

Calibration-Free Quantification of Lactic Acid and Lactic Acid Oligomers in Concentrated Aqueous Lactic Acid Solutions Using GC/Polyarc®/FID with Deans Switch

Application Note

Quantifying Unique Compounds

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Abstract

Quantification of compounds for which standards are not commercially available can involve costly synthesis of said compounds, or time-intensive approaches utilizing multiple analytical methods. Such a predicament is encountered when attempting to quantify the different lactic acid oligomers present in aqueous lactic acid solutions of a given total wt% lactic acid. In this application note, a simple, calibration-free method for quantifying lactic acid and its low molecular weight oligomers in an 88 wt% aqueous lactic acid solution is described. The data obtained with this method is compared with data obtained using other analytical methods.

Introduction

The most common detector for gas chromatography (GC) analyses of organic samples is the flame ionization detector (FID). Standard GC/FID analyses require calibrations for each analyte to be quantified because FID response is dependent on the chemical structure of the analyte. Often, standards are not commercially available or not easily synthesized and purified. For example, linear oligomers of lactic acid,

namely the dimer (Dp2), trimer (Dp3), and tetramer (Dp4), shown in **Figure 1**, are not commercially available and not readily synthesized and purified as pure compounds.

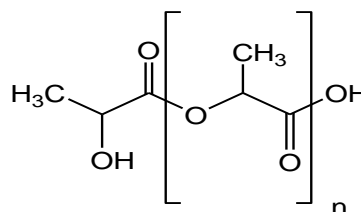


Figure 1. Lactic Acid (n=0) and Lactic Acid Oligomers (n=1,2,3).

It is well known that in concentrated aqueous solutions of lactic acid (>30 wt%), the monomer will undergo self-condensation to produce an equilibrium mixture of monomer and lactic acid oligomers.¹ The higher the wt% lactic acid, the higher the concentration of oligomers produced, and the higher the molecular weight of oligomers possible. The relationship between actual wt% lactic acid in a concentrated aqueous solution and the equilibrium oligomer distribution has been explored by Miller et al. using a combination of analytical methods, including titration, GC/FID/MS (after derivatization with silylation reagent), and HPLC/UV. The results obtained from this detailed study were used to create a thermodynamic model wherein a single parameter was proposed that “accurately represents oligomer distribution and titratable acidity over the full range of lactic acid concentrations”.² Because commercial standards of lactic acid oligomers are not available, the concentrations of such oligomers in concentrated aqueous lactic acid solutions can be approximated using equilibrium data. This in turn allows for the generation of calibration curves when attempting to quantify such oligomers in a given sample, after derivatization with suitable silylation reagent, by GC/FID.

The Polyarc[®] reactor is a catalytic microreactor that combines a catalytic combustion chamber, where all moles of carbon in eluting organic compounds are converted into a stoichiometric number of moles of carbon dioxide with >99.9% efficiency, which are subsequently converted to methane, again with >99.9% efficiency, in a catalytic reduction chamber prior to entry into FID. Since the only carbon-containing molecule entering the FID is methane, the Polyarc[®] reactor system alleviates the need to perform time-consuming calibrations by creating a uniform detector response for truly universal carbon detection.³ This allows for quantification of all carbonaceous compounds of interest in a sample containing an internal standard, of known concentration, in a single injection. In the following work, we apply this new capability to the quantification of lactic acid and lactic acid oligomers in a concentrated aqueous lactic acid solution without calibration. A comparison of results obtained using GC/Polyarc[®]/FID with results obtained from other analytical methods is also provided.

Experimental

All GC/FID and GC/Polyarc[®]/FID analyses were conducted on an Agilent 7890A GC/FID with a Zebron ZB-5HT (30 meter x 0.25 mm i.d.; 0.25 μm film thickness) or a J&W Scientific DB-5MS (30 meter x 0.25 mm i.d.; 0.25 μm film thickness) capillary column used for the separation. The same GC method parameters were used with both setups.

The high polarity and low volatility of lactic acid oligomers adversely impacts the ability to analyze and accurately quantify these analytes directly by GC/FID. The lactic acid monomer and linear lactic acid dimer are amenable to GC/FID analysis, but the chromatographic efficiency is poor with significant peak tailing observed using capillary GC columns of widely varying stationary phase polarity for the separation. To enhance the volatility of the highly polar lactic acid and lactic acid oligomer analytes and improve the chromatographic efficiency of the separation, concentrated lactic acid samples are first dissolved in acetonitrile and derivatized with N,O-bistrifluoroacetamide (BSTFA) with 1% trimethylchlorosilane (TMCS) at 50°C for 30 min. BSTFA is a commonly used silylation reagent that converts all carboxylic acid and hydroxyl groups in a molecule, such as lactic acid and lactic acid oligomers, to corresponding trimethylsilyl (TMS) esters and ethers, respectively. A Deans switch (SGE Analytical Science, 123703) was used to divert excess BSTFA present in derivatized lactic acid samples away from the Polyarc[®] reactor during the analysis. **Figure 2** shows the experimental setup used when analyzing BSTFA-derivatized lactic acid samples, where the Deans switch was used to toggle column flow between FID and Polyarc[®]/FID detection systems. Flow to the Polyarc[®]/FID occurred only after the excess BSTFA had eluted from the column. A solenoid valve (Parker 30CU02LV4) was supplied with 4 psi of N₂ and connected to the switch with a restrictor tube (Vici TSS107, 0.3 m x 0.18 mm i.d) placed between the two outlets.

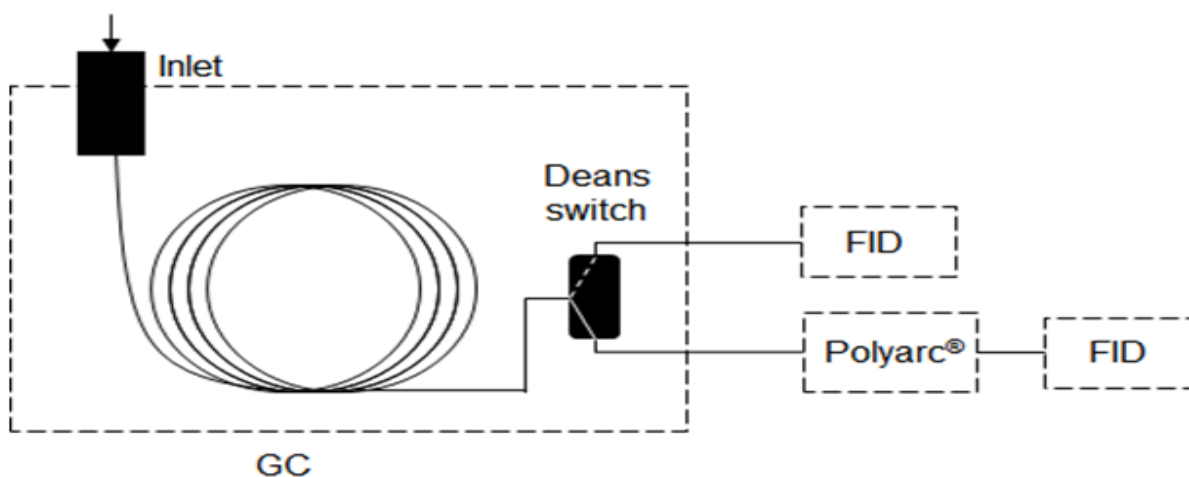


Figure 2. Experimental Setup for Detection by FID or Polyarc[®]/FID

GC conditions

Front inlet	Split/Splitless
Carrier Gas	Hydrogen
Inlet temperature	250 °C
Inlet pressure	10.279 psi
Septum purge flow	3 sccm
Oven	50 °C (2 min), 10 °C/min to 150 °C (0 min), 25 °C/min to 320 °C (2 min)
Column	ZB-5HT (30 m × 0.25 mm × 0.25 µm)
Syringe	10 µL
Injection volume	1 µL

FID conditions

Temperature	335 °C
H ₂	5 sccm
Air	350 sccm
Makeup	45 sccm (N ₂)
Sampling rate	50 Hz

Polyarc® reactor conditions

Setpoint	293 °C
H ₂	35 sccm
Air	5.0 sccm

Deans switch conditions

Makeup	4 psi (N ₂)
Restrictor	Vici TSS107 (0.3 m × 0.18 mm i.d.)

Results and Discussion

When quantifying the concentrations of lactic acid and lactic acid oligomers in concentrated aqueous solutions of lactic acid by GC/FID, a very laborious calibration must be conducted after samples have been derivatized with BSTFA, because authentic standards of Dp2-Dp4 do not exist. Actual calibration curves for Dp2-Dp4 are created based on the equilibrium concentrations of these oligomers in concentrated aqueous lactic acid solutions (e.g., 88 wt%) evaluated internally by capillary electrophoresis.

As shown in Figure 3, GC/Polyarc®/FID analyses of BSTFA-derivatized concentrated aqueous lactic acid solutions were characterized by extensive tailing of the residual, excess BSTFA peak, which provided a sloping baseline where the di(trimethylsilyl) derivative of lactic acid elutes. Not only did this analyte peak elute on a sloping baseline but it also exhibited peak tailing, which contributes to inaccurate peak integration. Minimal tailing was observed for these peaks when

analyzing the same sample by GC/FID. It was hypothesized that residual BSTFA in the sample generates significant silicon dioxide on the combustion catalyst surface within the Polyarc® that impacts combustion efficiency of later-eluting analytes, while also creating adsorption sites that contribute to the observed peak tailing. It was suggested by senior technical staff at ARC that a Deans switch device could be installed in series, prior to the Polyarc®/FID, to resolve the issue with excess BSTFA. The primary application of the Deans switch is in multidimensional GC (MDGC).⁴ For our purposes, the Deans switch would be used to divert early-eluting excess, unreacted BSTFA reagent from entering the Polyarc® reactor, so the performance of the Polyarc® for later-eluting analytes of interest was not negatively impacted. After BSTFA had eluted, the Deans switch would re-establish flow to Polyarc®/FID so later-eluting sample components of interest would be detected.

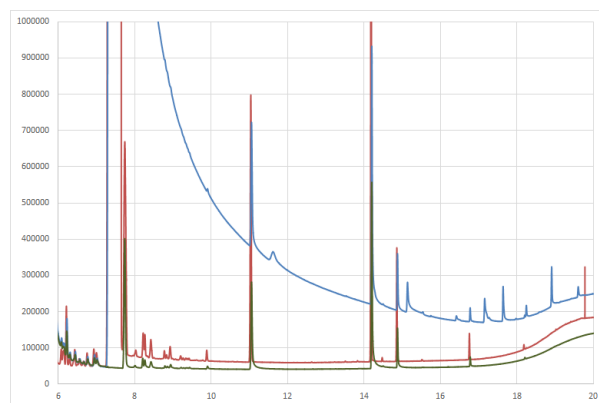


Figure 3. Overlay of chromatograms of a BSTFA-derivatized concentrated aqueous lactic acid solution, corresponding to FID-only (red), Polyarc®/FID (blue), and Polyarc®/FID with Deans switch (green).

Presented in **Table 1** are the concentrations evaluated for lactic acid monomer and lactic acid oligomers, specifically Dp2-Dp4, in 88 wt% solution of ADM lactic acid derivatized with BSTFA, using the GC/Polyarc®/FID with the Deans switch system presented in **Figure 2**. A known concentration of maleic acid internal standard is all that was needed to transform peak areas of analytes, relative to internal standard, into concentrations using a single injection. Results obtained with Polyarc®/FID are compared with values appearing in referenced technical paper, and results obtained by standard GC/FID analysis as well as capillary electrophoresis.

Table 1. Concentrations for lactic acid and lactic acid oligomers in an 88 wt% lactic acid solution

Analyte	Concentration (wt%)			
	Polyarc®/FID	Theoretical ¹	GC/FID ²	Capillary Electrophoresis ³
Lactic Acid	56.5	55.4	56.0	53.2
Dp2	22.6	23.6		26.4
Dp3	6.2	8.0		7.1
Dp4	1.5	2.5		1.3
Total ⁴	86.8	89.5		88.0

¹Taken from Reference 2

²Instrument was calibrated using crystalline lactic acid standard before analysis

³Distribution evaluated internally on 88 wt% ADM lactic acid solution at equilibrium

⁴Sum of concentrations of lactic acid and lactic acid oligomers

Conclusions

The results obtained show good agreement between the Polyarc®/FID calibration-free method and values appearing in a technical paper, as well as other methods of analysis. Proof of concept has thus been demonstrated for the accurate and fast quantification of lactic acid and lactic acid oligomers in samples of interest to those working with aqueous lactic acid or samples containing such oligomers as impurities.

References

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