

# Quantification of Effects of Common Variables on Determination of Volatile Acidity in Wine

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




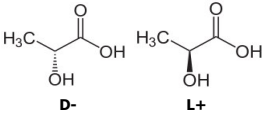
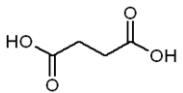
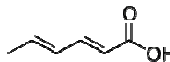
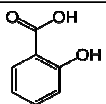
# Background

Volatile Acidity (VA) in wines is a regulated component, a microbiological monitor, and a sensory quality indicator. Although many acids in wine are volatile to varying degrees, the volatile acids of interest in this analysis are more specific than the terminology suggests, and should more accurately be described as the volatile short chain fatty acids (Table 1).

Many published methods describe a steam distillation and titration method for quantifying these acids in wine; they differ in many key points (Table 2 ).

Reported VA acetic acid recoveries relative to acid-specific tests range from 59 to 118%. Industry performance data from Collaborative Testing Services (CTS) shows a coefficient of variation for VA ranging from 10 to 30% with an apparent effect of wine type (Figure 1), perhaps due to pH, lactic, SO<sub>2</sub>, CO<sub>2</sub>, or sorbate levels.

# Table 1 Volatile Acids

The Volatile Acids of Wine and Their Relationship to "Volatile Acid" Analysis								
General Category		Specific Acid	MW	Formula	Structure	Range in wine	pKa	B.P. °C
Volatile Fatty Acids	Primary VA Component	Acetic	60.1	C <sub>2</sub> H <sub>4</sub> O <sub>2</sub>		0.1 to 1.4 g/L <sup>1</sup>	4.76	117.9
	Secondary VA Components (short chain fatty acids)	Formic	46.0	CH <sub>2</sub> O <sub>2</sub>		2.4 to 89 mg/L white <sup>3</sup> 3.3 to 22 mg/L red <sup>3</sup>	3.75	100.7
		Propionic	74.1	C <sub>3</sub> H <sub>6</sub> O <sub>2</sub>		0.1-6.6 mg/L whites <sup>3</sup> trace-13.9mg/L reds <sup>3</sup>	4.87	141.0
		Butyric	88.2	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>		0.3 to 3.1 mg/L whites <sup>3</sup> 0.3 to 2.9 mg/L reds <sup>3</sup>	4.83	163.5
		Valeric	102.1	C <sub>5</sub> H <sub>10</sub> O <sub>2</sub>		0 to 6 mg/L	4.82	186.1
Other Volatile Acids (not considered VA proper)	Non VA Microbial Components	Lactic D(-) = hexose fermentation byproduct; L(+) = hexose byproduct AND malic acid decarboxylation byproduct	98.1	C <sub>3</sub> H <sub>6</sub> O <sub>3</sub>		D(-) 0.1 to 1 g/L <sup>2</sup> 0.1 to .5 g/L <sup>3</sup> L(+) 0.3 to 6 g/L <sup>2</sup> 0.1 to 6 g/L <sup>3</sup>	3.86	D(-) 103 L(+) 119 DL 122
		Succinic	118.1	C <sub>4</sub> H <sub>6</sub> O <sub>4</sub>		0.5 to 7.5 g/L <sup>2</sup> 0.5 to 2 g/L <sup>3</sup>	4.21 5.64	235
	Non VA Other Component	CO <sub>2</sub> (dissolved as carbonic)	44.0	CO <sub>2</sub>	CO <sub>2</sub>	0 to 12 g/L (to 3.92 in still wines <sup>2</sup> )	6.37* 10.4	
	Non VA Preservatives	SO <sub>2</sub> (dissolved as sulfurous)	64.0	SO <sub>2</sub>	SO <sub>2</sub>	0 to 350 mg/L <sup>1</sup>	1.8 7.2	
		Sorbic	112.1	C <sub>6</sub> H <sub>8</sub> O <sub>2</sub>		0 to 300 mg/L <sup>1</sup>	4.76	228
		Salicylic	138.1	C <sub>7</sub> H <sub>6</sub> O <sub>3</sub>		Trace. No legal additions <sup>1</sup>	2.97	211

<sup>1</sup> (2007). Alcohol, Tobacco Products and Firearms. Code of Federal Regulations. 27 CFR 24.246: 637-641.

<sup>2</sup> Radler, F. (2002). Yeast-Metabolism of Organic Acid. Wine Microbiology and Biotechnology. G. H. Fleet. New York, Taylor and Francis.

<sup>3</sup> Amerine, M. A. and C. S. Ough (1980). Methods for Analysis of Musts and Wines. New York, John Wiley & Sons.

# Table 2 VA Methods

Method	CO2 Removal		SO2 control		pH adjustment	mLs	mLs	mLs	distillate to sample ratio	Precision g acetic/L		
	Sample	Distillate	Sample	Distillate		Sample Volume	Rinse Volume	Distillate Volume		error	repeatability	reproducibility
Amerine and Ough 1980	option <sup>1</sup>	option <sup>1</sup>	option <sup>1</sup>	YES	NO	10	none listed	100	10	---	---	---
AOAC Method 928.12 (HgO) 1979	NO	YES	HgO	NO	HgO	10	5 mLs H2O, 2 mL HgO, 5 mLs H2O	100	10	---	---	---
AOAC Method 940.19 (Ba(OH)2) 1988	? <sup>2</sup>	NO	Ba(OH)2	NO	H2SO4	50 <sup>3</sup>	5 mLs H2O	300	12	---	---	---
AOAC Method 964.08 1965	YES	NO	NO	NO	NO	25	5 mLs H2O	300	12	---	---	---
Cottrell et al 1985	YES	NO	H2O2	NO	NO	10	1 mL 1% H2O2, 15 mLs H2O	200 <sup>4</sup>	20	---	---	---
Ebeler 2003	YES	NO	H2O2	NO	NO	10	15 mLs H2O	100	10	---	---	---
Iland et al 2004	YES <sup>5</sup>	NO	H2O2	NO	NO	10	"small amount H2O"	100	10	---	---	---
Jacobson 2006	YES	NO	H2O2	NO	no	10	1 mL of 0.3% H2O2	100 <sup>6</sup>	10	---	---	---
OIV method	YES	NO	NO	YES	H2TA	20	none listed	250	12.5	---	0.04	0.08
TTB	"freshly prepared sample"	NO	option <sup>1</sup>	option <sup>1</sup>	NO	10	5 mLs H2O	100	10	---	---	---
Vinquiry	YES	NO	NO	YES	NO	10	"few mLs" H2O	125	12.5	---	---	---
Zoecklein et al 1994	option <sup>1</sup>	option <sup>1</sup>	NO	YES	NO	10	"rinse"	100	10	0.05	---	---

<sup>1</sup> effects discussed in text & various alternate treatments recommended

<sup>2</sup> not specified, but must occur due to sample processing

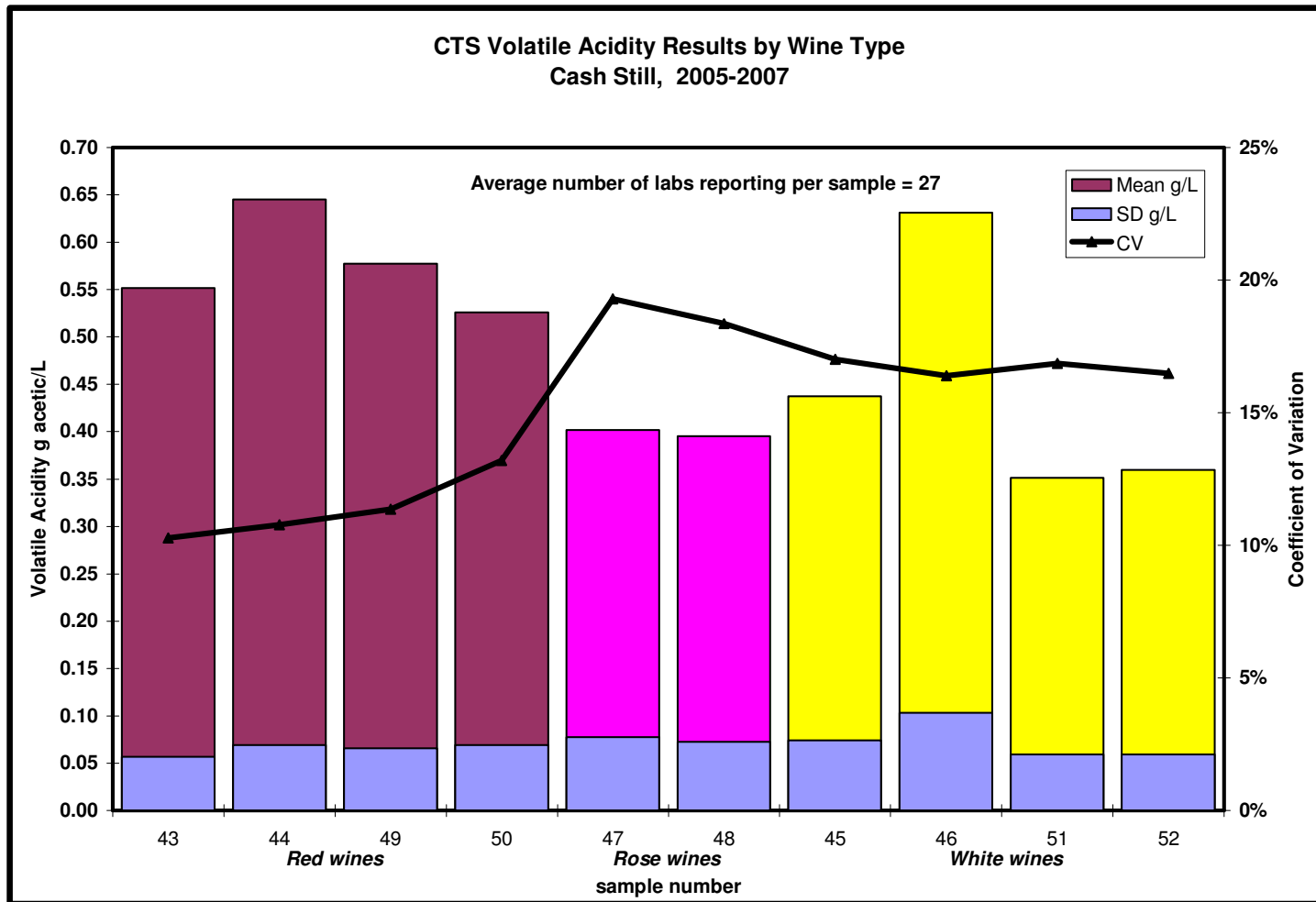
<sup>3</sup> 50 mLs of 50% sample= 25 mLs sample equivalents

<sup>4</sup> found 85% recovery at 100 mLs, 95% at 200 mLs

<sup>5</sup> specifies to only use vacuum, not heat

<sup>6</sup> specifies disregarding sample if >110 mLs

# Figure 1 CTS Results



# Documented Effects

Sulfur dioxide and sorbate levels interfere with VA results. Their control has been documented, but perhaps not practiced.

Researchers have reported only partial acetic acid recovery at the 100 mL (10x) distillate volume, large effects of pH between 3.5 and 4.5, and lactic carryover of at least 1.9%. Carbon dioxide and lactic acid effects on the steam distillation/titration method have been troubling chemists since 1940, but the method has not been improved.

## Evaluating Chemical and Operational Effects

Factorial design (Table 3) testing the impact of chemical and physical variables showed primary effects (Figure 2). Acid dissociation is significant at this range of pHs. The large impact of secondary fatty acids presumes they are present. Operational parameters of the still apparently influence the effects of lactic acid and CO<sub>2</sub> and were further investigated.

# Table 3 Variables Tested

Variable	In 13.5% Ethanol Solution saturated with KHTa	Specific	Low Level (-1)	Medium Level (0)	High Level (+1)	units
1	Primary VA Component	Acetic acid	0.00	0.70	1.40	g/L
2	Secondary VA Components	Butyric acid	0.0	1.5	3.0	mg/L
		Propionic acid	0.0	7.0	14.0	mg/L
		Formic acid	0.0	50.0	100.0	mg/L
3	Non VA Microbial Components	Lactic acid	0	3	6	g/L
		Succinic acid	0	4.5	9	g/L
4	Non VA Other Components	CO <sub>2</sub>	0	2	4	g/L
5	Chemical Variable	pH	3.00	3.75	4.50	pH units
6	Physical Variables	Boiling rate by distillate accumulation*	70	85	100	% line voltage
7		Distillate temperature	5	20	35	Centigrade
8		Distillate volume	10	12.5	15	multiple of sample size

\*Research and Development Glass Brand "Watersaver" 550 watt immersion coil heater AS PURCHASED:

=>100% line voltage, boiled 25 mL H<sub>2</sub>O sample to 150 mLs in ~12 minutes

**Coil length was reduced by 20% for this trial to allow a faster maximum boiling rate.**

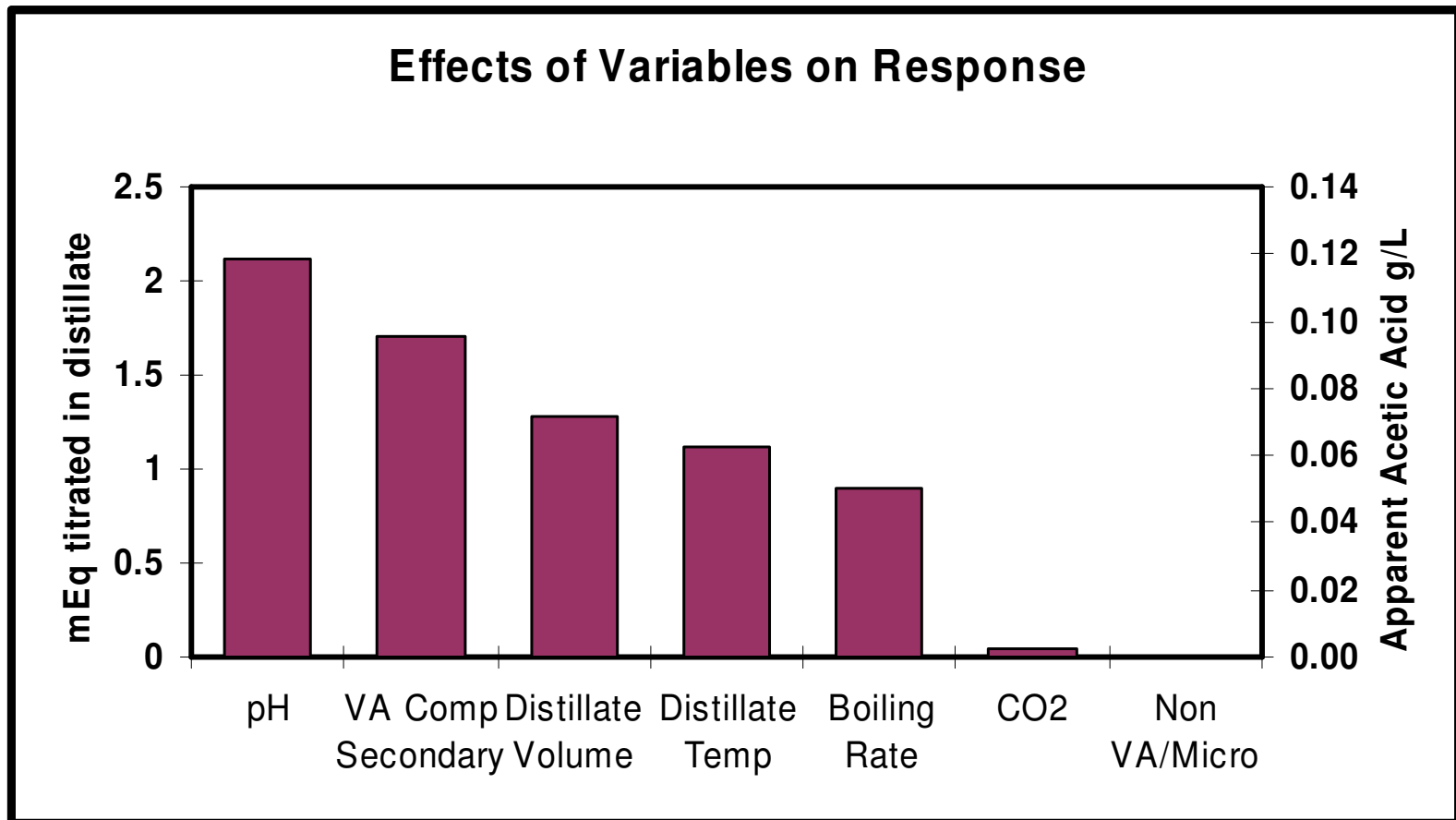
Control of the rate was obtained by adjusting line voltage to this shorter coil:

=>100% line voltage: 25 mL H<sub>2</sub>O sample to 150 mLs in ~7.5 minutes (10mL sample to 100 mL distillate in ~5 min)

=> 85% line voltage: 25 mL H<sub>2</sub>O sample to 150 mLs in ~10.5 minutes (10mL sample to 100 mL distillate in ~7 min)

=> 70% line voltage: 25 mL H<sub>2</sub>O sample to 150 mLs in ~17 minutes (10mL sample to 100 mL distillate in ~12 min)

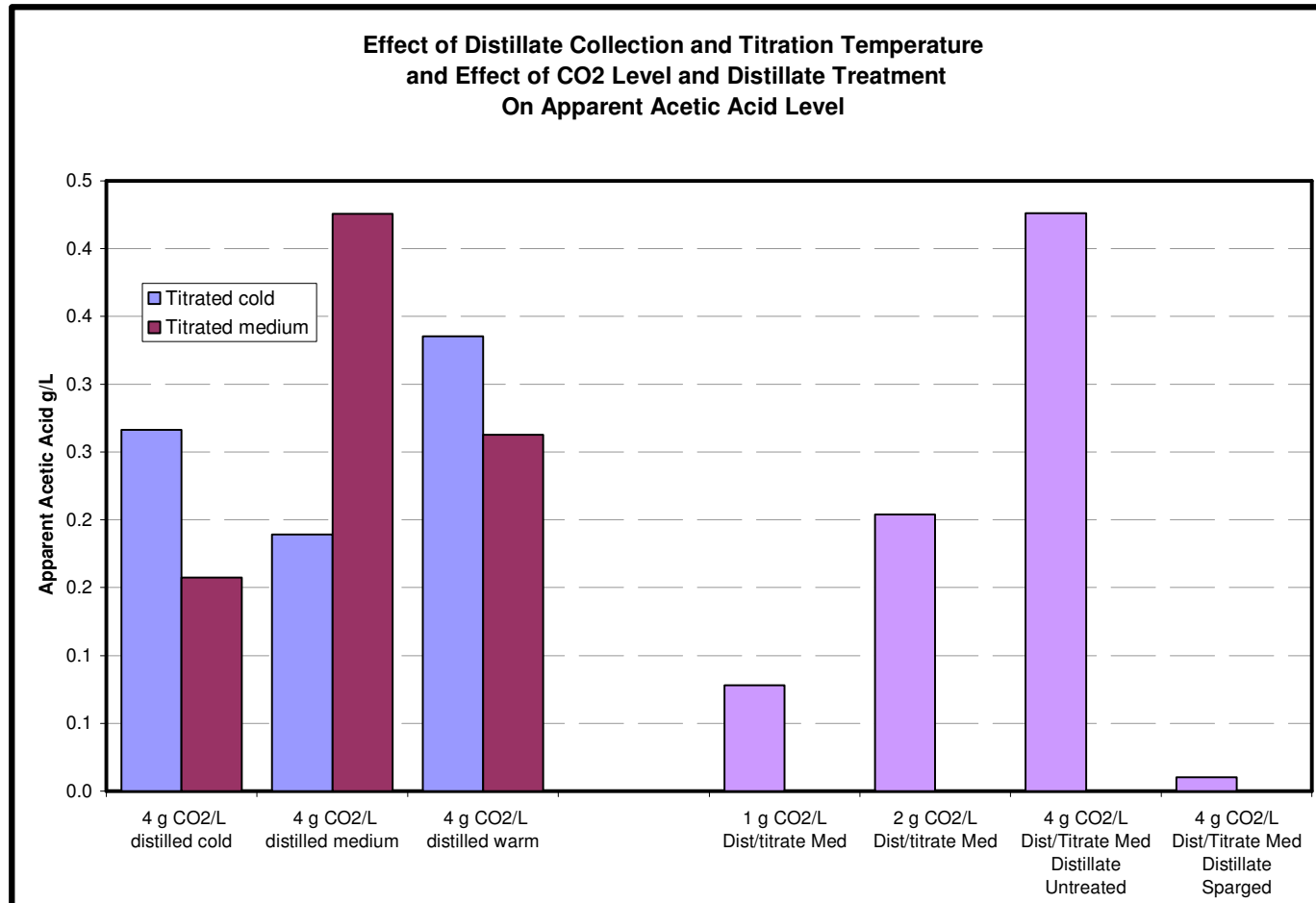
# Figure 2 Results



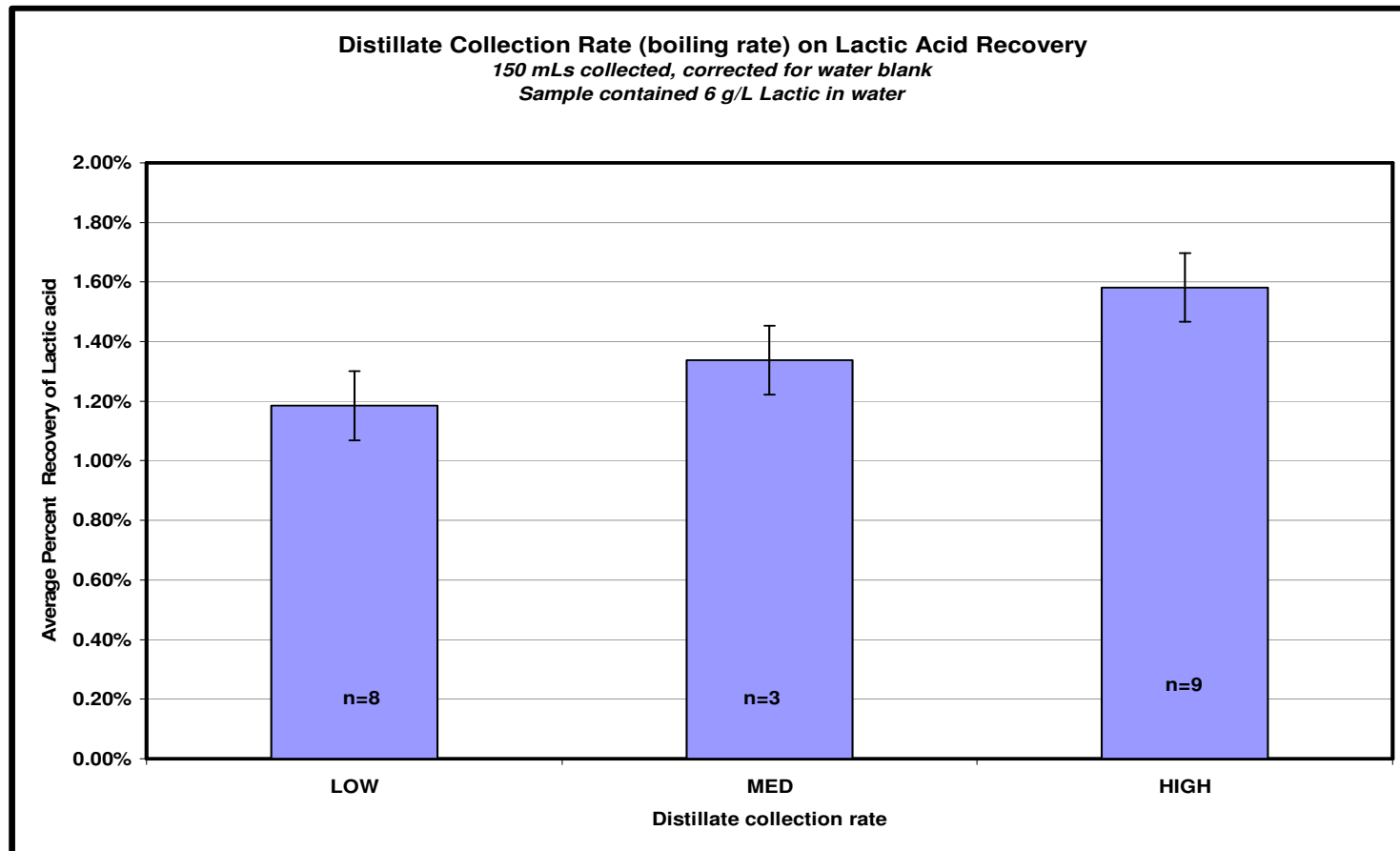
# Evaluating Chemical and Operational Effects

Lactic acid recovery increased with increasing boiling rates, and averaged lower than published data (Figure 3). CO<sub>2</sub> has a strong effect if it persists into the distillate (Figure 4), and the chemistry and kinetics of its solubility, acid equilibrium, and rate of hydration are not trivial. Complex samples which are not degassed prior to distillation may be difficult to degas with sparging (Figure 5). Titrations taking advantage of the difference in CO<sub>2</sub> and fatty acid pK<sub>a</sub>'s may be the best method for quantifying CO<sub>2</sub> in the distillate.

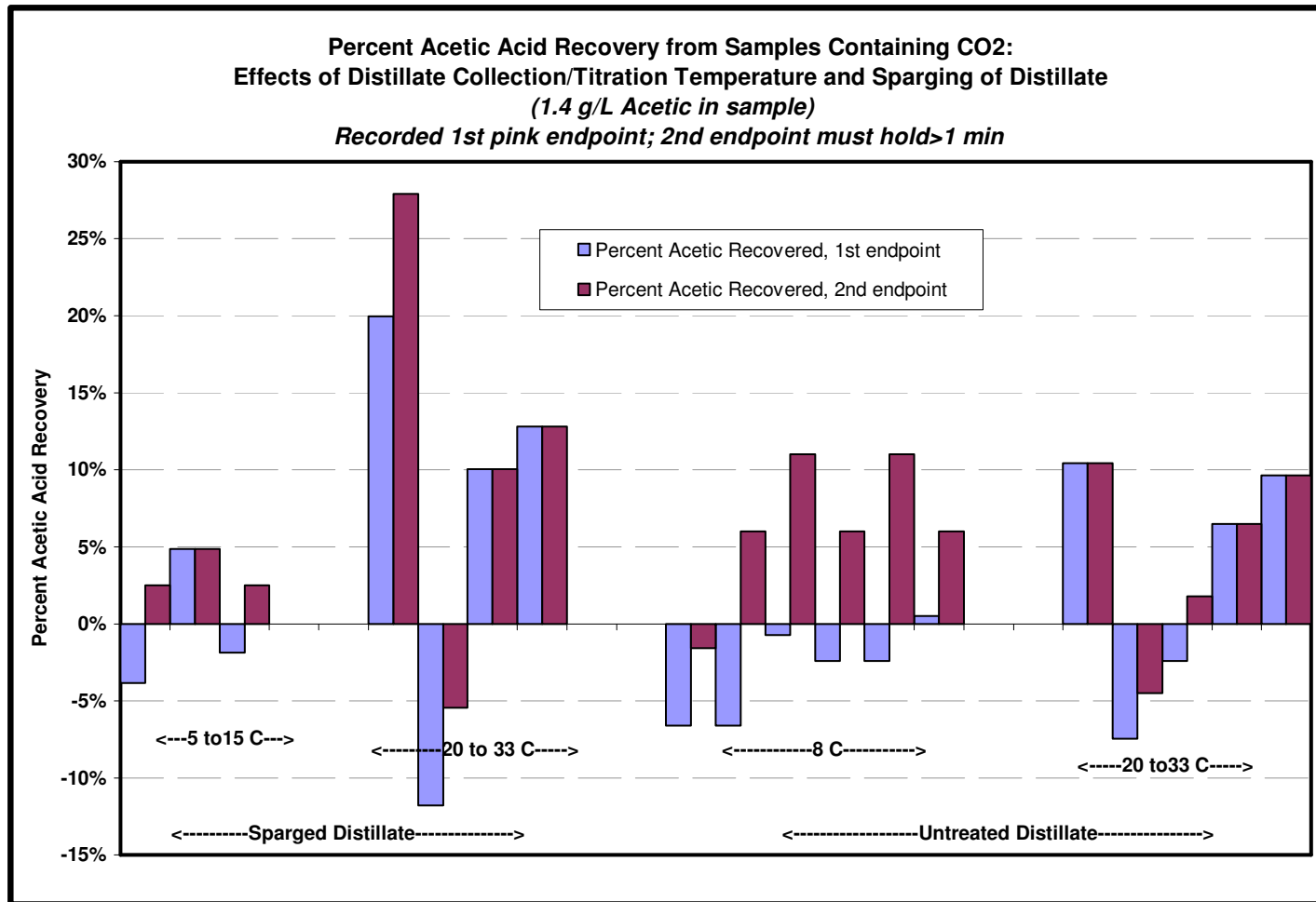
# Figure 4 CO2 Effects



# Figure 3 Distillation rate



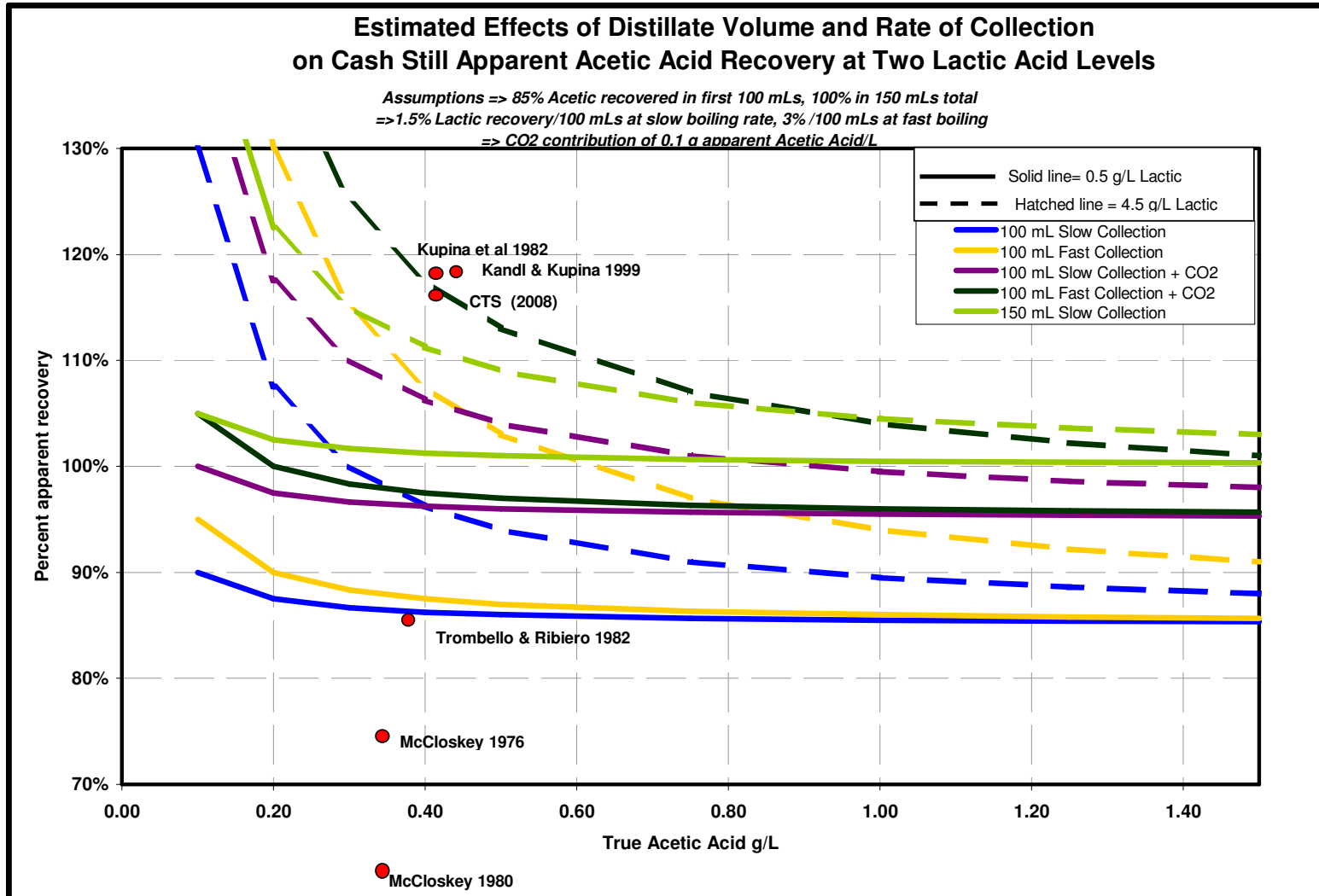
# Figure 5 Acetic Acid with CO2



# Conclusion

Distillate volume, boiling rate, lactic acid level, pH, and a contribution from dissolved CO<sub>2</sub> may explain the wide range of results reported using the Cash still (and its variants) for volatile acid analysis in wine relative to acetic acid levels (Figure 6).

# Figure 6 Summary of Effects



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