

ACETALDEHYDE and ISOVALERALDEHYDE (Gas Chromatography)

PRINCIPLE

Isovaleraldehyde (IVA) and acetaldehyde are released from the syrup with the aid of dilute phosphoric acid and heat. The liberated compounds are then measured using a headspace gas chromatographic system equipped with a flame ionization detector. Isovaleraldehyde and acetaldehyde results are reported at 11% D.S. for finished sweeteners. It is important to keep the headspace volume the same or it will have an effect on the final concentrations. Because of this, the volume of the preparation vials is important to maintain.

SCOPE

This method is applicable to the analysis of finished HFCS, 95 Dextrose, and most starch hydrolyzates (Note 1).

SPECIAL APPARATUS

1. Single column gas chromatograph equipped with a flame ionization detector and headspace autosampler (Note 2).
2. Capillary GC Column, Supelco Supel-Q Plot fused silica capillary column (30m, 0.53mm ID), or equivalent.
3. Sample vials (20 mL), caps, seals and crimping tools.
4. Repipettor capable of reproducibly delivering aqueous solutions to within $\pm 0.1\%$.
5. Analytical balance capable of measuring 0.0001 g.
6. Transfer pipettes.
7. Steam or water bath set at boiling.

ACETALDEHYDE and ISOVALERALDEHYDE — continued**REAGENTS**

1. Ultra-pure filtered water (Note 3).
2. Phosphoric Acid Solution: In a 1 L volumetric flask add 500 mL of Ultra-pure water. Then add 12.171 g of concentrated phosphoric acid (85%). Mix thoroughly. When mixed, fill to the mark with Ultra-pure water and mix.
3. Blank syrup standard (Note 4).
4. Isovaleraldehyde spiking solution: Accurately weigh 107 mg of 98% isovaleraldehyde into a clean 100 mL volumetric flask. Dilute to volume with Ultra-pure water. This is *Stock I* (1,050 ppm). Pipette 1.0 mL of Stock I into a clean 100 mL volumetric flask. Dilute to volume with Ultra-pure water. This is *Stock II* (10.5 ppm). Pipette 1.0 mL of Stock II into a clean 100 mL volumetric flask. Dilute to volume with Ultra-pure water. This is the *Spiking Solution* (105 ppb).
5. Acetaldehyde spiking solution: Purchase a certified solution of 5.62 ppm 4,6-0-ethylidene-D-glucose or weigh 56.2 mg of 4,6-0-ethylidene-D-glucose (Aldrich catalogue #E3275-4 or Pfaltz and Bauer catalogue #E11450) into a beaker, and transfer to a one liter volumetric flask using Ultra-pure water, dilute to volume. Then prepare a 5.62 ppm solution by diluting 100 ml of this solution to one liter with Ultra-pure water. Store capped in the refrigerator.

GC AND HEADSPACE PARAMETERS**GC Parameters**

1. Injector port temperature: 150°C
2. Detector temperature: 250°C
3. Oven temperature ramp profile:

ACETALDEHYDE and ISOVALERALDEHYDE — continued

Table 1 – Oven Temperature Ramp Profile

Step	Ramp Rate °C/min	Next Temp (°C)	Hold Time (min)	Run Time (min)
Initial		50	0	0
1	7.5	190	1.5	20.2

4. Gas pressure: 45 mL/min H₂, 450 mL/min air.

Headspace Sampler Parameters for capillary column

1. Carrier gas (He): ≈20mL/min at 12 psi. This will provide passive pressure/flow to the GC.
2. Needle temperature: 125°C
3. Transfer line temperature: 125°C
4. Oven temperature: 80°C
5. Pressurization time: 0.7 minutes
6. Injection time: 0.3 minutes
7. Thermo time: 60 minutes
8. GC cycle time: 23.5 minutes
9. Needle withdraw time: 0.5 minutes

PROCEDURE

Standard Preparation:

1. Prepare five vials with caps.
2. Weigh 1.5000 ± 0.0050 g of blank syrup into each vial.

ACETALDEHYDE and ISOVALERALDEHYDE — continued

3. Pipette purified water, the spiking solution of acetaldehyde and IVA, and phosphoric acid into each vial in the quantities shown in Table 2. The phosphoric acid is added last into each vial.
4. Immediately cap the vial and crimp it tightly. These vials should not be left for any period of time with stock solution in them unless they are capped.

Sample Preparation:

For 42 FX samples, weigh 1.6000 ± 0.0050 g of sample into a vial, and to this pipette 7.9 mL of deionized water, then 1.0 mL of phosphoric acid; cap and crimp tightly. Table 2 summarizes the 42 FX sample preparation.

For 55 FX samples, weigh 1.5000 ± 0.0050 g of sample into a vial, and to this pipette 8.0 mL of deionized water, then 1.0 mL of phosphoric acid; cap and crimp tightly. Table 2 summarizes the 55 FX sample preparation.

All vials are then mix thoroughly and then placed in a boiling water bath for 60 minutes. The samples are then transferred to the headspace analyzer and are ready to be analyzed on the GC.

CALCULATIONS

Results are calculated using a 5-point calibration curve. The calibration standards consist of the blank syrup and each of the four prepared standards. The blank is subtracted from each standard and then a line equation is created with the corrected standards (x-axis is the standard concentration, the y-axis is the peak area). The concentration of components, expressed in parts per billion of 11% D.S. finished product samples, can be determined by the following equation:

$Y = mx + b$ where:

Y = peak area

x = concentration of acetaldehyde or IVA in ng/g (ppb) of 11% ds syrup

m = slope

b = y-intercept

ACETALDEHYDE and ISOVALERALDEHYDE — continued**Tables**

Table 2 - Combined IVA/ Acetaldehyde GC Method Standards and Sample Composition

	Syrup (g)	Purified Water (mL)	Acetaldehyde Spiking Solution (ml)	IVA Spiking Solution (ml)	Phosphoric Acid (ml)
Blank	1.5	8.0	--	--	1.0
Std #1	1.5	7.6	0.2	0.2	1.0
Std #2	1.5	7.2	0.4	0.4	1.0
Std #3	1.5	6.8	0.6	0.6	1.0
Std #4	1.5	6.0	1.0	1.0	1.0
55 FX	1.5	8.0	--	--	1.0
42 FX	1.6	7.9	--	--	1.0

Table 3 - Combined IVA/ Acetaldehyde GC Method Standard Concentrations

Standard	Acetaldehyde Concentration (ppb)	IVA Concentration (ppb)
Blank	0	0
#1	22.9	2
#2	45.7	4
#3	68.6	6
#4	114.3	10

NOTES AND PRECAUTIONS

1. Sulfur dioxide levels in excess of 10 ppm may interfere.
2. Different GC's and headspace units may use different parameters.
3. High quality water (over 18 mega-ohms) must be used to prepare all reagents and blanks to avoid contamination.
4. The blank syrup should have low or non-detectable levels of IVA and acetaldehyde.
5. This method typically reports slightly higher acetaldehyde results than CRA Method E-1.
6. The variability of IVA and acetaldehyde results obtained from this method are similar and typically better than those reported from methods ISBT 12.0 and CRA method E-1 respectively.

ACETALDEHYDE and ISOVALERALDEHYDE — continued

REFERENCES

1. CRA Acetaldehyde Method E-1. CRA Method E-1 “Acetaldehyde”:
Revision Dec. 19, 2006.
2. ISBT Isovaleraldehyde Method 12.0. “Isovaleraldehyde (IVA)”:
Revision March 2003.