

# Courtesy Copy

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## Volatile Acids in Wines Using An Automated Segmented Flow Analyzer

### Scope and Application

Volatile acidity in wine may be indicative of spoilage. Wines may be screened for acetic acid by GC and those over the legal limit are confirmed by this method. Acetic acid is a natural fermentation product with levels ranging from below 200 ppm to above 1000 ppm in blenders and to above 4000 ppm in flavors and wine vinegar. This method may be used in the analysis of wine.

#### Regulatory Tolerances:

Volatile Acidity, calculated as Acetic Acid and exclusive of sulfur dioxide (27CFR4.21) —

0.14 % by volume max. in **red wine** when starting brix  $\leq$  28

0.17 % by volume max. in **red wine** when starting brix is  $>28$

0.12 % by volume max. in **white wine** when starting brix  $\leq$  28

0.15 % by volume max. in **white wine** when starting brix is  $>28$

### Levels and Limitations

Analyte	Detection Limit (g/100mL)	Quantitation Limit (g/100mL)	Linear Range (g/100 mL)	Interferences
Acetic Acid	0.001	0.012	0.012 – 0.18	None

### Equipment

#### Instrumentation:

Segmented Flow Analyzer equipped with volatile acids cartridge, Astoria-Pacific or equivalent

#### Glassware and Supplies:

Class A 1L, 500 mL, and 100 mL volumetric flasks

Volumetric pipets as needed

Other standard laboratory glassware—beakers, erlenmeyers, etc.

### Reagent and Standards

#### Reagents:

**VA Reagent kit part no. 80-9100-13C-Astoria Pacific OR**

Sulfuric Acid (FW 98.08) – **ACS Grade**

Antifoam B – **ACS Grade**

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Volatile Acid (VA) Color Reagent Kit (part no. 80-9150-04) – Astoria-Pacific  
Sodium Acetate (FW 82.04) – **ACS Grade**  
Triton TX-10, 4 oz (part no. 90-0760-04) – Astoria-Pacific

***The following are not included in the kit and should be purchased separately:***

***30% Hydrogen Peroxide (FW 34.01), Aqueous Solution – ACS Grade  
Acetic acid, 99% or better- ACS Grade***

Deionized (DI) water – 18 mega ohm or better  
***NBA standard Level 2 (see TM 200)***

## **Reagent and Sample Preparation and Handling:**

***(Also refer to instructions provided with VA Reagent Kit if used)***

### 0.91 M Sulfuric Acid Distillation Reagent (1L) see Safety Notes

1. Add 50 mL of sulfuric acid to approximately 800 mL in a 1 L flask.
2. Cool and dilute to volume.
3. Mix well.
4. Store for up to 3 months.

### Sodium Acetate Reagent Solution (100 mL)

1. Dissolve 1.639g sodium acetate in 80 mL DI water in a 100 mL volumetric flask.
2. Dilute to volume and mix well.
3. Store for up to 4 months.

### Peroxide/Acetate Reagent (100mL)

1. Add 2 mL 30% hydrogen peroxide, 1 mL sodium acetate solution and 2-3 drops antifoam B to approximately 80 mL DI water in a volumetric flask.
2. Dilute to volume and mix well.
3. Prepare fresh daily

### Sampler Wash

1. Deionized water

### Color Reagent

1. Add 100 mL VA color reagent to 800 mL DI water contained in a 1L flask.
2. Dilute to volume and mix well.
3. Add 2 mL Detergent TX-10.
4. Store refrigerated for up to 3 months.

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## Start-up/Shutdown Solutions

1. Distillation and Peroxide lines: DI water
2. Color reagent line: Add 2 mL detergent TX-10 to 1L DI water in a flask and bring to volume. Store for up to 3 months.

## Stock Calibrant 1.2 g/100 mL as acetic acid (500 mL)

1. Dissolve 8.195 g sodium acetate in approximately 400 mL DI water in a 500 mL volumetric flask.
2. Dilute to volume and mix well.
3. Store refrigerated for up to 4 months.

## Working Calibrants

1. Transfer 1, 6, 10 and 15 mL of the stock calibrant (1.2 g/100 mL) into 100 mL volumetric flasks containing approximately 50 mL DI water.
2. Quantitate to volume with DI water.
3. These will correspond to the following concentrations (g/100 mL): 0.012, 0.072, 0.12, and 0.18. Prepare daily.

## Sample Preparation

1. Generally, for wine samples, no sample prep is needed.
2. If particulate matter is present in the wine, filter.
3. Degas carbonated wines before analysis.

## Procedures

1. Follow instrument manufacturer's operating guidelines to set up the instrument.
2. Check for stable baseline, ensure that bubble spacing and size are consistent before proceeding with analysis.

## Quality Control

1. Run the four working calibrants plus a DI water blank, which will represent the zero calibration point.
2. Determine the **regression of the 3<sup>o</sup> polynomial equation**
3. Analysis should only proceed if the correlation coefficient is greater than 0.99.
4. Run control standards after every 6-7 samples, insert blanks to check for carryover after every suspected high sample.
5. **Run NBA 0.1 g/100mL standard (NBA Level 2) or freshly prepared 0.1 g/100 ml acetic acid standard as one of the controls.**

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- 6. Run a LCS with every batch. See logbook for specifications.**
- 7. The instrument also carries out an autowash (blank) automatically after every 10 samples and reports carryover results.**
- 8. Any samples above the range of the standard curve need to be diluted and retested.**

## Troubleshooting

**If the regression equation is bad, or control standards do not work: check the following**

- 1. Improper equilibration of segmented flow analyzer –check bubble size, shape, distance**
- 2. Blocked tubings**
- 3. Standards not prepared correctly**
- 4. Standards not set up correctly (tubes out of order)**
- 5. Old reagents/standards**

**If all else fails, consult with primary analyst or contact vendor.**

## Sources of Uncertainty

- 1. Dilution errors during standard preparation**
- 2. Errors in reagent preparation**
- 3. Instrument not working correctly- see troubleshooting**
- 4. Old reagents**
- 5. Carryover from previous sample**

## Calculations

Use the operating system software (a third degree polynomial equation is used) to calculate volatile acidity as acetic acid.

## Reporting Results

Report the results as follows:

<b>Component</b>	<b>Sample Type</b>	<b>Units</b>	<b>Precision</b>	<b>Format</b>
Acetic Acid	All	g/100mL	2 decimals	x.XX

## Safety Notes

Consult the MSDS for any chemicals used that are unfamiliar. All chemicals should be considered hazardous - avoid direct physical contact.

Sulfuric acid releases a great amount of heat when mixed with water. Wear safety glasses, gloves, apron, and work in the hood.

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## References

AOAC Methods XVII, 964.08, 978.12

Astoria-Pacific International Astoria Analyzer Volatile Acidity Method 305-A470-A00(Method A470).

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## Required Training, Certification and Re-certification.

1. In-house training by a certified chemist in segmented flow analysis and software(fas-pac) operation. Training on segmented flow analysis (in-house or vendor provided).
2. Initial certification is achieved by running **LCS** samples in triplicate **within 3 SD and one high and one low level spike (ex. 0.04 g/100 mL and 0.1 g/100mL). Spike recovery results should be within 10% of expected values.**
3. Annual proficiency testing.