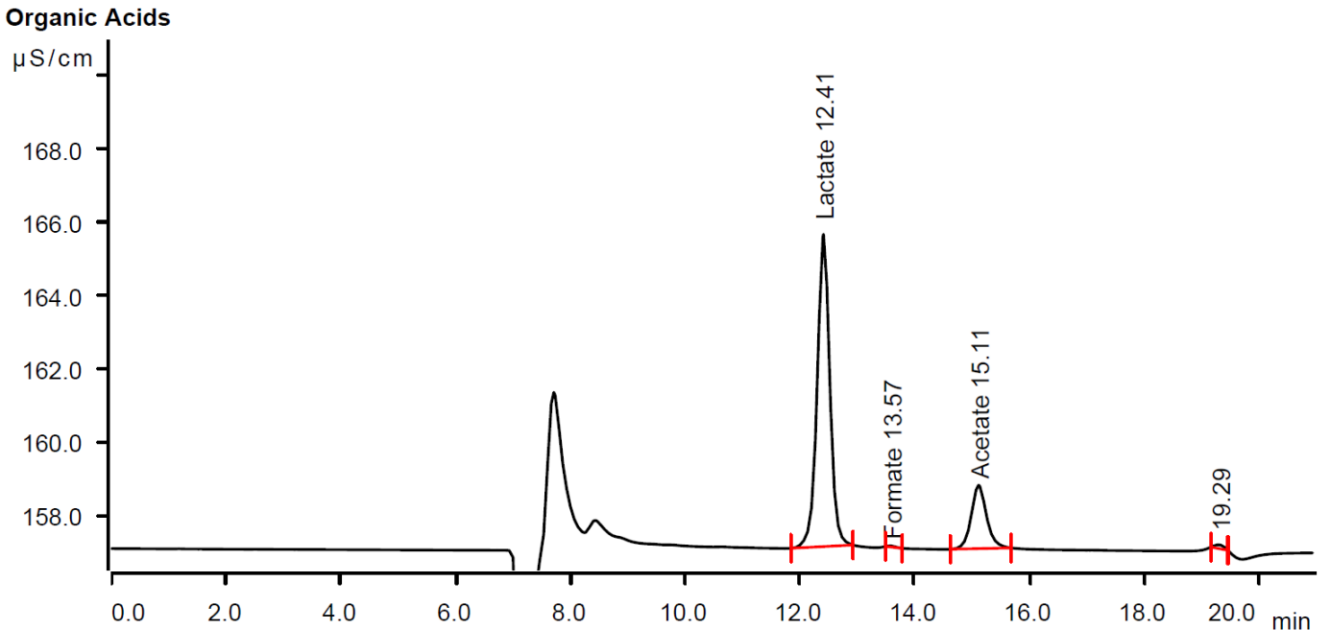
Appendix:

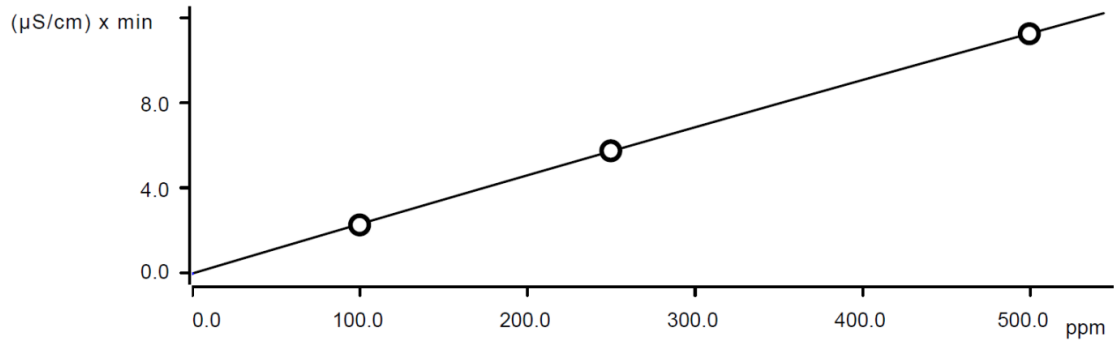
1. Chromatogram
2. Calibration curves

Appendix 1: Chromatogram



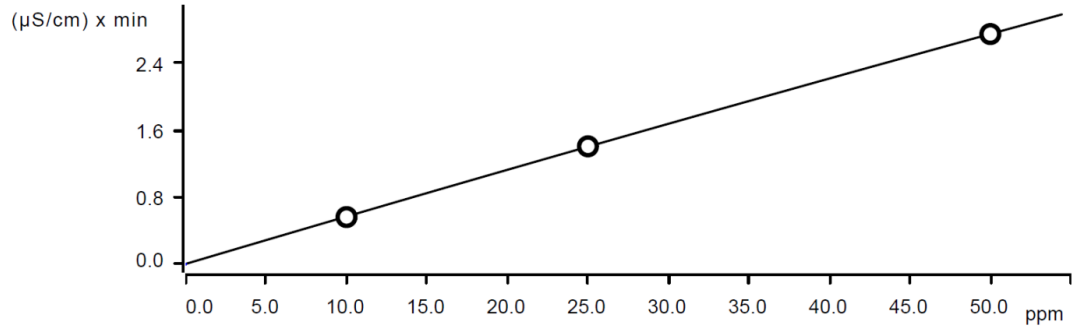
App7endix 2: Calibration curves

Lactate (Organic Acids)



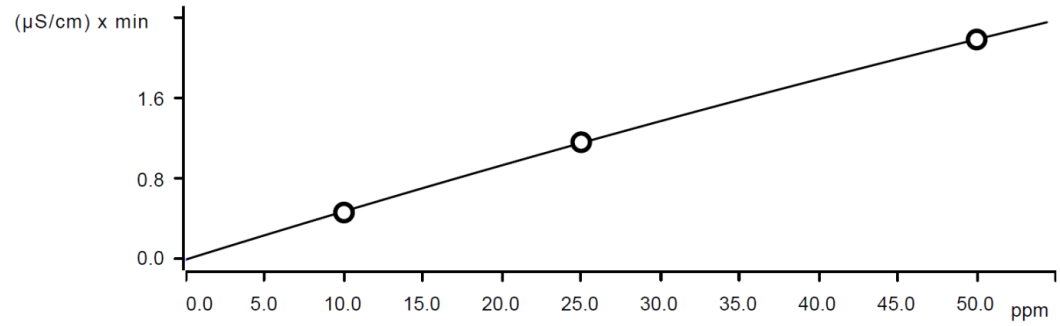
Function: . . . . . *A = 1.16563E-3× Q - 4.28177E-9× Q²*
Relative standard deviation . . . . . *0.708184 %*
Correlation coefficient *0.999975*

Formate (Organic Acids)



Function: . . . . . *A = 2.86261E-3× Q - 1.09356E-7× Q²*
Relative standard deviation . . . . . *0.493722 %*
Correlation coefficient *0.999988*

Acetate (Organic Acids)



Function: . . . . . *A = 2.43320E-3× Q - 2.56479E-7× Q²*
Relative standard deviation . . . . . *1.074973 %*
Correlation coefficient *0.999937*