Analysis of Pinot Noir Wines by HS-SPME GC/Q-TOF: Correlating Geographical Origin with Volatile Aroma Profiles

Philip L. Wylie[1], Anna K. Hjelmeland[2], Ron Runnebaum[3] & Susan E. Ebeler[3]

1) Agilent Technologies, Wilmington, DE 19808
phil_wylie@agilent.com

2) Agilent Technologies, Santa Clara, CA

3) Department of Viticulture and Enology
University of California, Davis, CA

Outline

Larger Pinot Noir study objectives

HS-SPME GC/Q-TOF sub-study

HS-SPME method

GC/Q-TOF method

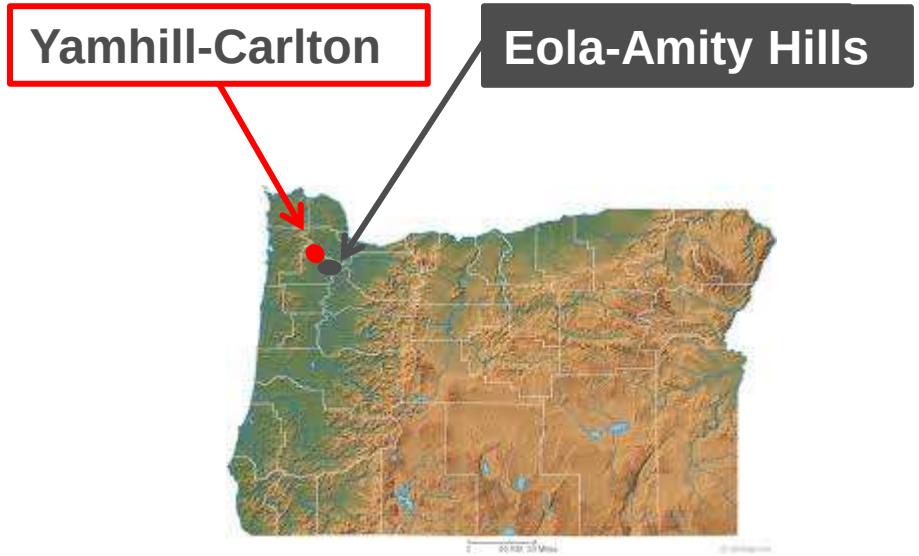
HS-SPME GC/Q-TOF Results

Statistical results

Larger Study Supported by Jackson Family Wines

- Obtain *Pinot noir* grapes from 15 different vineyards (2015)
  - Same grape clones
  - Same root stock (10 vineyards)
  - Different soils and microclimates
- Deliver grapes to UC Davis winery
- Make four replicate wines from each vineyard
  - Same enological practices used for all wines
- Analyze all wines to see how soil & microclimates affect wine
  - **Volatile Analysis- HS-SPME GC-qTOF (comparing to GC-MS acquired data)**
  - Elemental Analysis- ICP-MS
  - Sensory Analysis- Descriptive Analysis
  - Polyphenolic Analysis- LC-DAD

American Viticultural Areas in CA and OR Providing Grapes

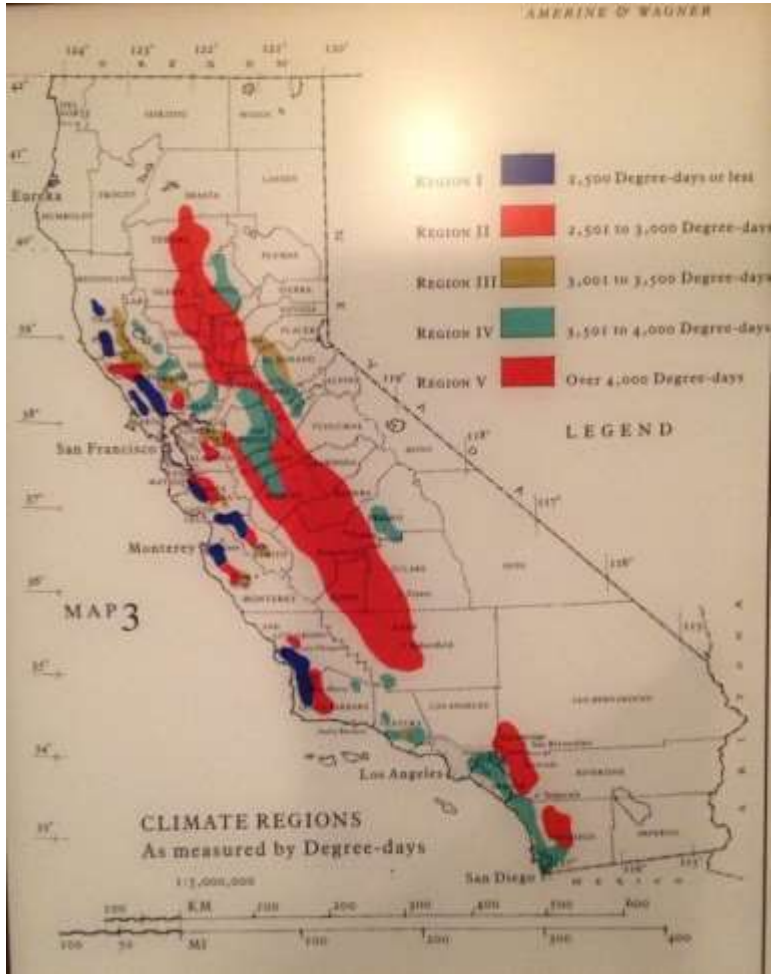


Macro weather data available. Need climate in the vineyard where the grapes are grown

One degree day per degree Fahrenheit over 50 °F.
Summed from April 1 – Oct. 31

Other considerations:
Pinot noir grown in cooler regions, some with coastal/marine influence

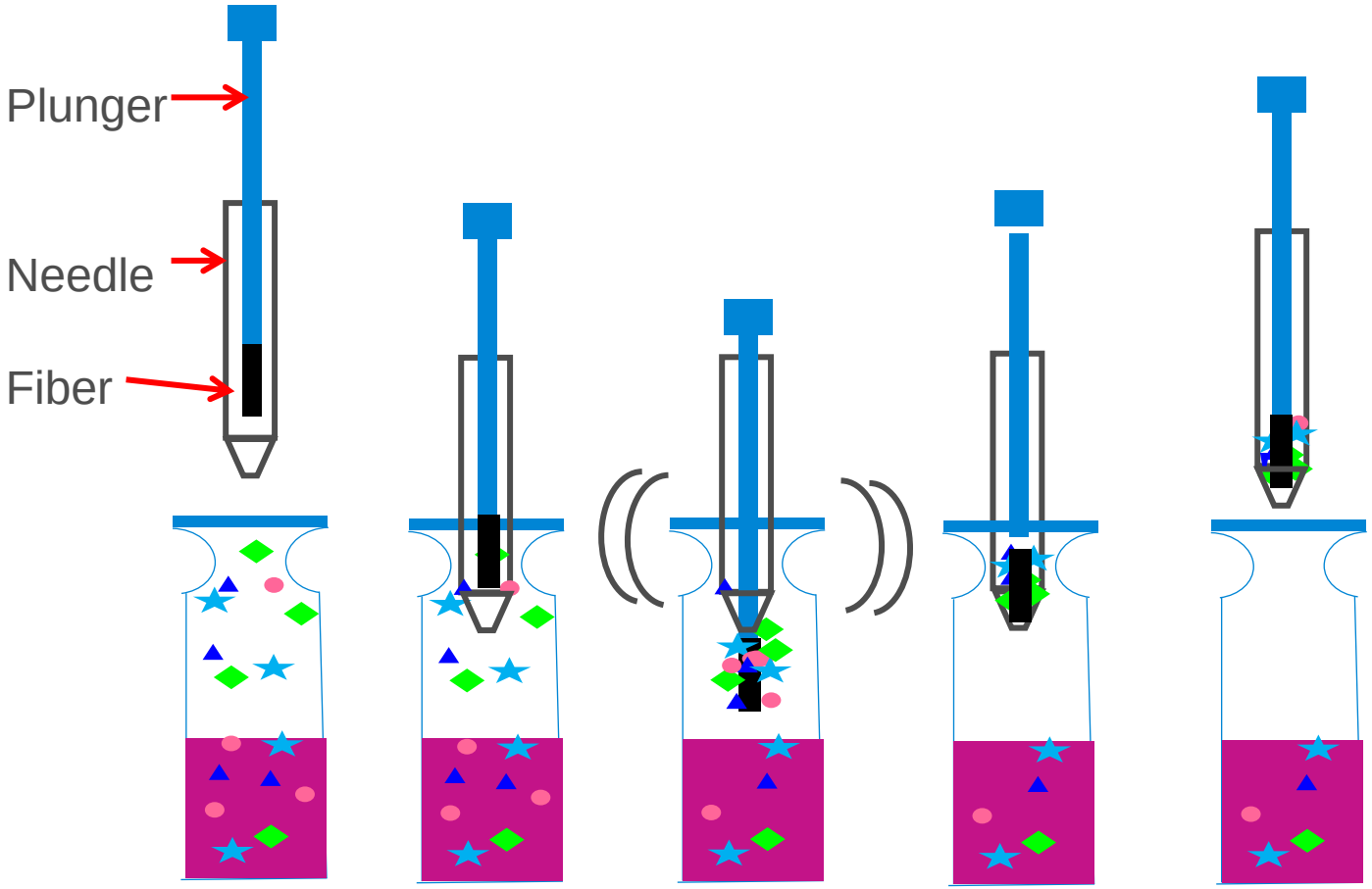
Altitude from near sea level to 2000 ft.



Winkler Heat Index regions in California

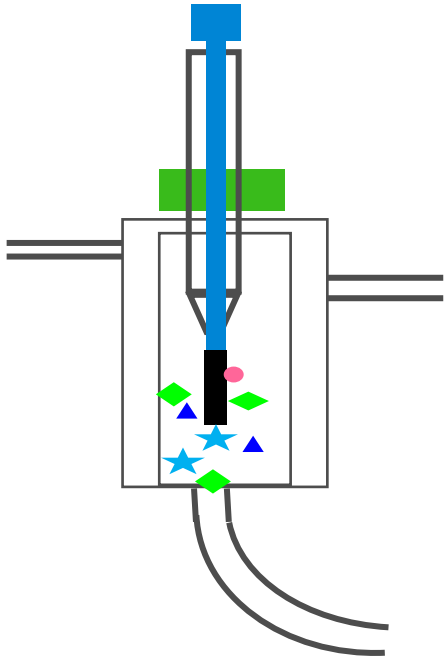
Recording weather stations will be installed to get microclimate data

Solid Phase Microextraction (SPME) Steps



5 min
30°C
Pre-equilibration

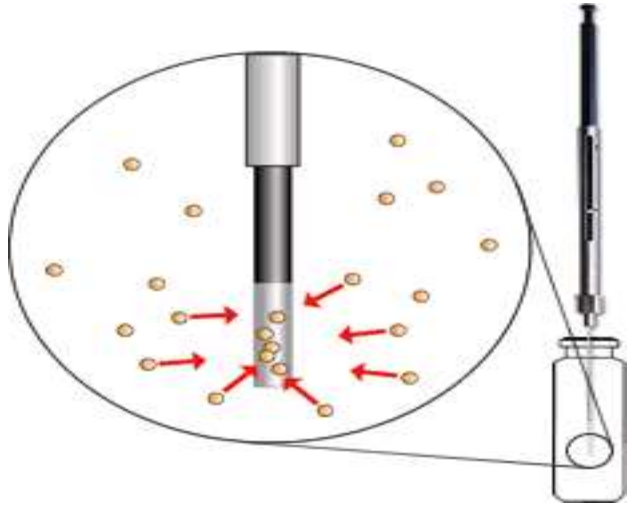
45 min
30°C
Sample Extraction



2 min
250°C
GC Injection

We have used HS-SPME GC/QQQ for ultra-trace analysis of haloanisoles in wine

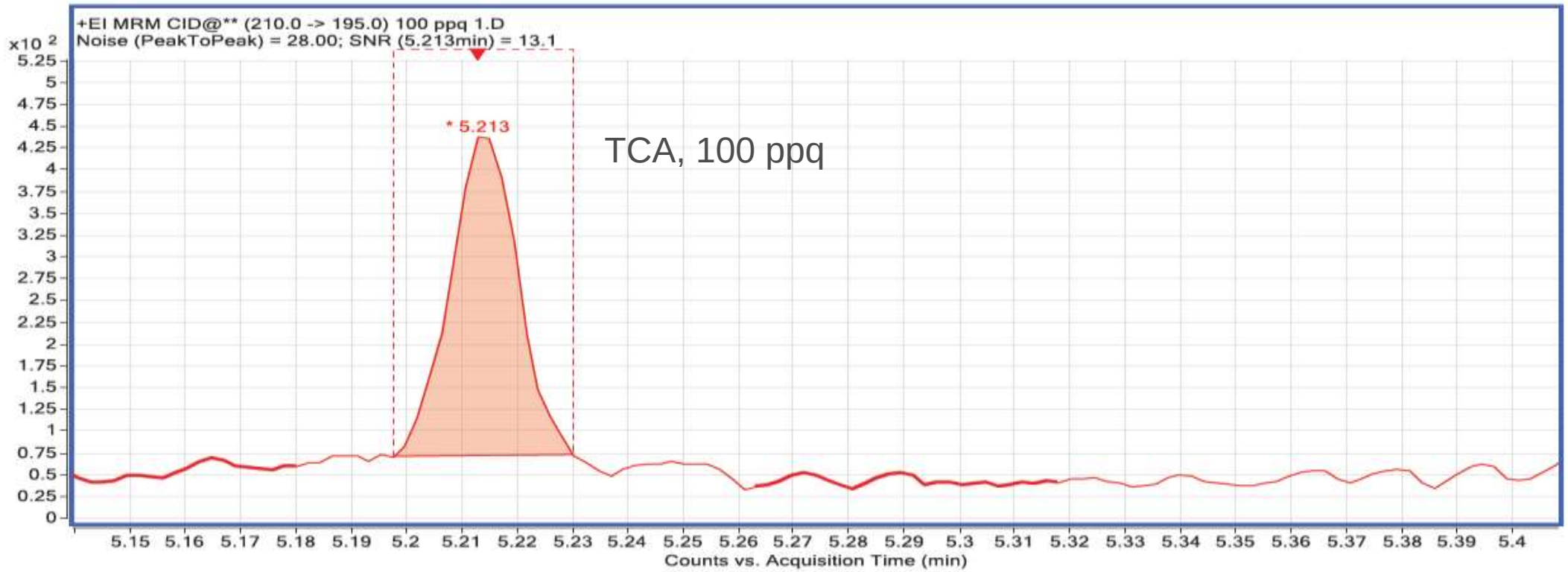
- GC conditions: initial 40C, ramp @ 30C/min to 280, hold for 3 Min, flow rate 1.2 mL/min
- Extraction conditions: SPME headspace, 100 µm PDMS, pre-extraction agitation @ 500 rpm & 40 C for five minutes, extract 10 minutes at 500 rpm & 40 C
- Injection: Splitless, desorb at 280 for 11 min
- Internal standards: d5-TCA, d5-TBA & C13-6 PCA; – for TeCA, C13-6 PCA was used as the internal standard



Solid Phase Micro Extraction

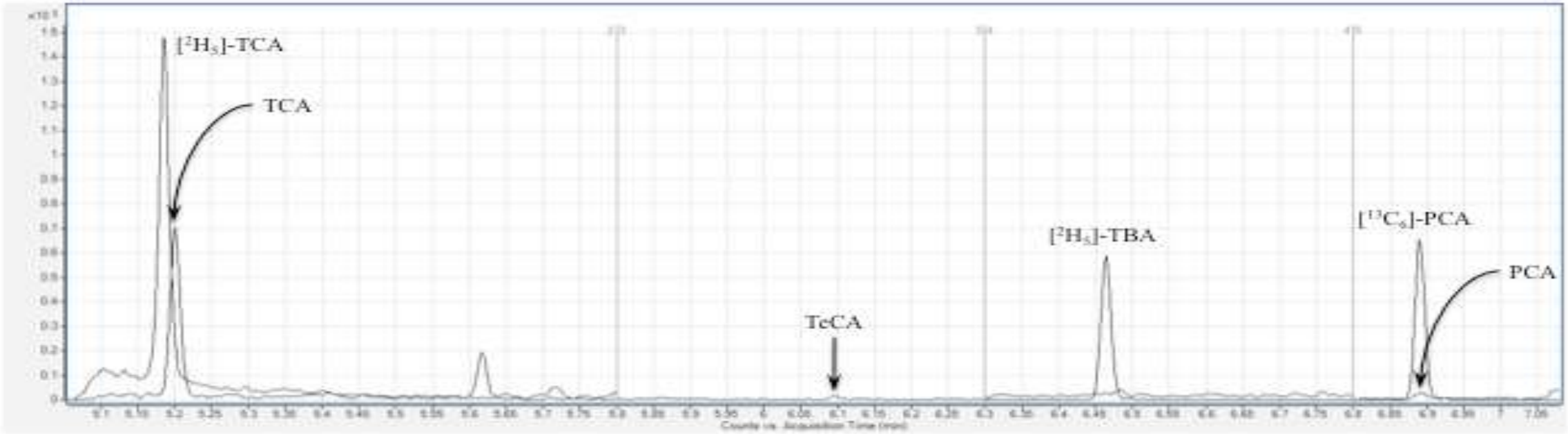
TCA at 0.1 ng/L 210→195 m/z S/N 13.1

100 ppq is equivalent to 1 second in 320,000 years



TCA in Customer Complaint Wine – Measured TCA = 2.3 ng/L (ppt)

Figure 1: Chromatogram of customer complaint wine A. Calculated TCA level 2.3 ng/L.



HS-SPME GC/Q-TOF Method

7200 Accurate Mass High Res. GC/Q-TOF

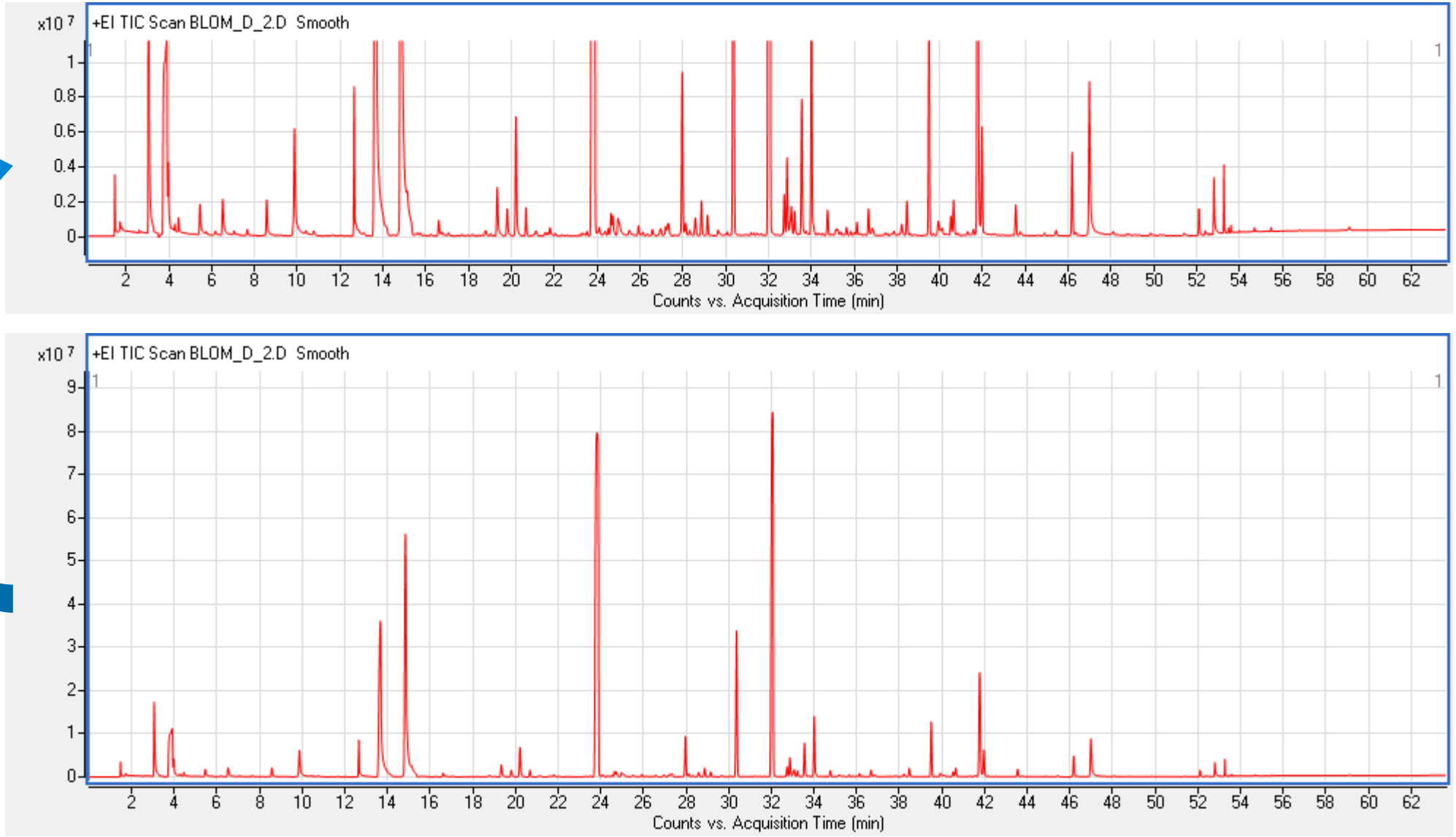
PAL 3 Autosampler for SPME,
Liquid or HS Injections

TOF mode @ 5Hz
30 m X 0.25 mm X 0.25 µm DB-WAXETR
40°C (5 min); 3°C/min→180°C (min);
30°C/min→240°C (10 min)

100 µm, 1 cm Fiber
Pre-extraction sample equilibration = 5 min @ 30°C
Headspace extraction = 45 min @ 30°C
Fiber desorption in MMI inlet = 2 min @ 240°C
Fiber conditioning = 10 min @ 250°C

Typical HS-SPME GC/Q-TOF chromatogram of *Pinot noir* wines in the study

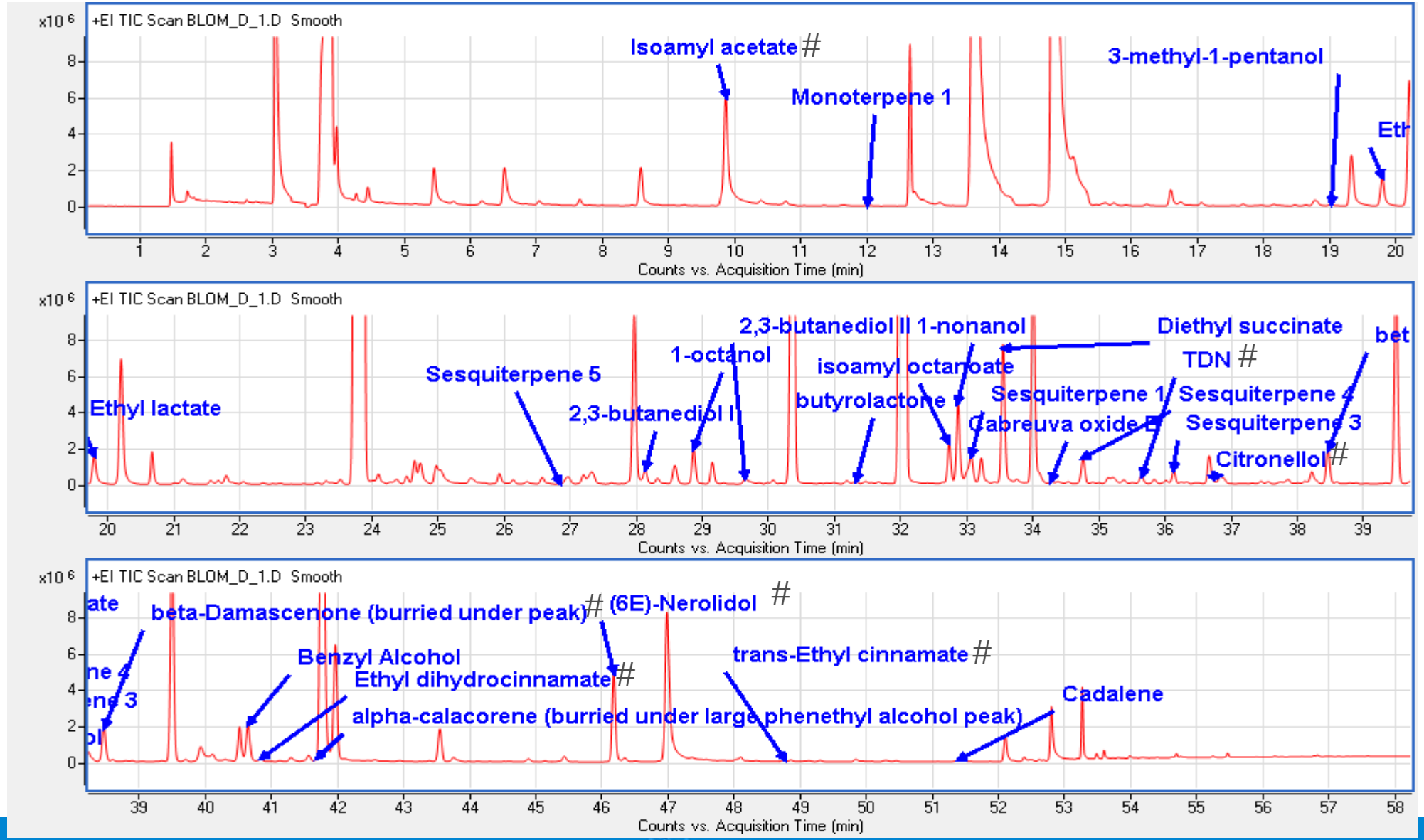
Zoom Abundance



Identification of *Pinot noir* wine volatiles

- Many of the compound were identified by running authentic standards (using RT Locked method on a different GC/MS).
- **For other compounds, we:**
  - Used spectral matching of high resolution accurate mass spectra to NIST 14 unit mass library
  - Calculated Retention Index (RI) values
  - Compared observed RI value to other published values (polar column)
  - Used knowledge of characteristic red wine volatiles with aroma impact
- Compounds found by spectral searching and RI comparison are *tentatively identified*.

Most significant compounds labeled (some identities are tentative)



Statistical Analysis

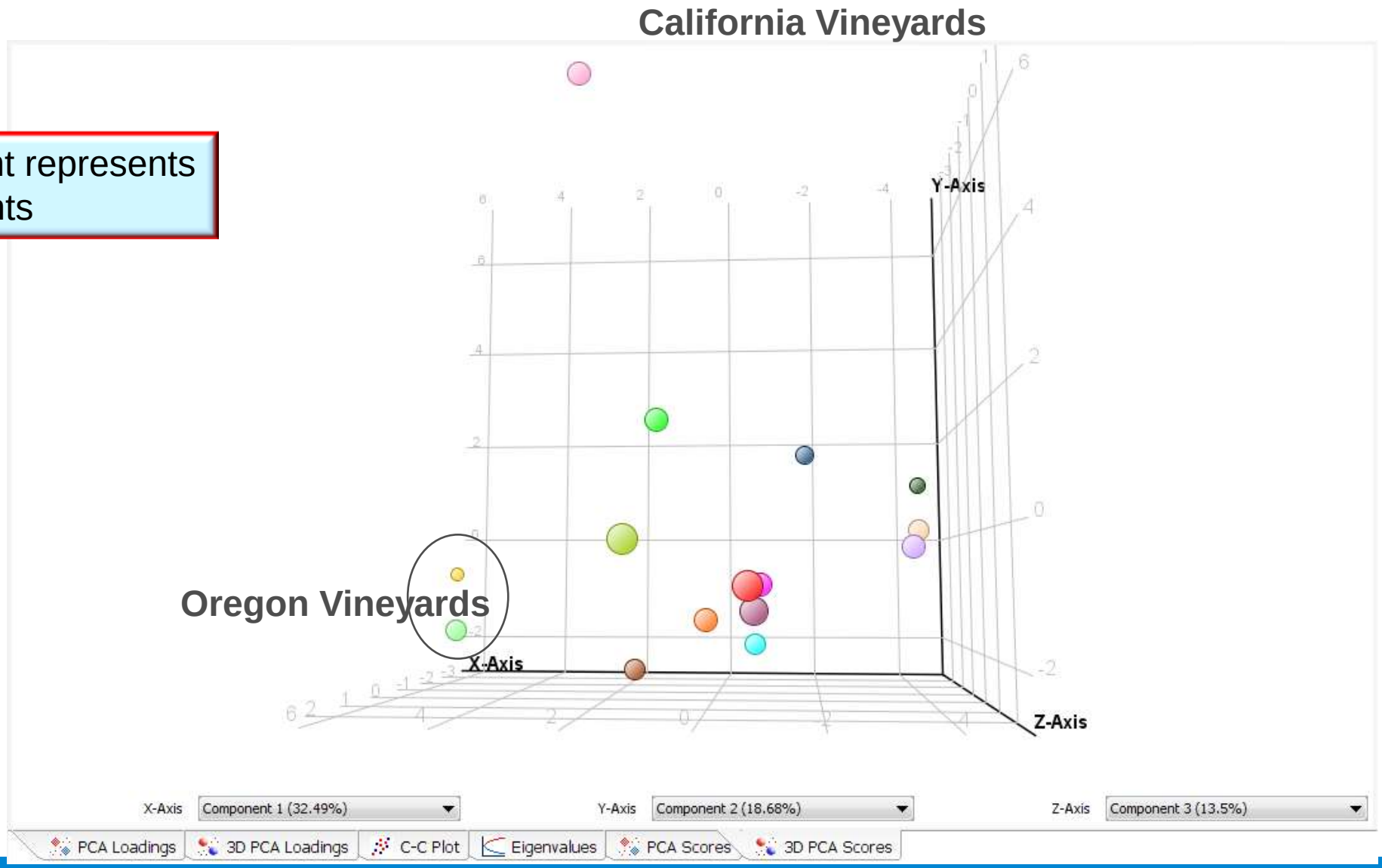
Use MassHunter Quant to produce table of Analyte response/ISTD response for 65 identified and tentatively identified compounds

ANOVA used to determine which compounds differed significantly by vineyard

PCA Scores and Loadings plots done using Mass Profiler Professional

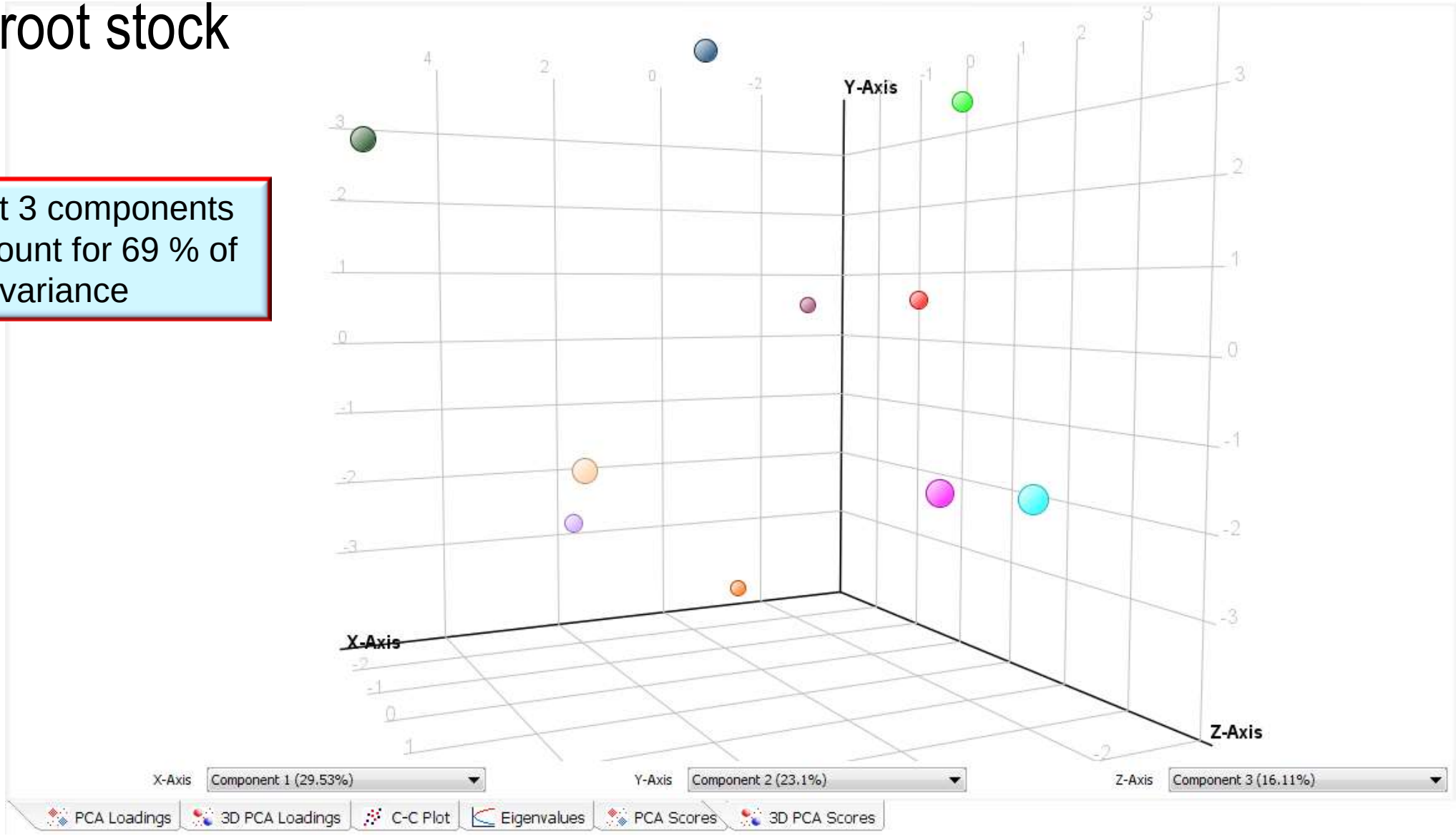
PCA Scores Plot Averaging 3 GC/MS replicates for three wine replicates

Each data point represents
9 measurements

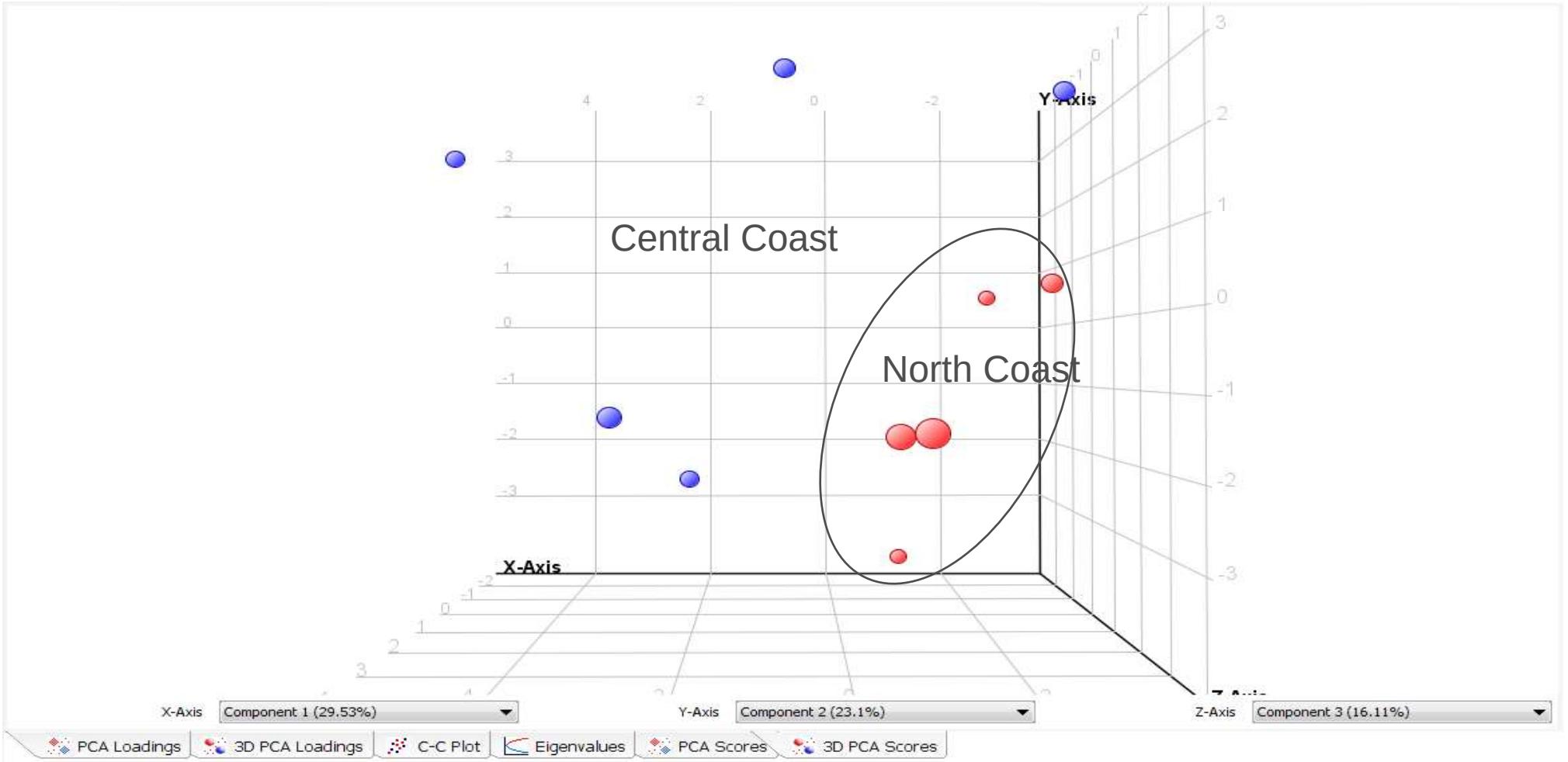


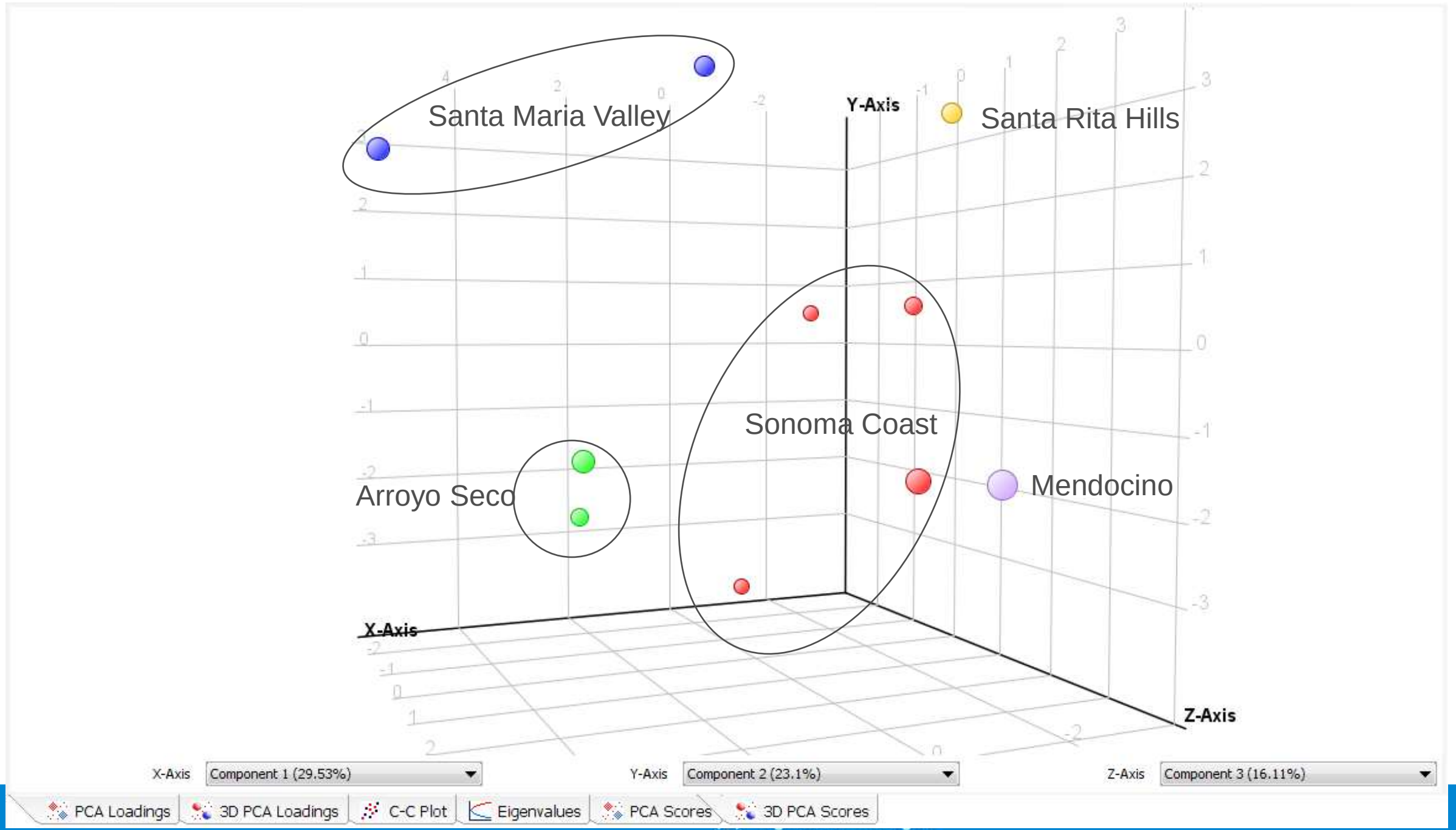
PCA Scores Plot for 10 vineyards with same combination of grape clone and root stock

First 3 components account for 69 % of the variance

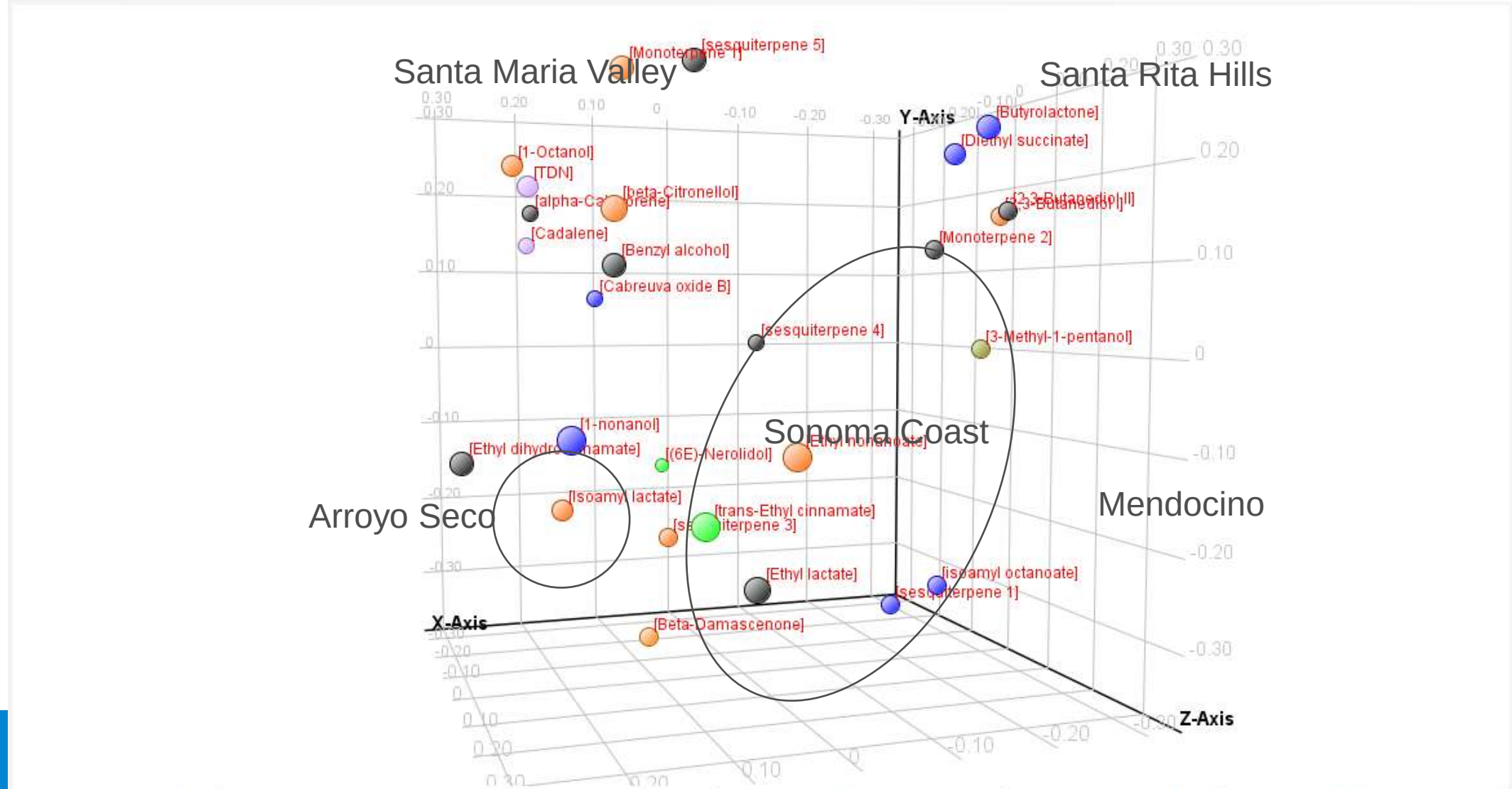


North CA Coast wines cluster away from Central CA Coast wines




Santa Maria Valley
Santa Rita Hills
Sonoma Coast
Arroyo Seco
Mendocino
Y-Axis
X-Axis
Z-Axis
X-Axis
Component 1 (29.53%)
Y-Axis
Component 2 (23.1%)
Z-Axis
Component 3 (16.11%)
PCA Loadings
3D PCA Loadings
C-C Plot
Eigenvalues
PCA Scores
3D PCA Scores

PCA Loadings Plot of significant compounds



Conclusions

- Four batches of wine were made from Pinot Noir grapes harvested from 15 different vineyards in California and Oregon (3 batches used for GC/Q-TOF analysis)
- Three replicate HS-SPME injections made for each wine batch
  - 15 vineyards X 3 wine batches X 3 replicates = 135 analyses
  - 9 analyses for each vineyard
- Volatile profile differs between vineyards in Oregon, CA north coast and CA central coast
- All vineyards could be separated in PCA
- This approach could be useful in evaluating regional differences in botanicals

What still needs to be done

- ❑ Obtain climate information for each AVA and each vineyard
  - ❑ Placing recording weather station at each vineyard location
- ❑ Correlate GC/Q-TOF results with
  - ❑ Vineyard microclimate
  - ❑ Low resolution GC/MS results
  - ❑ Metals analysis
  - ❑ Polyphenolic analysis by LC/DAD
  - ❑ Sensory Analysis
- ❑ Continue investigation over multiple years
- ❑ Add more vineyards with same grape clone and same root stock

Thanks to my coauthors: Anna Hjelmeland, Ron Runnebaum & Susan Ebeler

Thanks to Jackson Family Wines for Support and for contributing the grapes

Thank You!

UC Davis Department of Viticulture and Enology Teaching & Research Winery

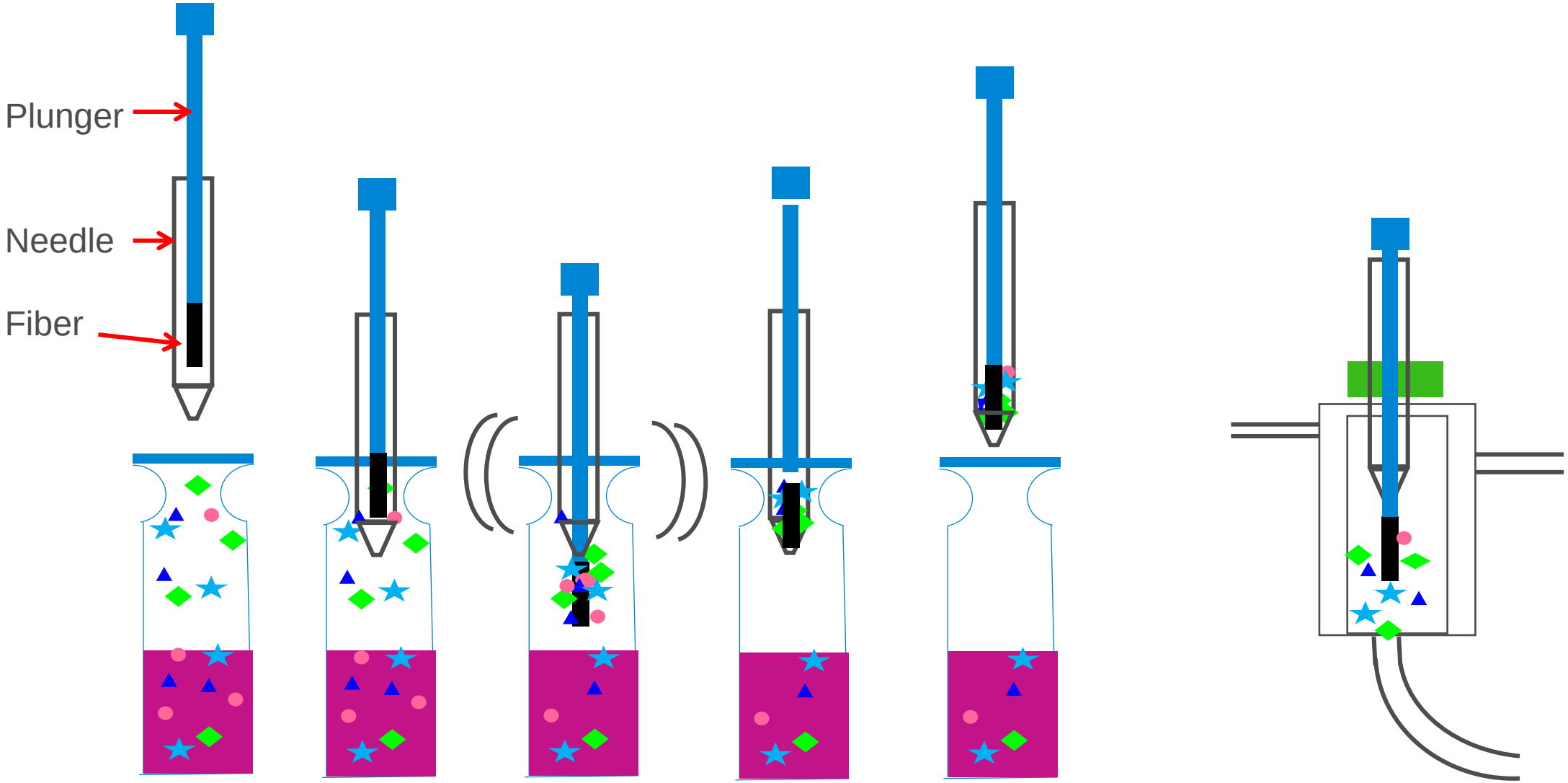
152 research Fermenters

Highly automated e.g. temperature control with minimal gradients

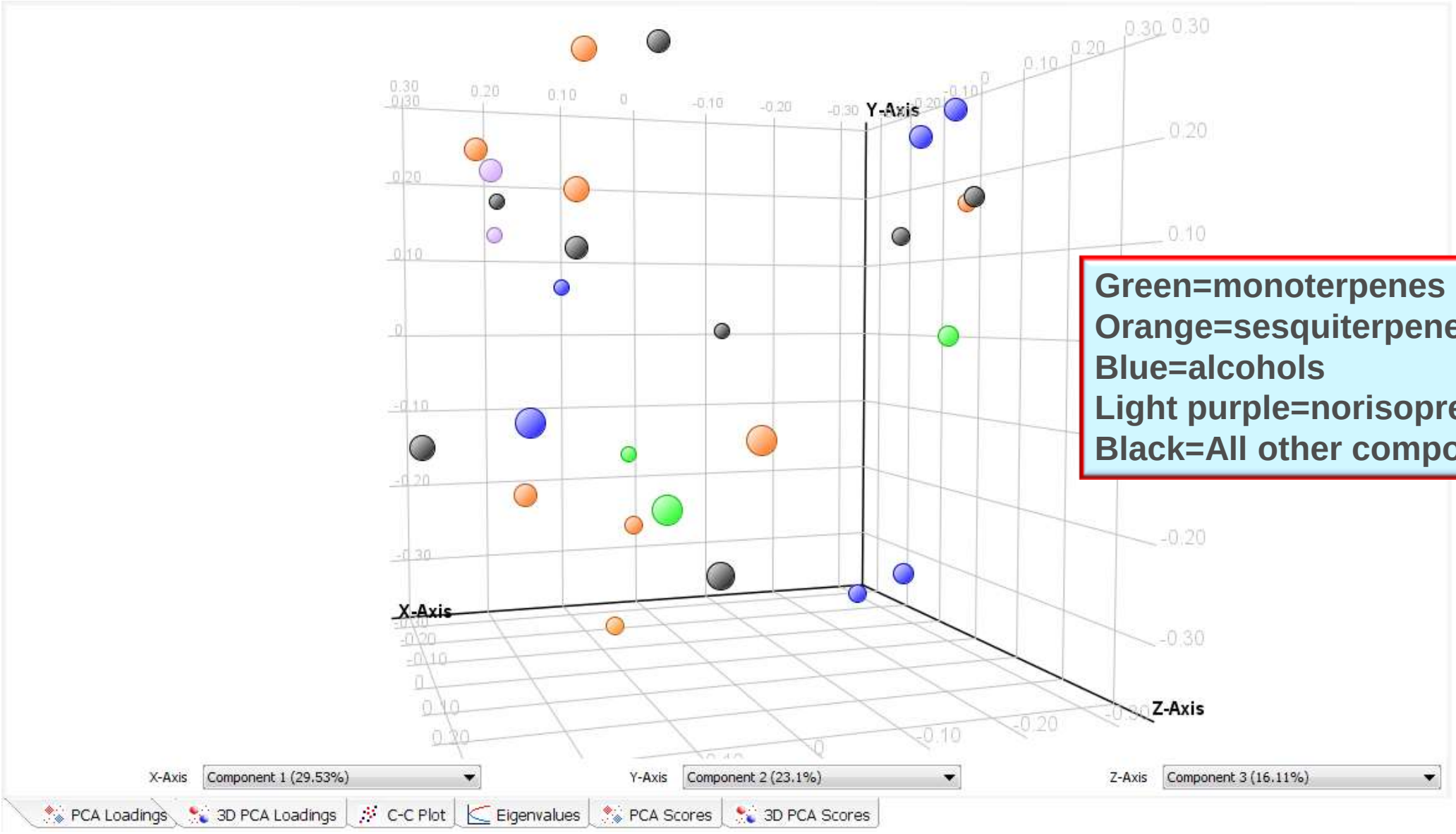
14 500-gallon fermenters

UC Davis Departments of Viticulture and Enology and Food Science Share the Robert Mondovi Institute – LEED Platinum building Complex

Solid Phase Microextraction (SPME) Steps



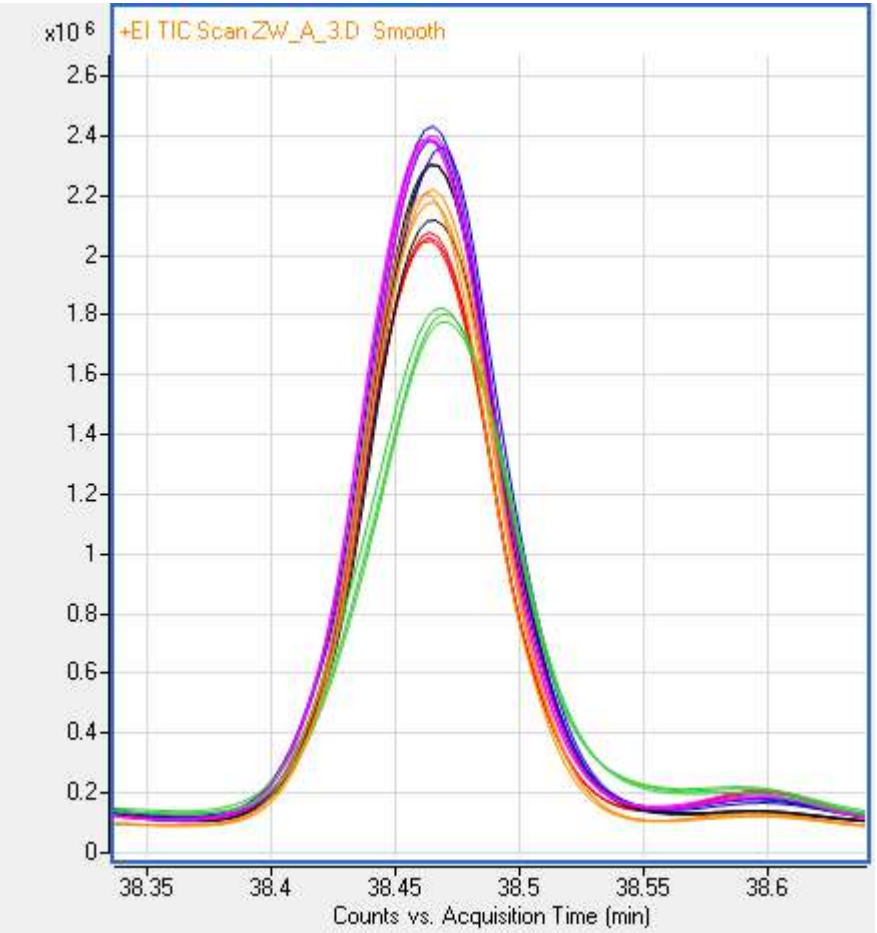
PCA Loadings Plot of the significant compounds



Green=monoterpenes
Orange=sesquiterpenes
Blue=alcohols
Light purple=norisoprenoids
Black=All other compounds

Not very discriminating peak in the TIC (left). EIC of β-Damascenone (m/z = 121) shows more discrimination

TIC



EIC m/z =121