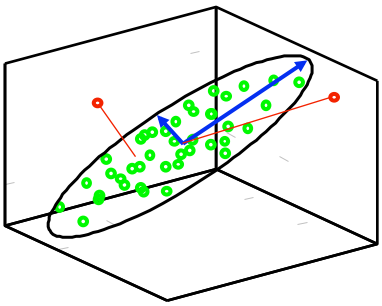
Chemometrics

Without Equations
(or hardly any)



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Biometrics

Technometrics

Econometrics

Psychometrics

Chemometrics

Chemical

Measurements

While worrying about the "Metrics" part, do not forget the "Chemo" part.

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Chemometrics - Use of Mathematics, Chemistry, and Logic to Perform:

- Experimental Design - How to take measurements in such a way as to maximize the chances of obtaining the desired information at the least cost.
- Data Analysis - How to get as much of the information out of a set of measurements as possible and relate *measurements* made on a *chemical* system to the *state* of the system

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This Workshop Concentrates on Two Aspects of Data Analysis:

- Exploratory Data Analysis and Pattern Recognition
- Regression

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Mathematical Tools Used in Chemometrics Can be Very Sophisticated.

- Multivariate Statistics
- Matrix and Tensor Algebra
- Eigenfunction - Eigenvalue Problems
- Neural Networks, Support Vector Machines
- Discriminant Analysis
- Etc.

But the Software Can Take Care of the Calculations -

Provided We Understand Conceptually What the Software is Doing.

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Outline (Part 1)

- Introduction
- Exploratory Data Analysis & Pattern Recognition Motivation
- Principal Components Analysis
- SIMCA
- Summary

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Outline (Part 2)

- Regression Motivation & Rational
- Classical Least Squares (CLS)
- Multiple Linear Regression (MLR)
- Principal Components Regression (PCR)
- Partial Least Squares Regression (PLS)
- Summary

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Outline (Part 1)

- Introduction
- Motivation for Exploratory Data Analysis & Pattern Recognition
  - what is Exploratory Data Analysis?
  - what is Pattern Recognition?
  - relevant measurements
  - some statistics definitions
- Principal Components Analysis
- SIMCA
- Summary

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What is Exploratory Data Analysis and Pattern Recognition?

It is important to examine data patterns to:

- Identify trends, clusters, and other patterns that should be explained and understood before modeling.
- Ensure the data represents all types of samples that the model will be expected to handle in application.
- Detect and correct or remove faulty samples which might endanger the reliability of the model when applied to future samples.
- Often the final goal is to identify trends or class membership.

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Pattern Recognition Applications

- How do I know I am producing a product within or outside of specification(s)?
- Am I producing the same product at all my plants?
- What new source of a feed-stock is most like the source I just lost?
- How should I modify my formulation to correct for a new feed-stock to produce product closest to my traditional product?

These are just different ways of asking the same question:

How do if I know when two items are the same or when they are significantly different?

To Answer Any Such Questions I Need Information - Data

Data is usually easy to find:

- Quality Monitoring
- Product Research
- Customer Surveys
- etc.

Data from designed experiments is usually best

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Chemical Instrumentation Has Caused a Data Explosion

FTIR Spectra
60 samples from
3500-700 cm^{-1} @ 4 cm^{-1} resolution

84,000 Pieces of Information!

Vis-NIR-XRF Hyperspectral image

606 x 659 pixels = 399,354 samples
from 1241 channels / wavelengths

495,598,314 Pieces of Information!

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Most of This Information is Either:

- Irrelevant to the problem we want to solve
- Redundant
  - redundancy is a problem for some types of analyses
  - opportunity for other types

Need to find a way to sift through this data and find the parts that will lead to a solution to the problem

Trivial Example: How can one tell the difference between a cat and a dog?

Perhaps Count the Number of Feet?

Fluffy	4
Spot	4
Puff	4

Rex	4
Lassie	4
Rover	4

These numbers are all the same.

No Variance in this Variable

Therefore no information in these numbers.

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A number only has value when it is compared with other numbers

Since we are looking for the difference between dogs and cats, we need a numerical descriptor that changes, *i.e.* has variance

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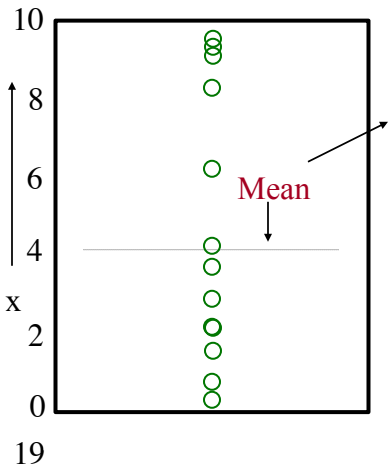
Before Defining Variance
Need to Define Mean:



Bar signifies the mean value

$$\overline{X} = \frac{9.4 + 4.1 + 0.6 + 3.5 + \ldots + 2.7}{15} = 4.0$$

- Degrees of Freedom
- Number of samples
- Number of pieces of information available

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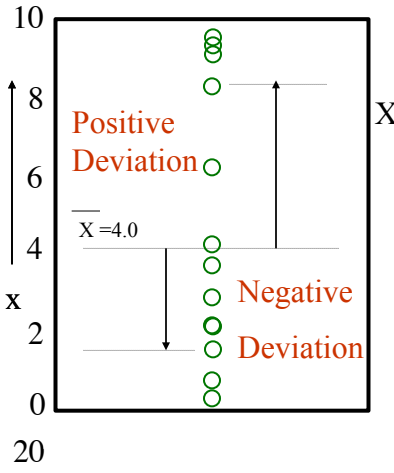
Calculating Variance



Deviations squared to make positive

$$X_{Var} = \frac{(9.4-4.0)^2 + (4.1-4.0)^2 + \ldots + (2.7-4.0)^2}{15 - 1} = 11.7$$

- Lost one Degrees of Freedom

Why?

Conservation of Information

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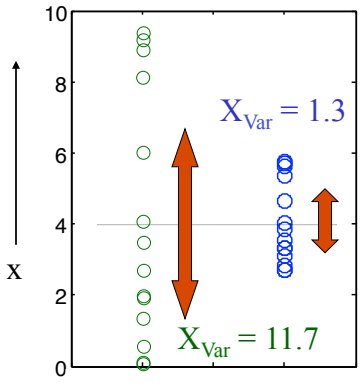
Both data sets have the same mean



but they have different variance

Note: Standard Deviation

$$X_{std} = (X_{var})^{1/2}$$

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But in statistics ...

Variance = Information

Note: Many methods are concerned with capturing the maximum sum of squares or with minimizing the residual sum of squares (*i.e.* least squares)

Back to the Cats and Dog problem

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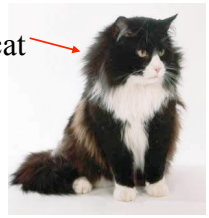
Perhaps length of fur can be used to tell cats from dogs?

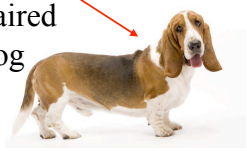
Long haired cat



Short hair cat

Large Variance

Short haired dog



Long haired dog

But still little Discrimination Power between cats and dogs.

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Variance = Information

But is the information relevant to the problem or stated objective?

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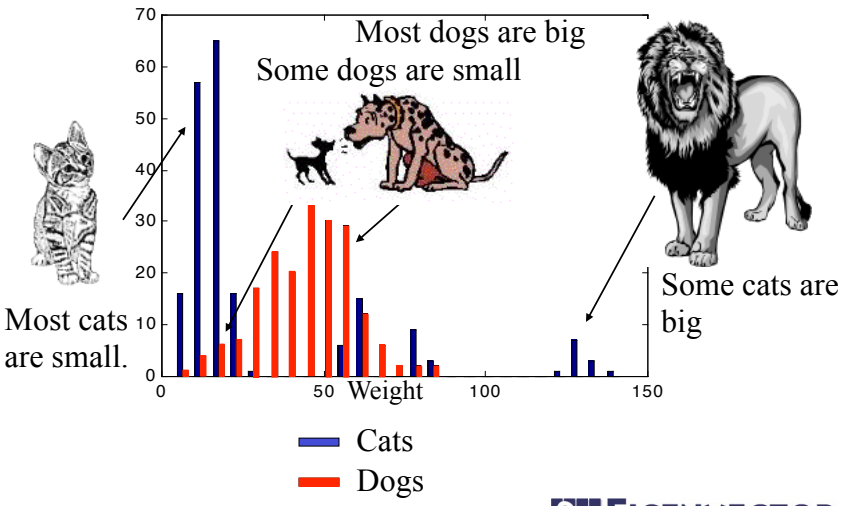
Perhaps Weight of Animal?



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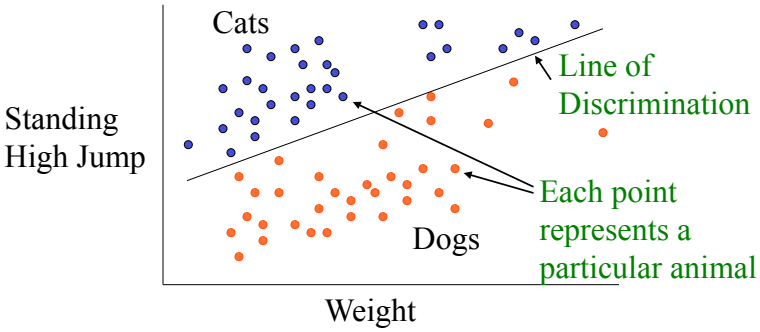
Perhaps two variables *Together*?



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Some Questions:

- If two variables were better than one, how about three or more?
- How to determine which variables are relevant to the problem?
- How to determine the underlying variables that define the problem and the solution?
  - I want to understand the process (*i.e.*, the underlying structure of the data).

A more realistic example…

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Table of 20 Solutions Analyzed for Ca^{2+} and Cl^- Concentrations.

	$[Ca^{2+}]$	$[Cl^-]$
1	0.1254	0.2435
2	0.4542	0.9097
3	0.7190	1.4361
4	0.8965	1.7946
5	0.2770	0.5536
⋮	⋮	⋮
20	0.9944	1.9886

20 Rows

Samples

20 x 2 Matrix

table of numbers

2 Columns

Variables

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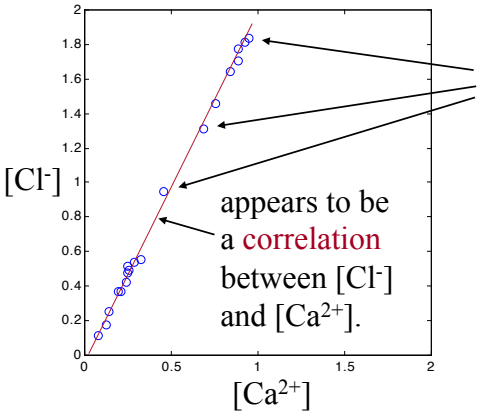
Plot [Cl^-] versus [Ca^{2+}]



Each point represents a sample in two-variable space.

The existence of strong correlation implies there aren't two variables, but two forms of the same underlying variable.

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How to find this underlying variable?

Next topic:

Principal Component Analysis

PCA

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Outline (Part 1)

- Introduction
- Pattern Recognition Motivation
- Principal Components Analysis (PCA)
  - What is PCA?
  - Scores and Loadings
  - Interpretation
  - Examples
- SIMCA
- Summary

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The Problem in Dealing with So Many Variables is that I Can Only Think in 2 or 3 Dimensions

Is there a way to concentrate the information from *many* variables into a *few* (hopefully 2 or 3) underlying variables?

Hint: Variance = Information

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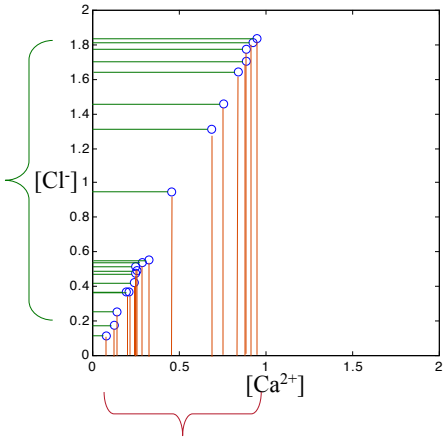
Plot the Cl^- Concentration vs. the Ca^{2+} Concentration

$[Cl^-]_{var} = 0.4557$

Can do the same with $[Cl^-]$.



$[Ca^{2+}]_{var} = 0.1135$

Can read the $[Ca^{2+}]$ of each sample by projecting it on the [Ca2+] axis.

Can then calculate the variance of the [Ca2+].

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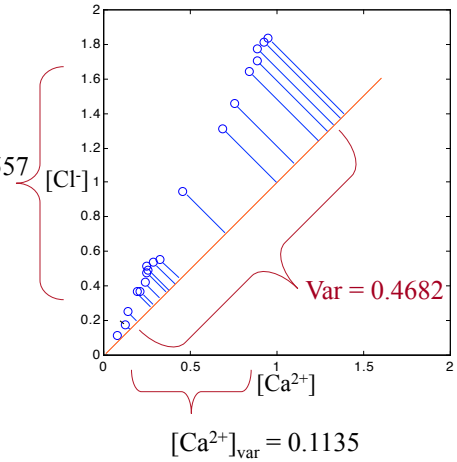
Find a New Axis of Maximum Variance:

$[Cl^-]_{var} = 0.4557$



$[Ca^{2+}]_{var} = 0.1135$

There are an infinite number of new axes that I can create.

I can project my samples onto this new axis and calculate the amount of information captured by this axis (i.e. variance.)

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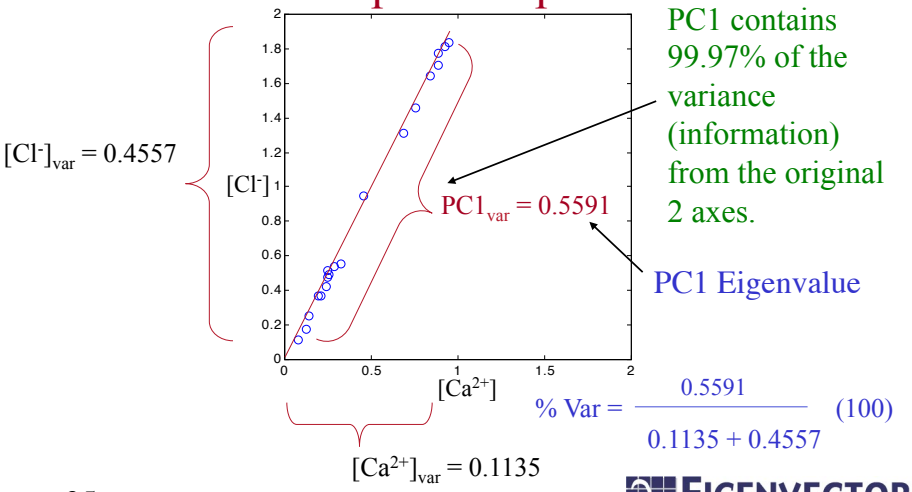
Only One New Axis Which Captures the Maximum Variance:

Principal Component



PC1 contains 99.97% of the variance (information) from the original 2 axes.

PC1 Eigenvalue

$$\% \text{ Var} = \frac{0.5591}{0.1135 + 0.4557}(100)$$

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How Was PC1 Constructed?

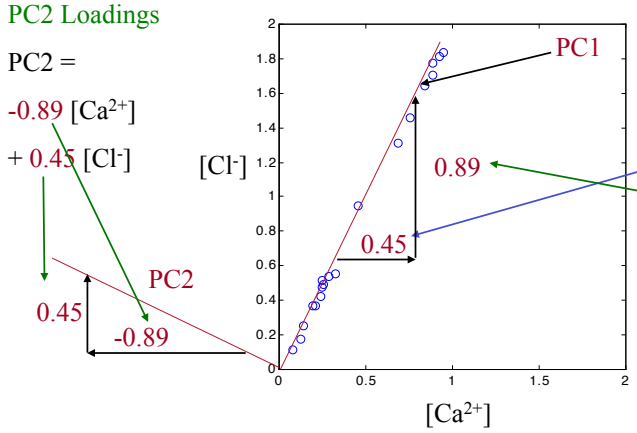
Linear Combination of Original Axes

PC2 Loadings

PC2 =

$-0.89\ [Ca^{2+}]$

$+ 0.45\ [Cl^-]$



PC1 =

$0.45\ [Ca^{2+}]$

$+ 0.89\ [Cl^-]$

Loadings

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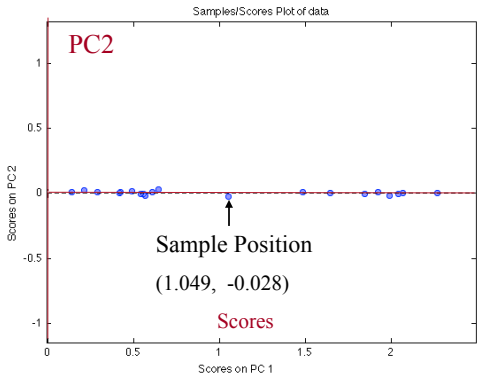
The Positions of the Data in the PC Coordinate System (Scores)



Same as last plot except rotated to make PCs new axes

(PCs are a new coordinate system)

PC1

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Summary of Ca^{2+}/Cl^- Example

$$PC1 = 0.45\,[Ca^{2+}] + 0.89\,[Cl^-]$$

- Loadings for PC1 in a ratio of 1:2
  - One part Ca^{2+} and two parts Cl^-
- PC1 related to $[CaCl_2]$
  - ~ 99.97% variance is systematic
- PC2 related to random measurement error
  - ~ 0.03% noise

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Important Mathematical Term: Chemical Rank of the Matrix:

The number of independent, underlying, meaningful sets of information in a data set.*

- Calcium ion, chloride ion example
  - Started with two variables: $[Ca^{+2}]$ and $[Cl^{-}]$
  - Ended with one meaningful variable: $CaCl_2$
  - Chemical Rank = 1

*This is really the definition of "pseudo-rank." The mathematical rank is the number of linearly independent rows or columns.

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Summary of What We Have Done and Learned

- Variance = Information
- Many variables contain information, but much of the information may be redundant (correlated) or irrelevant.
- Use linear combinations of the original variables to create new variables (Principal Components, PCs) that combines redundant information → PCA
- The Rank of a Matrix is the number of Principal Components that describe other than random noise.

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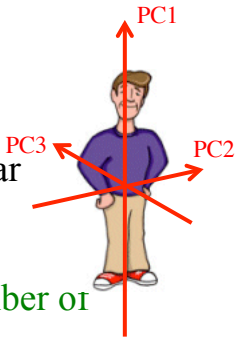
How Does PCA Find the PCs?

- The 1st principal component (PC) passes through the origin and the maximum variance of the data.
- The 2nd PC is orthogonal (perpendicular or independent) to PC 1 and passes through the second most variance.
- The process is continued until the number of new PCs = number of old variables.
  - PCs ≤ min(number of samples, number of variables) = mathematical rank of the data



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What Does PCA Give Me?

- Most of the variance (information) is concentrated in the first few PCs.
  - Some may be relevant to the problem of interest
- Small random noise is sifted into the later PCs
  - and may be thrown away - data filtering
  - or used in a residuals analysis
- Important Assumption:
  - The signal/noise is > 1
  - *i.e.,* most of the variance is from sources other than random noise

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What Does PCA Give Me?

- Loadings: Compositions of the new PC axes in terms of the old variables. May be able to interpret the loadings in chemical terms.
  - Loadings ⟷ Variables
- Scores: The position of the samples in the new PC coordinate system. The closer samples are to each other in the first few PC space, the more they are alike.
  - Scores ⟷ Samples
- Eigenvalues - The variance stored in each of the Principal Components
  - Eigenvalues can then be used to calculate the % of the information stored in each PC.

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Say 100 Times Before You Go to Sleep Tonight!!

Loadings ⟷ Variables

Scores ⟷ Samples

Eigenvalues ⟷ Variance

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Arch Example: a Little More Complicated Problem

Track trading and migration patterns of prehistoric tribes. Samples of two unknown obsidian artifacts were compared to samples obtained from three obsidian quarries using XRF.

Quarry	Number of Samples	Row Index
K	10	# 1-10
SH	23	#11-33
AN	21	#34-54
Unknowns	2	#55 & 56
Total	56	

BR Kowalski, TF Schatzki, FH Stross. "Classification of archaeological artifacts by applying pattern recognition to trace element data." *Anal. Chem.*, **44**(13); 2176-2180 (1972).

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20 Obsidian Samples Were Analyzed for 10 Metals*

	Fe	Ti	Ba	Ca	K	Mn	Rb	Sr	Y	Zr
1	1173	417	54	961	441	47	135	55	60	145
2	1164	404	56	916	446	42	120	58	45	148
3	1030	373	59	920	487	38	128	53	58	138
4	1077	373	55	888	455	38	97	51	54	145
5	1080	403	53	919	442	41	133	60	45	155
•	•	•	•	•	•	•	•	•	•	•
•	•	•	•	•	•	•	•	•	•	•
•	•	•	•	•	•	•	•	•	•	•
56	880	156	36	279	388	37	103	15	53	143

56 x 10 Matrix

*X-Ray Fluorescence (ppm)

D.F. Stevenson, et. al., *Archaeometry*, **13**, 17 (1971).

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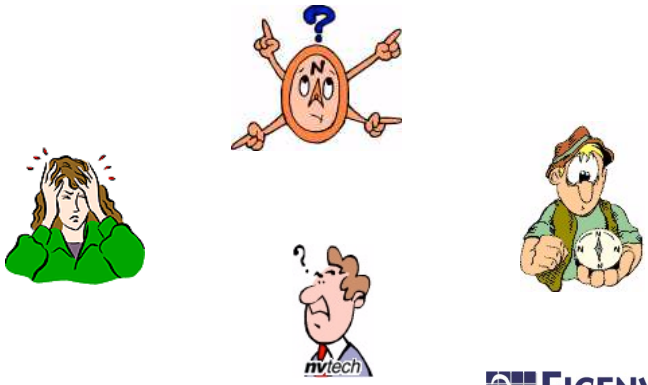
Plot Each Sample as a Point in 10-Dimensional Space



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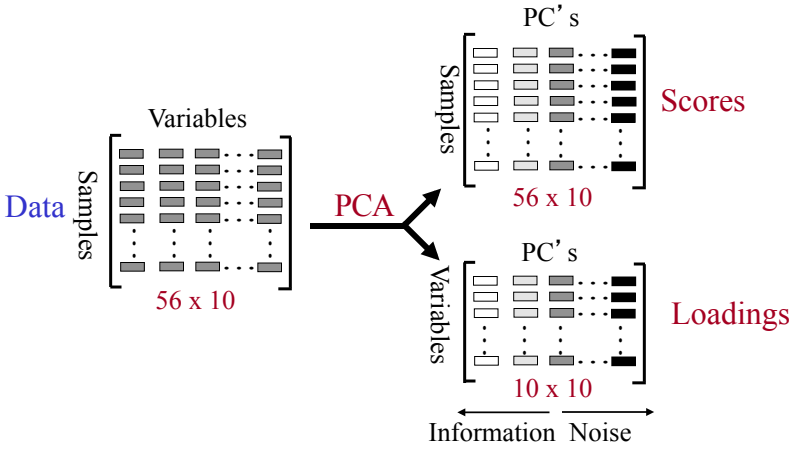
Let's Do It the Right Way

PCA



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When Washing Clothes
Sometimes Detergent is Not Enough

You may need Pretreatments

And You Need a Different Pretreatment for Different Stains

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Before PCA do:
Data Preprocessing

- None
- Mean centering
- Autoscaling

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PCA of GLASS - No Preprocessing

```
Principal Components Analysis Model
X-block: arch  56 by 10
Included: [ 1-56 ]  [ 1-10 ]
Preprocessing: None
Num. PCs: 5
```

Percent Variance Captured by PCA Model

Principal Component Number	Eigenvalue of Cov(X)	% Variance Captured This PC	% Variance Captured Total
1	2.36e+06	97.71	97.71
2	3.44e+04	1.42	99.13
3	1.93e+04	0.80	99.93
4	1.24e+03	0.05	99.98
5	2.08e+02	0.01	99.99

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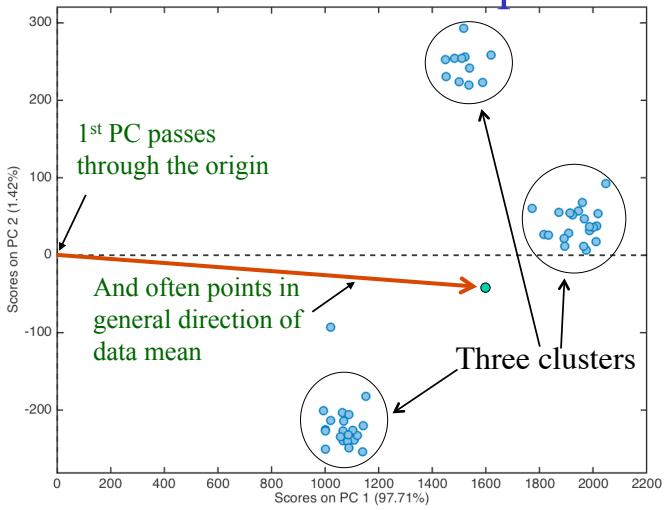
PCA Scores Plot - No Preprocessing



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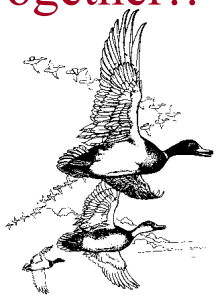
If we know and label the source of the known samples (Learning Set), we can identify the clusters and the memberships of the unknowns.

Birds of a Feather Flock Together!!



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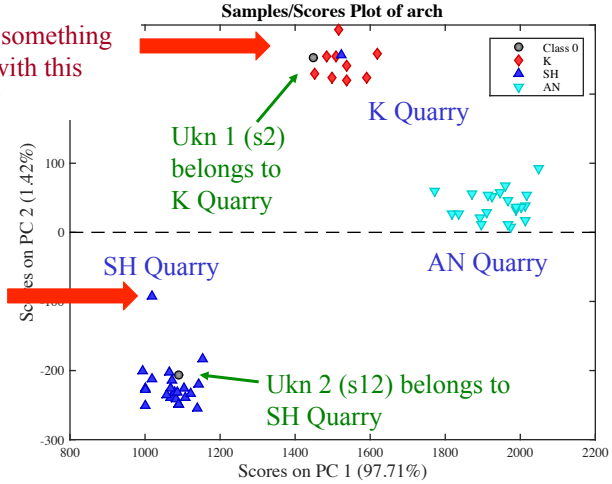
PCA Scores Plot - No Preprocessing

Is there something wrong with this picture?



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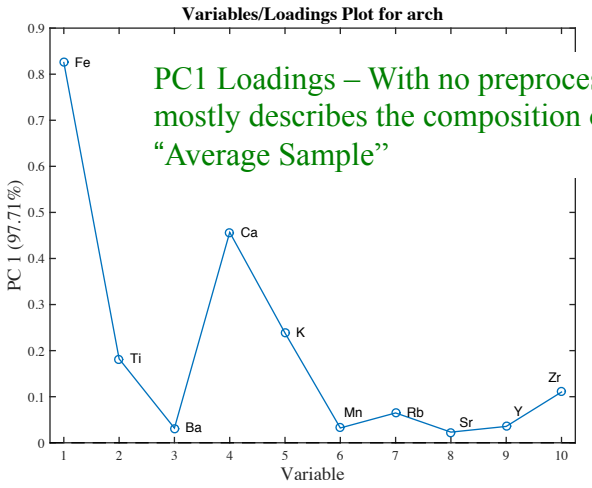
Loadings Plot



PC1 Loadings – With no preprocessing it mostly describes the composition of the "Average Sample"

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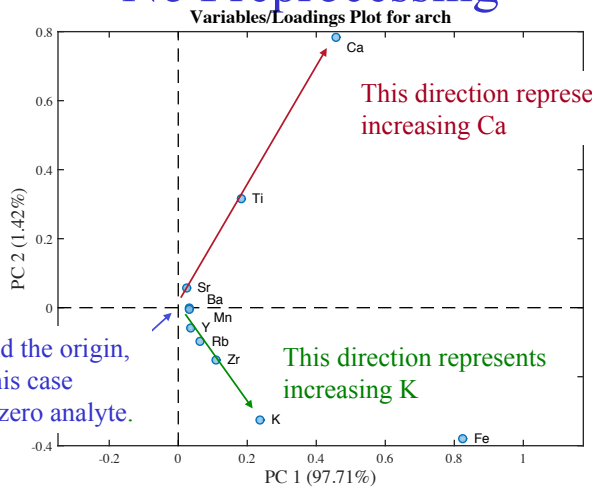
PCA Loading Plot PC 2 vs PC 1 - No Preprocessing



This direction represents increasing Ca

Always find the origin, which in this case represents zero analyte.

This direction represents increasing K

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Loadings Plots are like a Signpost to Locations in the familiar world of measured variables

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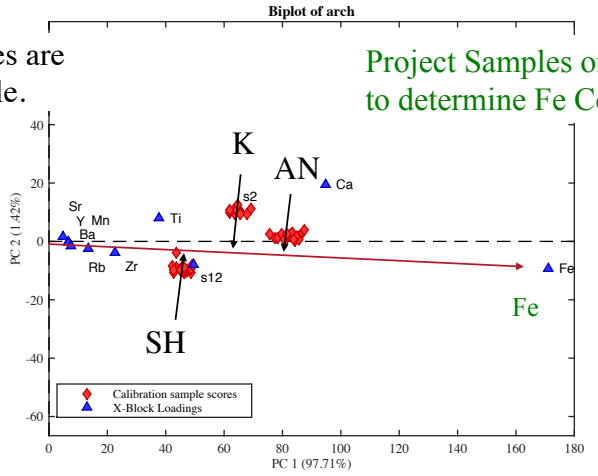
Easier to Understand by Superimposing Scores and Loadings - Biplot

Note the axes are equal in scale.

Project Samples onto Fe line to determine Fe Content.

PC 1+2 ~99.1%



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Interpret this Biplot:

- According to the first two PCs:
  - the AN Quarry has the highest Fe content,
  - the SH Quarry has the lowest Fe content, and
  - the K Quarry has ~average Fe content.
- Difference in Fe content is one source of discrimination between the quarries.

What we have learned

- The first two PCs can discriminate between the three quarries.
- PC 1 contains 99% variance, but mostly describes what the quarries have in common.
- But we really want to understand the difference between the quarries…

Data Preprocessing

- None
- Mean centering
- Autoscaling

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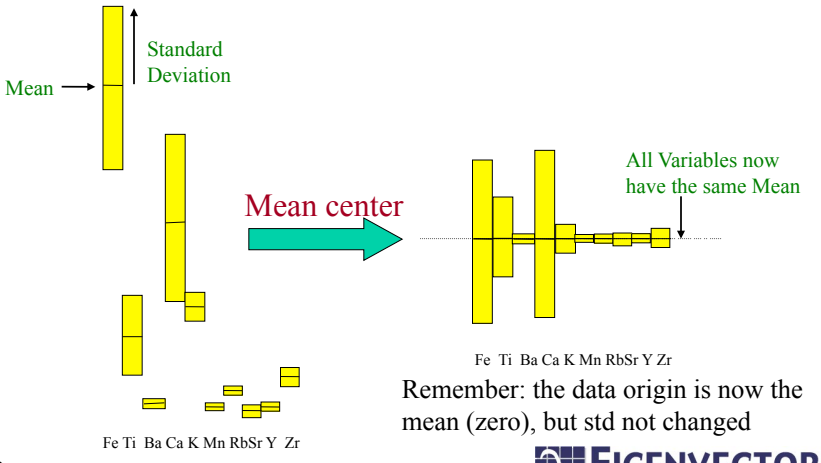
Repeat the PCA with Mean Centered Data



Remember: the data origin is now the mean (zero), but std not changed

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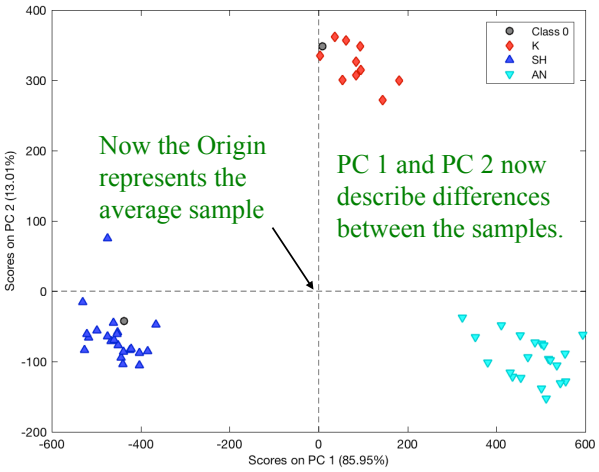
Scores Plot - Mean Centering



PC 2 13% of Variance

PC 1 86% of Variance

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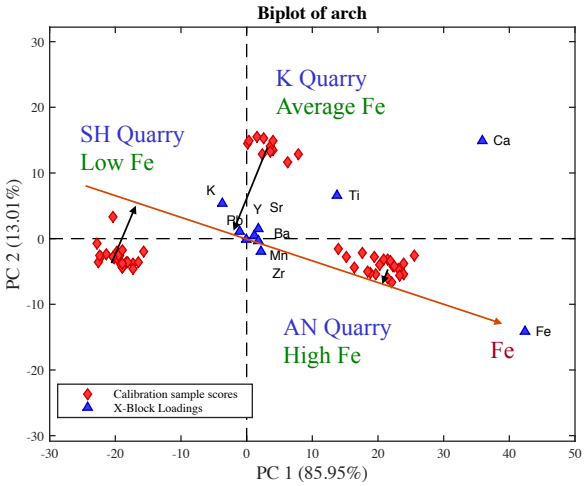
Biplot - Mean Centering



Note axes have equal scale

PC 1+2 ~98.97%

Fe is important in discriminating between the Quarries in PC 1 - PC 2 space.

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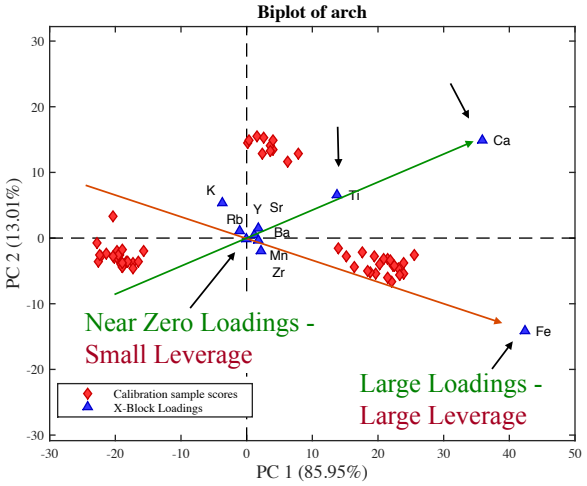
Biplot - Mean Centering



Fe, Ca and Ti are important in discriminating between the Quarries in PC 1 - PC 2 space.

Large Leverage

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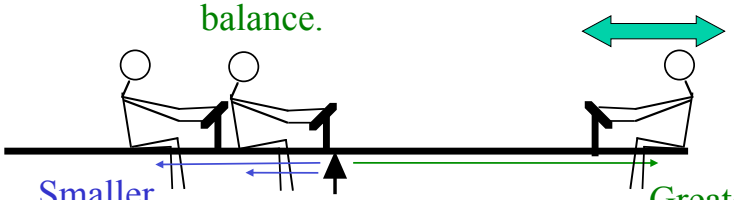
Leverage: Distance from Origin

Greater Leverage because the slightest change in position will greatly upset the balance.



Smaller Leverages

Origin

Greater Leverage

Leverage is measured by a quantity T^2.

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Variables with Small Leverages:

- Have little to say about the differences between the three quarries.
- And/or have relatively small signals (*i.e.*, low concentrations) compared to signals from other variables.
- Both of these situations result in small variances - little information

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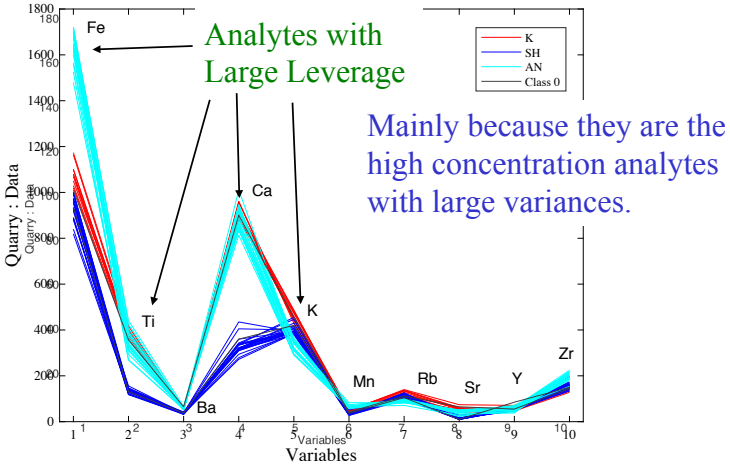
Plot the Data: Analyte Profile



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Do all four participants in this decision have the same Leverage?

Much More Fair!

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Data Preprocessing

- None
- Mean centering
- Autoscaling

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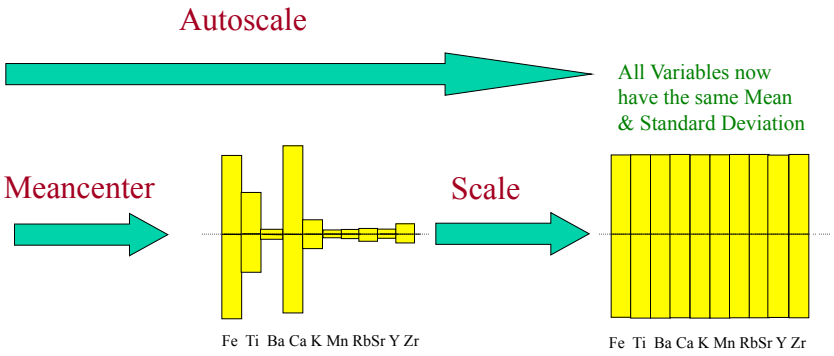
Autoscaling



Remember: autoscaling includes mean centering

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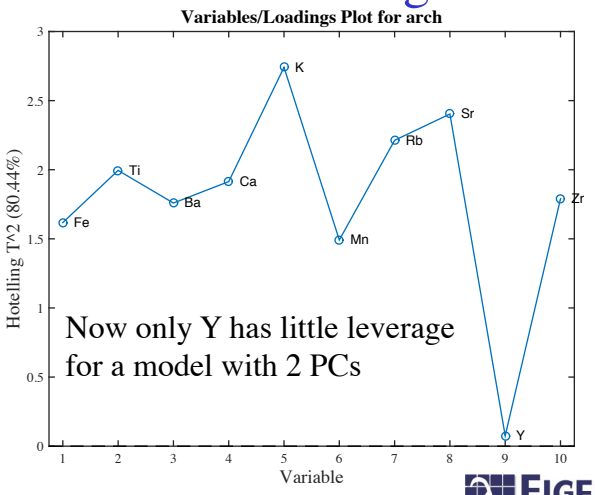
Leverages of Variable after Autoscaling



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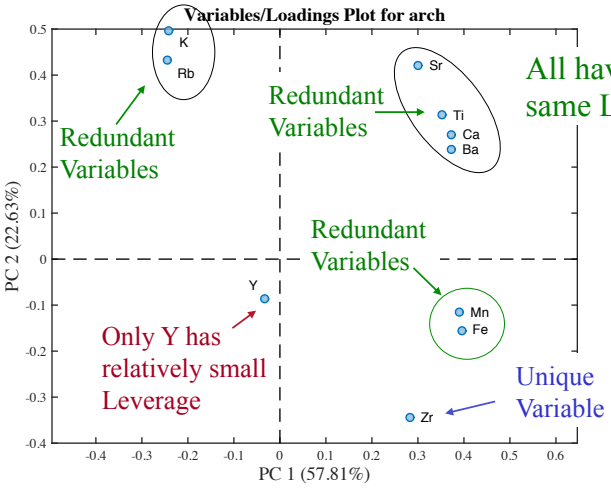
Loadings Plot - Autoscaling



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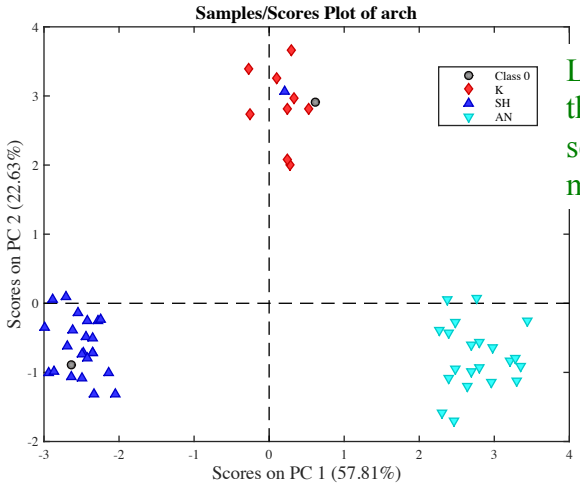
PCA Scores Plot -Autoscaling



Looks much the same as the scores plot for mean centering

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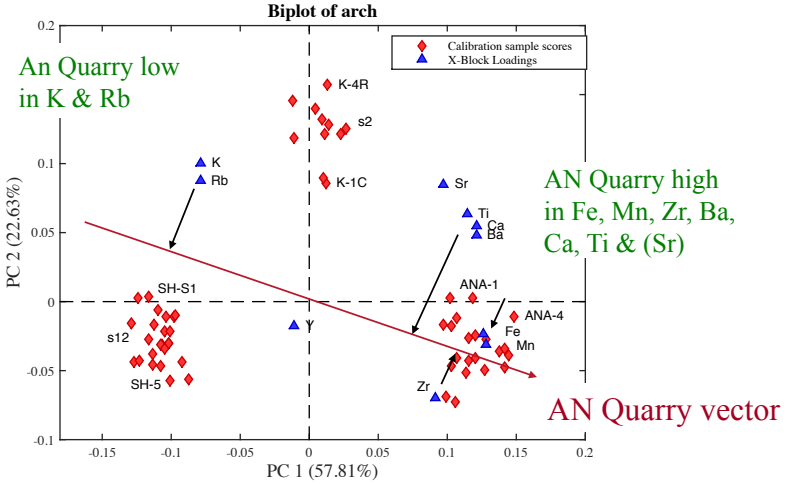
Biplot - Autoscaling



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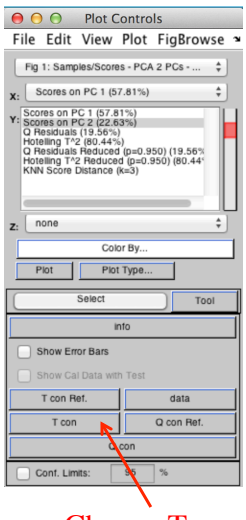
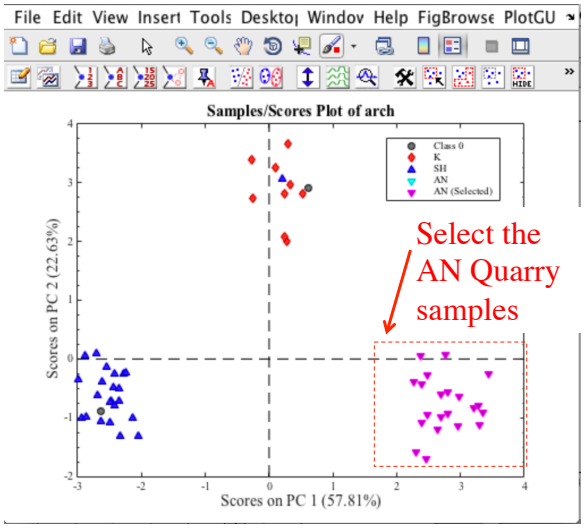
T- Contribution of Variables to Samples





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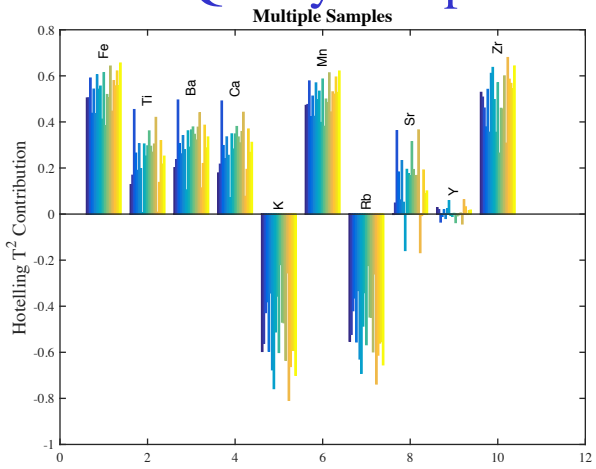
Variable Leverage of AN Quarry Samples



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What Can I See From Biplot or T-Con?

(In 2 PC - Autoscaled space)

- AN Quarry:
  - High in Fe, Mn & Zr
  - Moderately high in Ba, Ca, Ti & (Sr)
  - Low in K & Rb
- SH Quarry:
  - Low in Fe, Ti, Ba, Ca, Mn & Sr
- K Quarry:
  - High in K, Rb & Sr
  - Moderately high in Ti, Ba & Ca
  - Low in Zr

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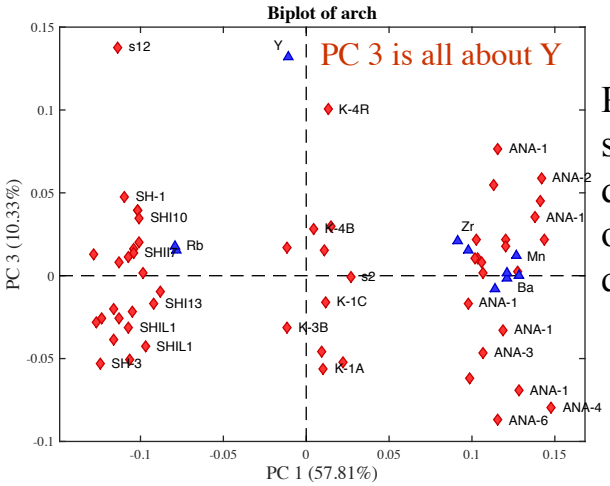
Also Check Higher PCs



PC 3 Scores show no discrimination of samples by quarry

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When Should We Mean Center or Autoscale?

- In exploratory analysis, nearly always mean center
  - Remember autoscaling includes mean centering.
- Autoscale when
  - variables are of different units *e.g.,* cm, g, C, atm, etc.
  - you are confident that low-signal variables still have good signal-to-noise ratios
  - not often in spectra - usually all same units *e.g.* abs
- Autoscaling weights each variable equally
  - Don't want to scale up noise.
- When in doubt, try it and see what happens

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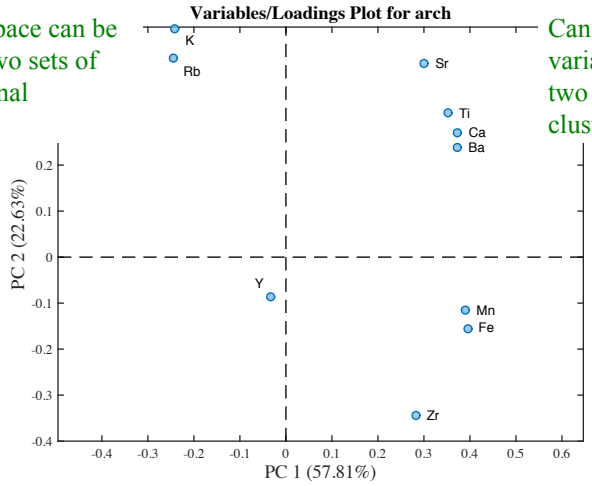
Examine Loadings Plot Again

Much of this space can be described by two sets of nearly orthogonal variables.



Can choose one variable from each of two ~ orthogonal clusters.

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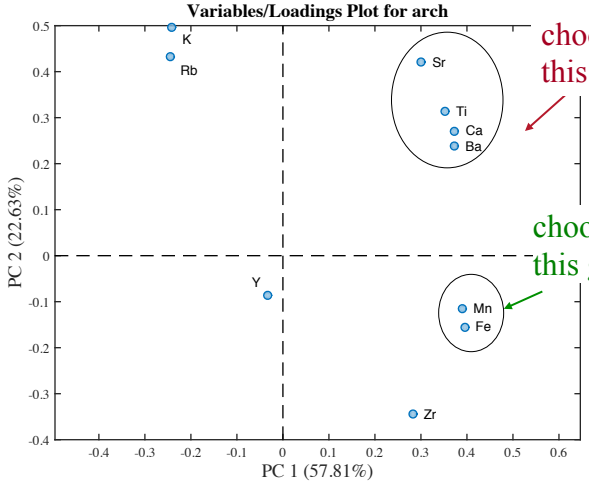
Examine Loadings Plot Again



choose Ca from this group

choose Fe from this group

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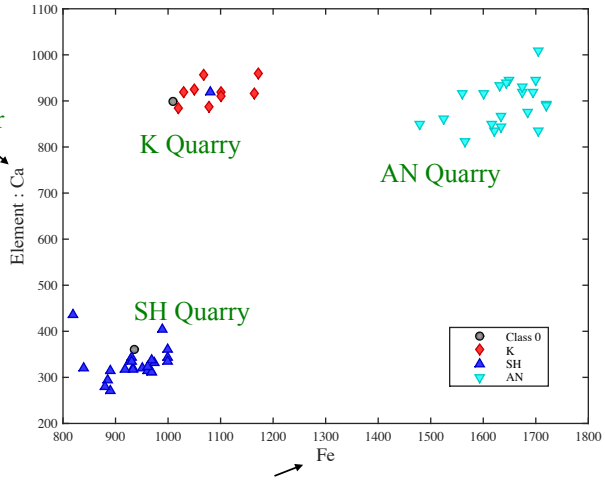
Plot [Ca] vs. [Fe] - Variable Selection

Could also use Sr, Ti or Ba here



Could also use Zr, Mn, Rb or K

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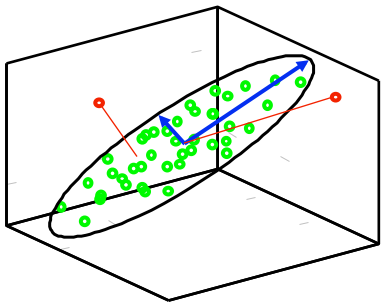
What Have We Accomplished?

- Concentrated discriminating power of 10 variables into 2 PCs containing 80% of the information using autoscaling
- Have determined characteristics of each quarry that differentiates it from other quarries.
- If lab analyses were difficult and/or expensive, found that we can differentiate quarries with just 2 variables.



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Yet a Little More Complicated Problem

- We wish to use FT-IR spectra and pattern recognition to distinguish authentic olive oil from counterfeit or adulterated olive oil.
- This time we shall see some special properties associated with Spectral Data.
- We shall also learn a new pattern recognition technique called SIMCA.

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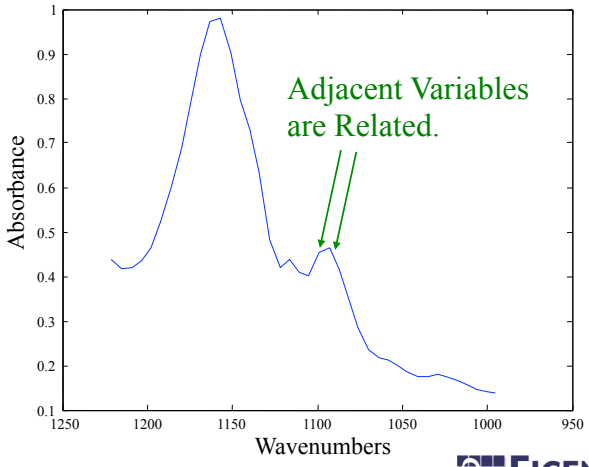
What is Special about Spectral Data?



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- PCA treats each variable as unrelated and unordered until it discovers relationships between variables.
- We could rearrange the order of the variables and PCA would not care.
- As chemists we know that absorbance in a spectrum must contain smoothly varying values.
- This will lead to special pretreatment techniques not available to other types of data.

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Olive Oil Samples
Learning Set:

Sample	Number of Samples	Sample Indices
Corn Oil	9	# 1-9
Olive Oil	15	# 10-24
Safflower Oil	8	# 25-32
Corn Margarine	4	# 33-36

Took FT-IR spectra (3600 - 600 cm^{-1}) of these oils using a fixed pathlength NaCl cell.

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Test Set:

Sample	No. Samples	Indices
Corn Oil*	9	# 1-9
Olive Oil*	15	# 10-24
Safflower Oil*	8	# 25-32
Corn Margarine*	4	# 33-36
Corn Oil in Olive Oil	5	# 37-41
5, 10, 20, 30 & 40%		
Almond Oil	1	(#42)
Peanut Oil	1	(#43)
Sesame Oil	1	(#44)

* New Samples

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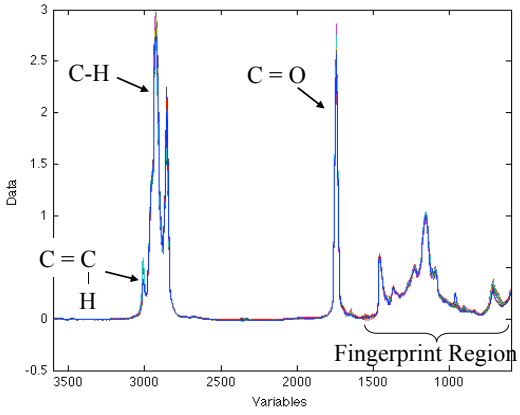
FTIR Spectra of 36 Sample Learning Set

Notice how spectra look alike.



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D.B. Dahlberg, et. al, *Applied Spectroscopy*, **51**, 1118 (1997).

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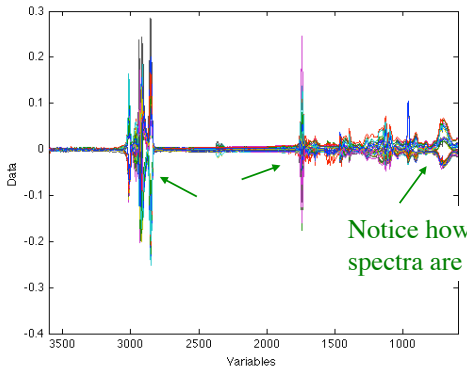
Do PCA on the Learning Set

- Examine full wavelength range
- Mean-center



Notice how differences between spectra are emphasized

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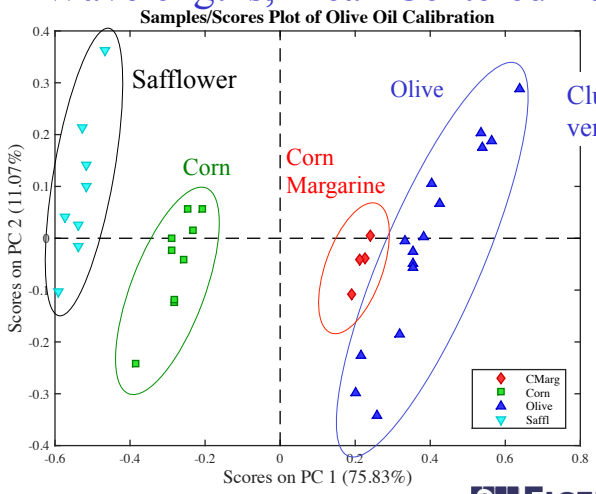
Scores Plot

All Wavelengths, Mean Centered Data



Clusters are not very tight

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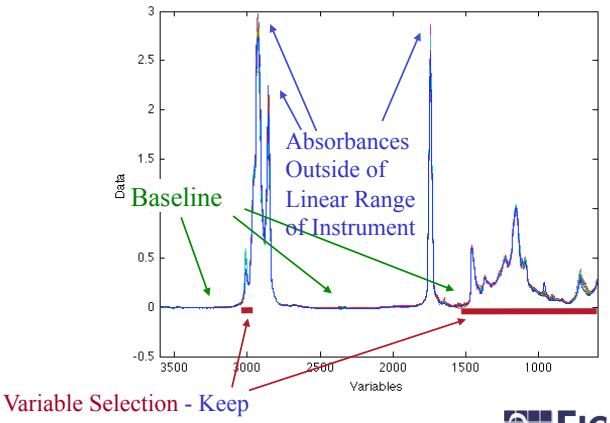
How Can the Model be Improved?



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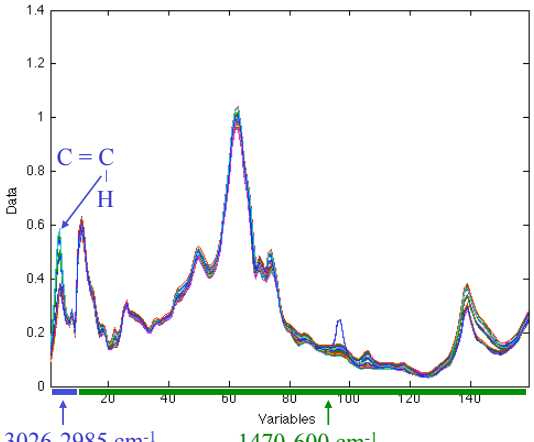
FTIR Spectra of Learning Set Selected Variables



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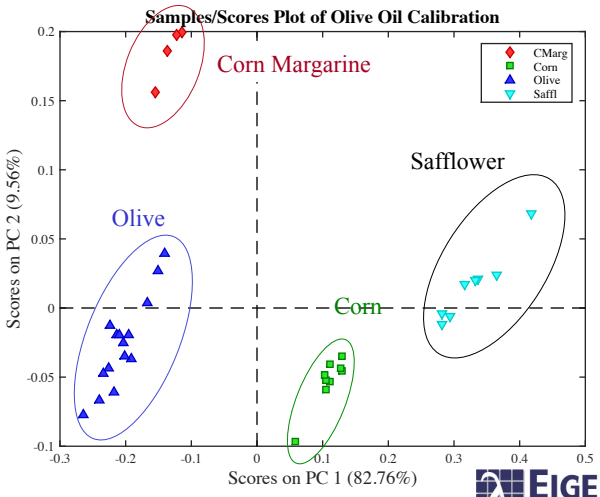
Scores Plot
Selected Wavelengths, Mean Centered Data



Better, but still too much spread

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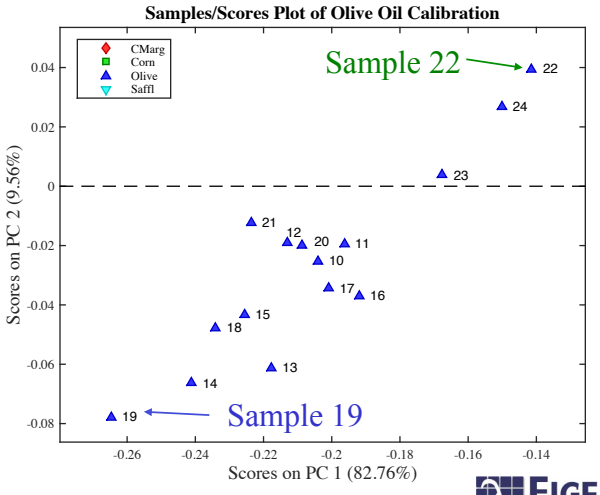
Scores Plot
Zoom In on Olive Samples



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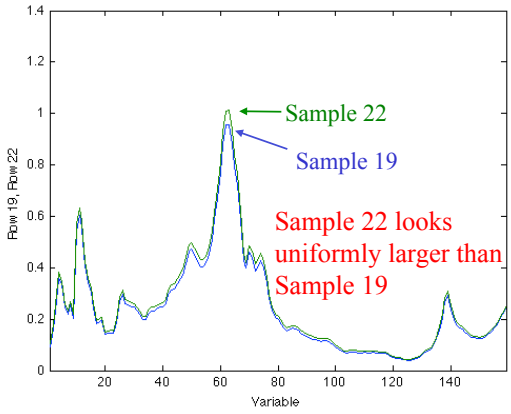
Spectra (Selected Wavelengths)
Samples 19 & 22



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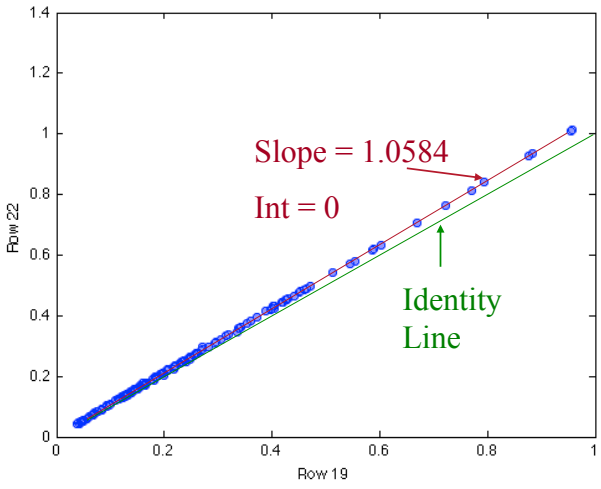
Plot Sample 22 vs. Sample 19



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Multiplicative Effect
Two Spectra are Identical except one is a Multiple of the Other

- Changing sample pathlength, *e.g.* changing light scattering with particle size.
- Changing sample density, *e.g.* changing temperature of sample.
- Changing gain of the instrument.

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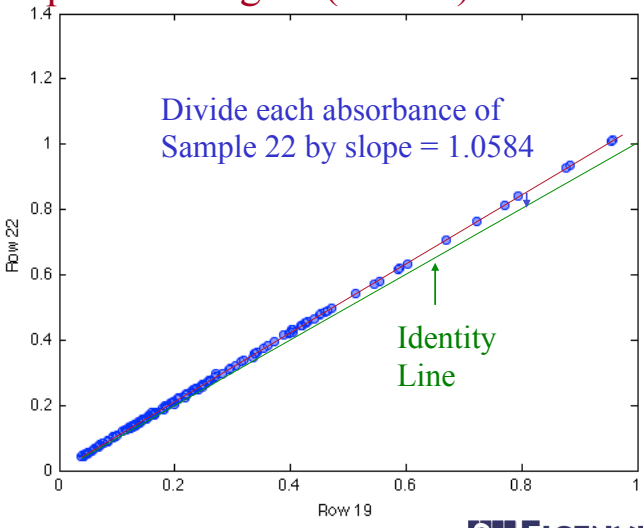
MSC
Multiplicative Signal (Scatter) Correction



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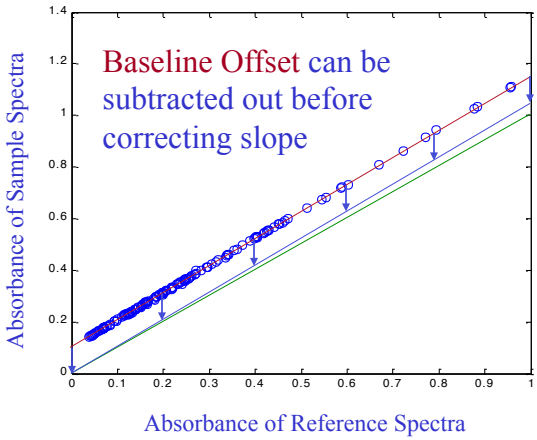
If there is also an Offset



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What to use as a Reference Spectrum?

- Anything we want that looks like the spectra in the Learning Set.
- Usually choose the Mean Spectrum of the Learning Set.
  - The same spectrum subtracted when doing mean centering of all samples, learning set, test set and real samples.

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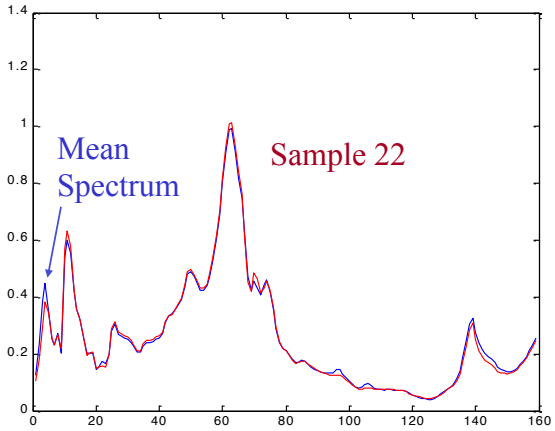
Mean Spectrum and Sample 22



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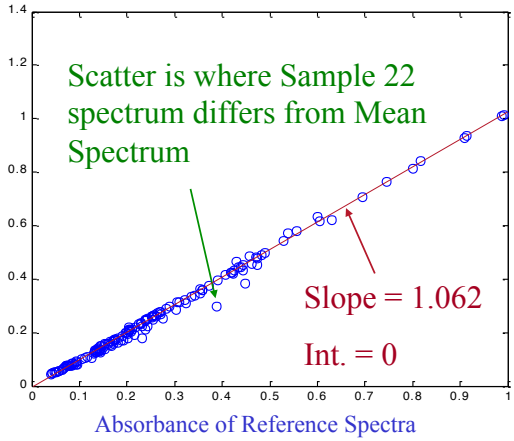
Sample 22 vs. Mean Spectrum



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Applying MSC

- Software will do MSC on each sample of the Learning Set
- You need to specify the Learning Set and the Reference Spectrum
  - usually mean spectrum of Learning Set

Geladi P, MacDougall D, Martens H., *Appl. Spectrosc.*, **39**(3), 491-500 (1985)

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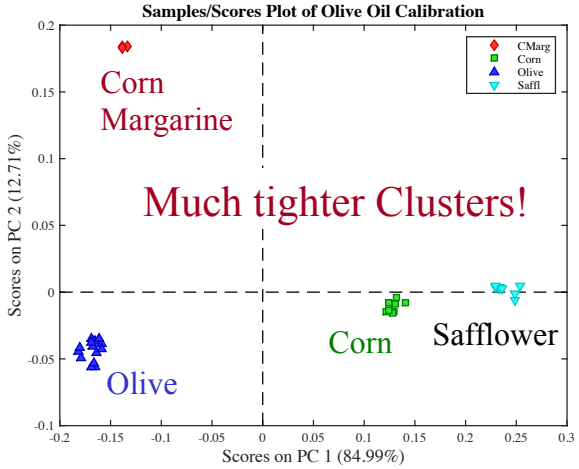
Scores Plot

Selected Wavelengths, MSC & Mean Centered



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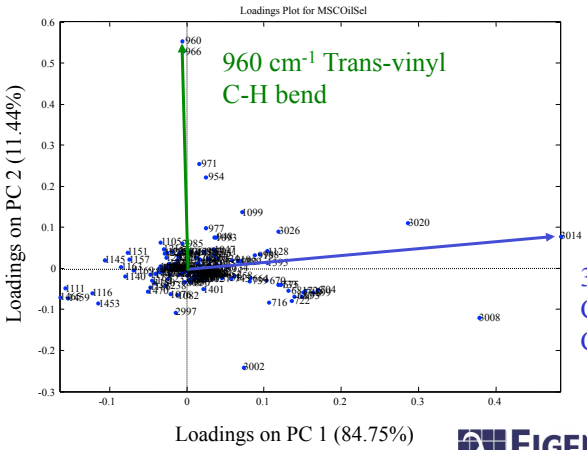
Loadings Plot
Selected Wavelengths, MSC & Mean Centered



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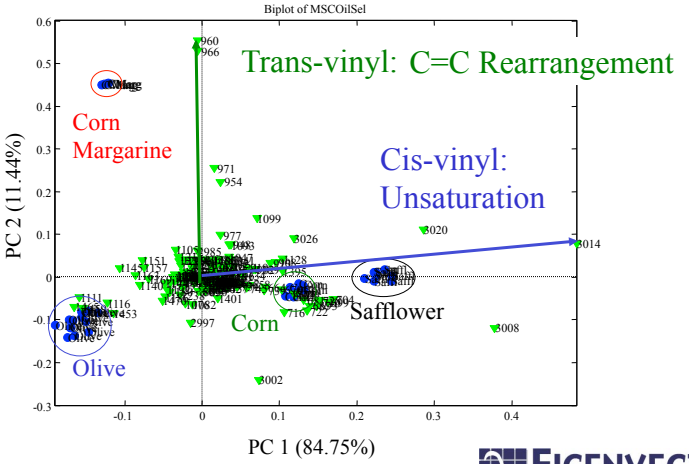
Biplot
Selected Wavelengths, MSC & Mean Centered



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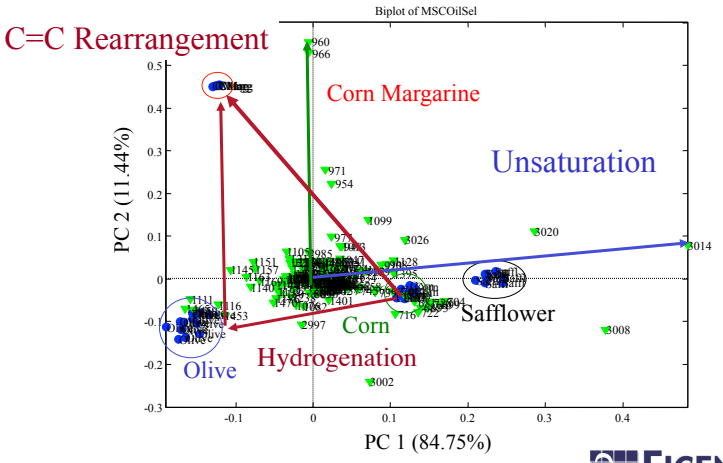
Biplot
Selected Wavelengths, MSC & Mean Centered



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Pattern Recognition

- Which samples are most like/unlike other Samples (Scores)
  - Outlier detection
  - Sample(s) most like target sample
  - Cluster analysis
- Which variables are most like/unlike other Variable (Loadings)
  - Variable selection
  - Detect underlying phenomena

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Pattern Recognition Can Relate Samples and Variables (Biplot)

- Determine which variables are responsible for sample similarities and differences.
  - Yields knowledge about a system
  - Leads to improvements in the system process, product, experimental improvements

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Can PCA be used to Detect Substitute or Adulterated Olive Oils?

- Bring in the Test Set
  - New samples of Olive, Corn, Corn Marg. & Safflower
  - Other Oils
  - 5, 10, 20, 30 & 40% Corn Oil in Olive Oil
- MSC
- Mean center
- PCA

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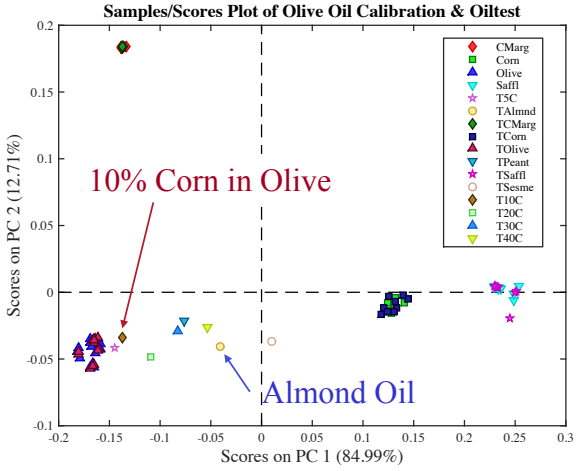
Score Plot Learning Set plus Unknowns "T"



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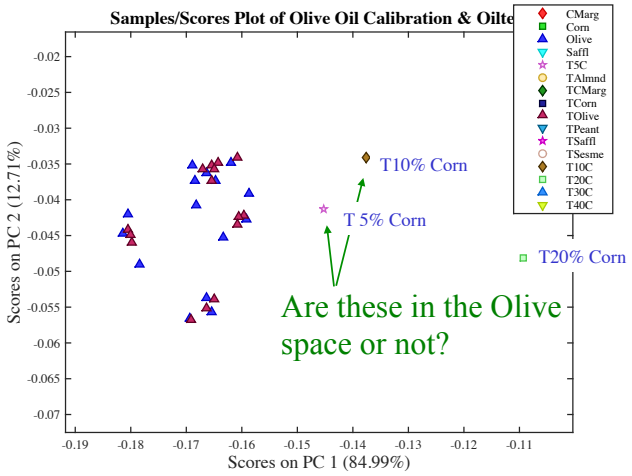
Closer View of Olive Oils



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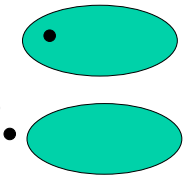
Need a method to put statistical Boundaries around the Olive Oil

If a sample falls within the boundaries, it is Olive Oil.

If a sample falls outside the boundaries, it is not Olive Oil.



One Technique is:

Soft Independent Method of Class Analogy

SIMCA

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Outline (Part 1)

- Introduction
- Pattern Recognition Motivation
- Principal Components Analysis
- SIMCA
  - Soft Independent Method Class Analogy
- Summary

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How SIMCA works

- Perform PCA on a Learning Set representing ONE class, *e.g.* Olive Oil
- Verify that you have a proper Learning Set.
  - samples are representative of all olive oils to be classed

How?

- Choose the number of PCs that are sufficient to describe this set.
  - This is the Model of that class.
- Set statistical Confidence Limits, *i.e.* boundaries for the Model.
- Determine if an unknown lies within or outside these limits.
- Repeat this process for Each class.

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Determine the Number of PCs That Are Sufficient to Describe Olive Oil

Perhaps You Know Your Analytical Technique is Good to ± 10%

Where does the Model end and the noise begin?

What is the Rank of the data?

	Eigenvalue of Cov(X)	% Variance This PC	% Variance Cumulative
1	2.72e-04	40.43	40.43
2	2.07e-04	30.80	71.23
3	7.08e-05	10.51	81.74
4	6.58e-05	9.77	91.51
5	2.71e-05	4.02	95.53
6	1.02e-05	1.51	97.04
7	9.45e-06	1.40	98.45
8	3.30e-06	0.49	98.94
9	2.67e-06	0.40	99.33
10	1.64e-06	0.24	99.58
11	9.75e-07	0.14	99.72
12	7.73e-07	0.11	99.84
13	5.69e-07	0.08	99.92
14	5.31e-07	0.08	100.00

Terminate Model with 3 PCs

Beginning to Model 10% Noise

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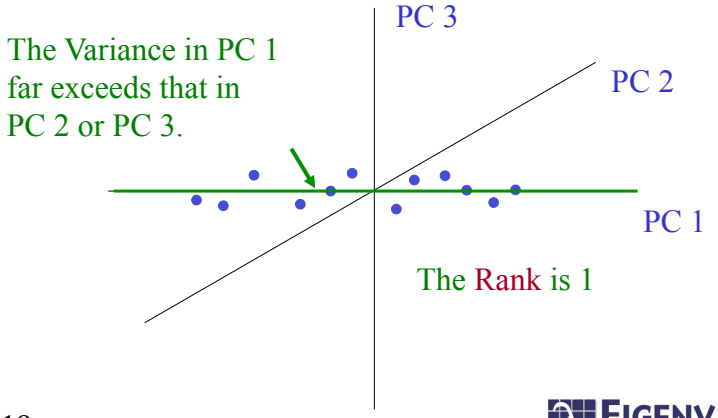
Another Approach is to Examine Distribution of Variance among the PCs



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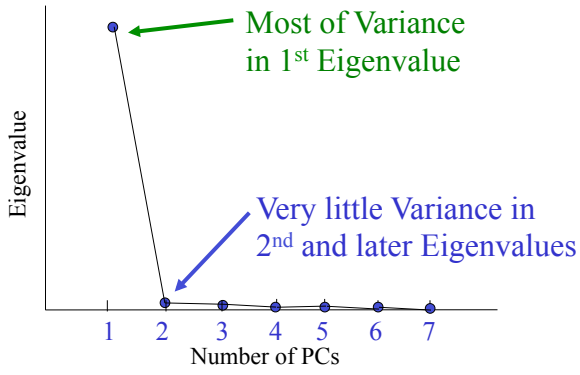
Plot Eigenvalues



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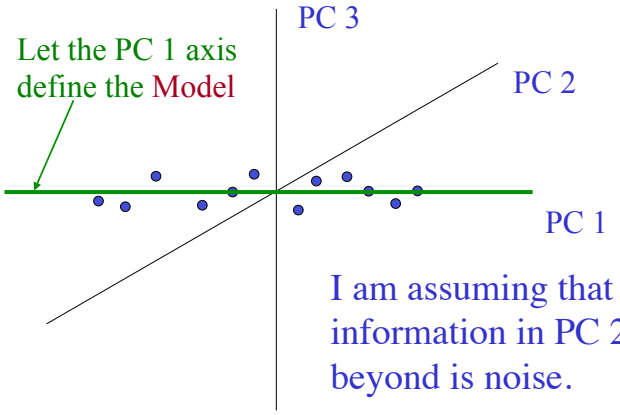
Create One PC Model with Boundaries



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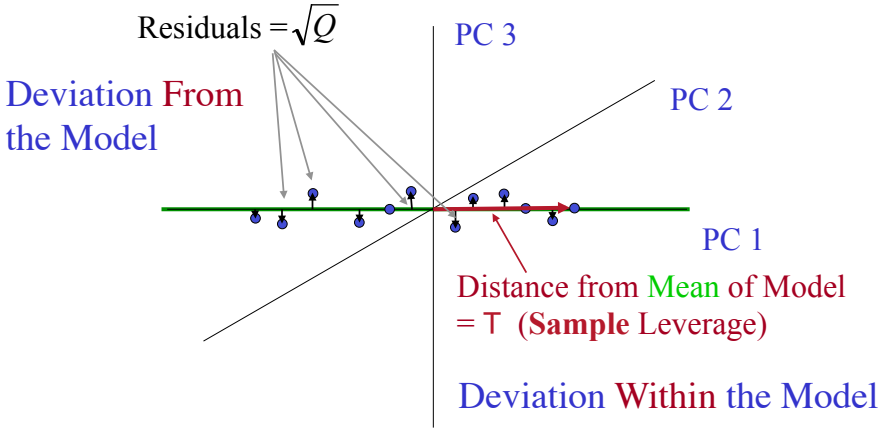
Individual Points Deviate from the Group in Two Ways



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Methods of Measuring Distance

- Euclidean Distance: Shortest distance between two samples. Here we're interested in the distance from the sample to the mean.
- Mahalanobis Distance: Distance scaled along each axis by dividing by the Standard Deviation of that axis corrected for the covariance between the axes. (this is also how T^2 is calculated).
- Mahalanobis Distance has several statistical advantages over Euclidean Distance.

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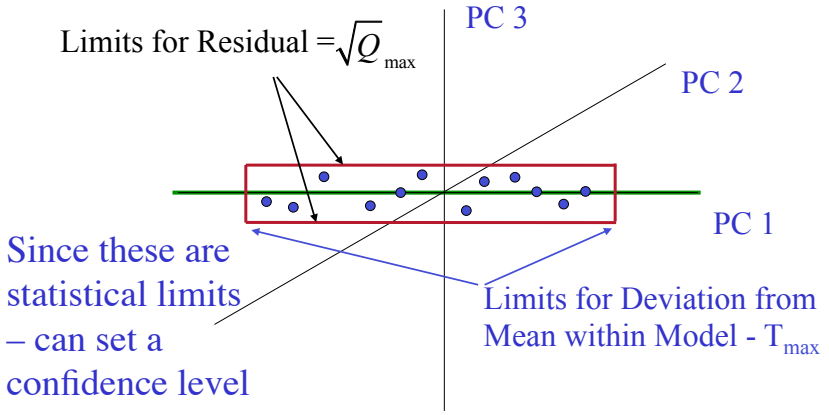
Set Class Boundaries Based on Deviation of Learning Set from Idealized Model



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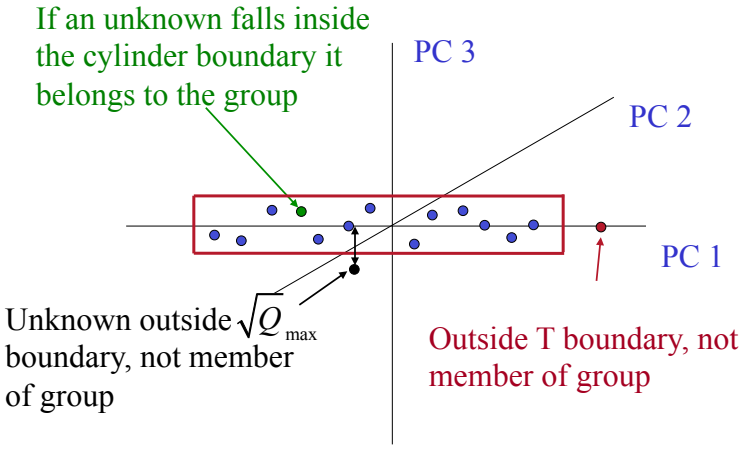
Place Unknown into PC Space



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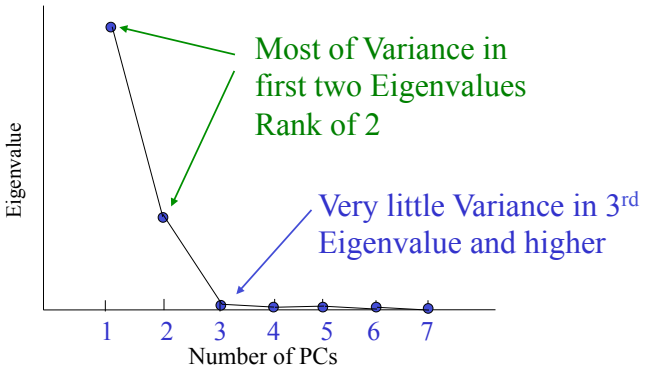
What If the Eigenvalue Plot Looks Like This:



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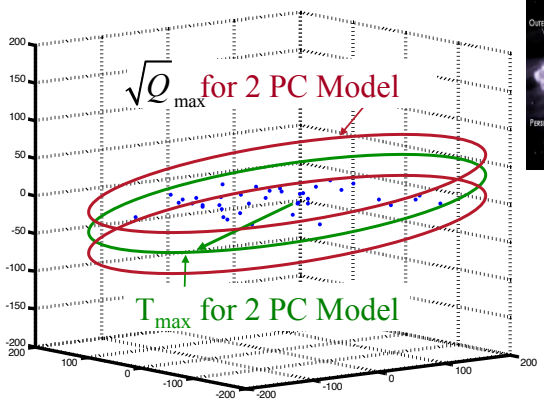
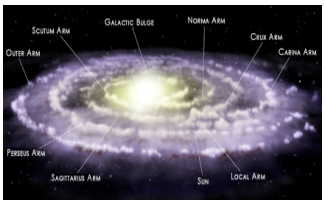
Use Two PC Model





How many PCs would you use to describe the distribution of stars in the Milky Way?

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An Additional Method to Determine the Number of PCs to Keep

Cross Validation

What is the best method to study for a chemistry exam?

Use homework problems to make up practice exams

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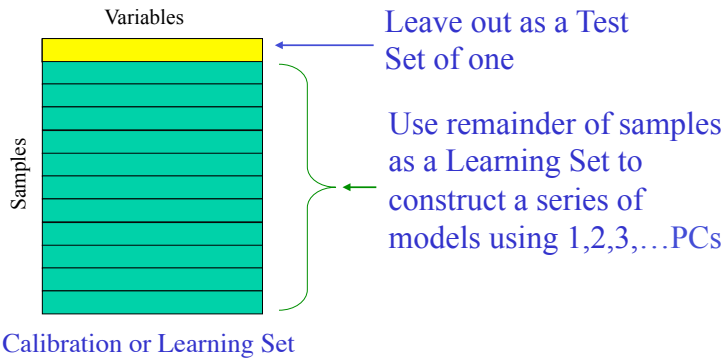
Many Patterns of Cross-Validation

- Leave-One-Out Cross-Validation



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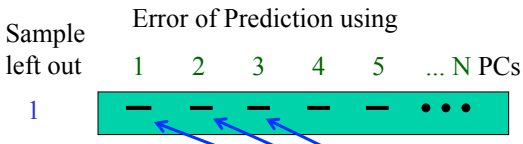
Leave One Out Cross-Validation

For each of these models uses the scores and loading to predict the values of variables (e.g. the spectra) for the sample left out.



$$Error^2 = \| X_{observed} - X_{pred}(\text{using } 1,2,3\ldots PC\text{'s}) \|^2$$

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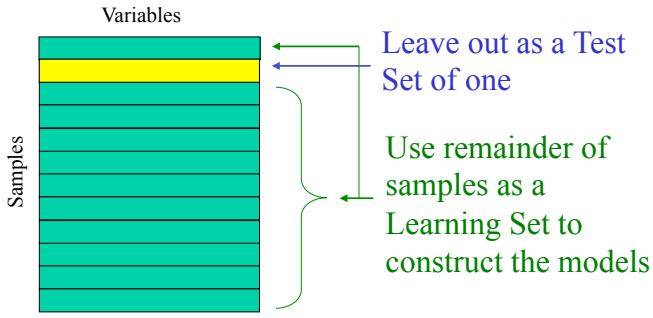
Many Patterns of Cross-Validation

- Leave-One-Out Cross-Validation



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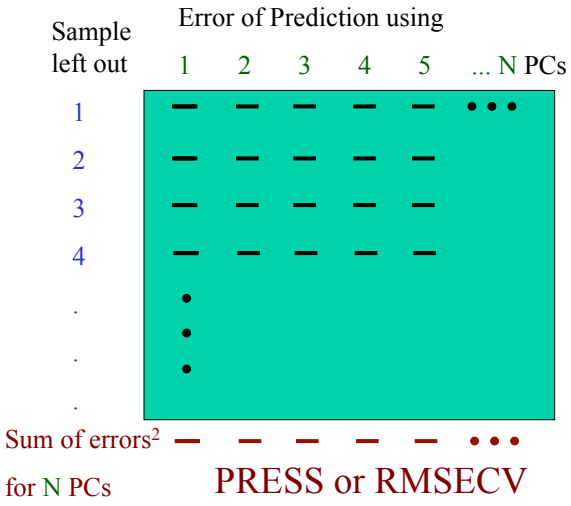
Leave One Out Cross-Validation



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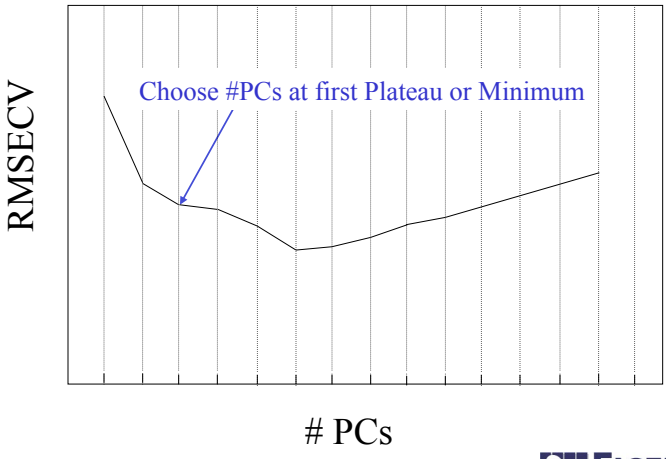
Construct PRESS Plot



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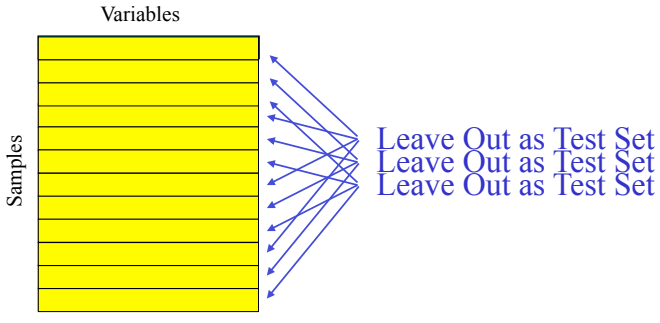
Many Patterns of Cross-Validation

- Leave-One-Out Cross-Validation
- Venetian Blind



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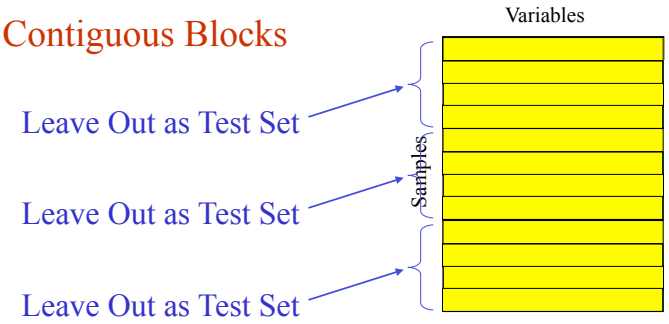
Many Patterns of Cross-Validation

- Leave-One-Out Cross-Validation
- Venetian Blind
- Contiguous Blocks



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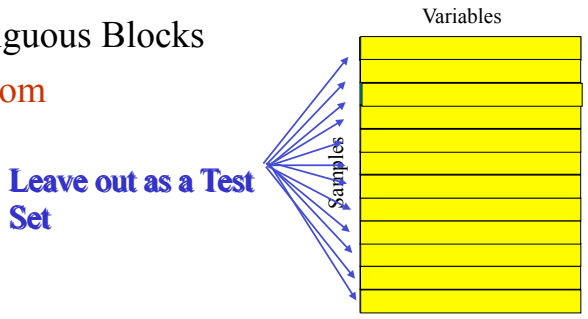
Many Patterns of Cross-Validation

- Leave-One-Out Cross-Validation
- Venetian Blind
- Contiguous Blocks
- Random



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Which CV method do I use?

- Use Leave-one-out CV for fewer than about 16 samples in the Calibration (learning) Set.
- Your choice for larger Calibration Sets.
- Number of Splits = approximately the square root of the number of samples (24 sample, use 5 splits)
  - $1/5^{th}$ of the data is left out at a time
- Make sure that a representative set is always left behind to construct good models.

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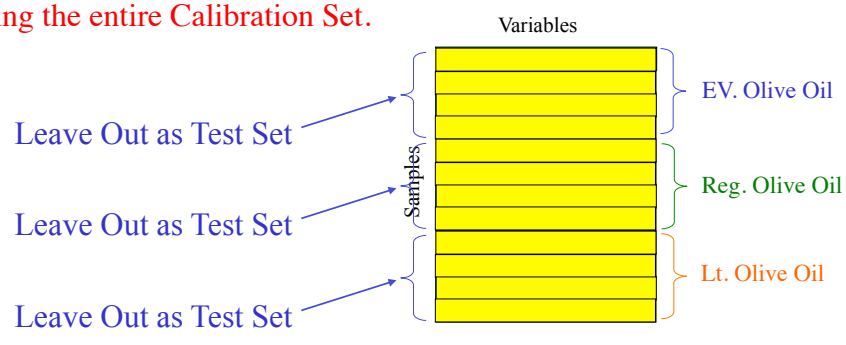
For example, if you chose Contiguous Blocks for Olive Oil Model

Once the number of PCs has been determined, the model is created using the entire Calibration Set.



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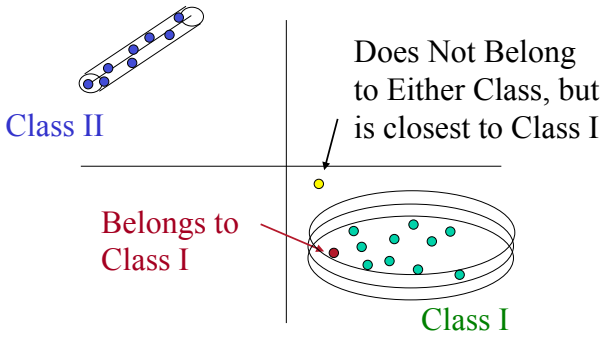
SIMCA tells if a Sample falls within the Boundaries of a Class



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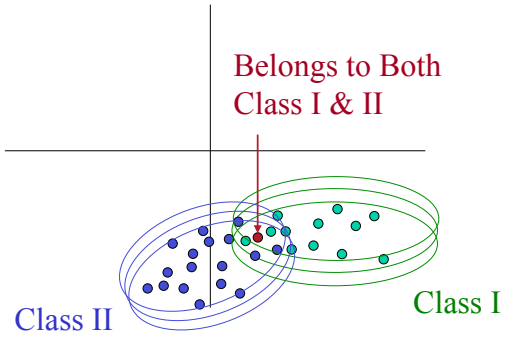
With SIMCA - A Sample May Belong to Two or More Classes at the Same Time



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You Can Change the Rules for Failing to Be in the Group

- Outside T only
- Outside Q only
- Either Outside T or Outside Q
- Outside some combined limit, *e.g.* $\sqrt{(T^2 + Q^2)}$

These pictures have been using this rule

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According to these Rules SIMCA is the Only Modern Classification Method that Allows a Sample to Belong to More than One Class or No Class at All

But Rules are Made to Be Broken

Or at least changed according to our needs

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Sample Classification Predictions

- Class Prediction Member by Class
  - Class Prediction Member – each class
  - Class Prediction Member – unassigned
  - Class Prediction Member – multiple

  } Traditional SIMCA Rules
- Class Prediction Strict
- Class Prediction Most Probable
- Class Prediction Probability by Class

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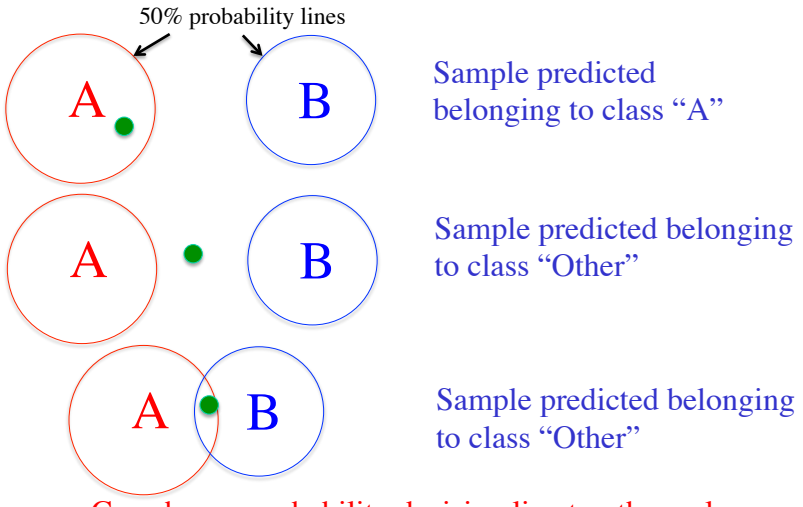
Class Prediction Strict



Can change probability decision line to other values

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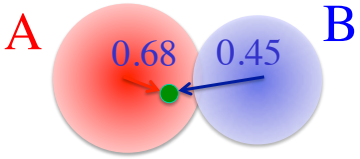
Class Prediction Most Probable



Sample predicted more probable belonging to class "A" than to class "B"

A "soft class" model

Prob A=90% & Prob B=89%, Then assigned to Class "A"

Prob A=0.02% & Prob B=0.01%, Then assigned to Class "A"

"Multiple" Class and "Unassigned" Class assignments not allowed

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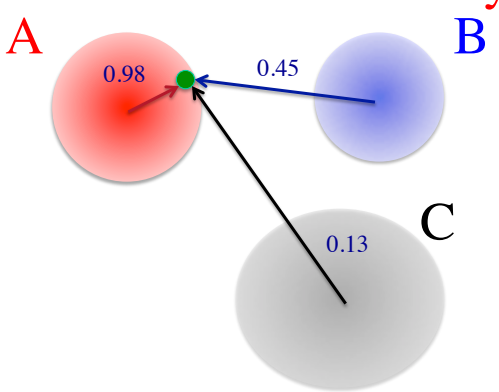
Class Prediction Probability by Class



Probability calculated for each class.

Useful when you need to report a confidence of assignment or need to derive special rules for class assignment.

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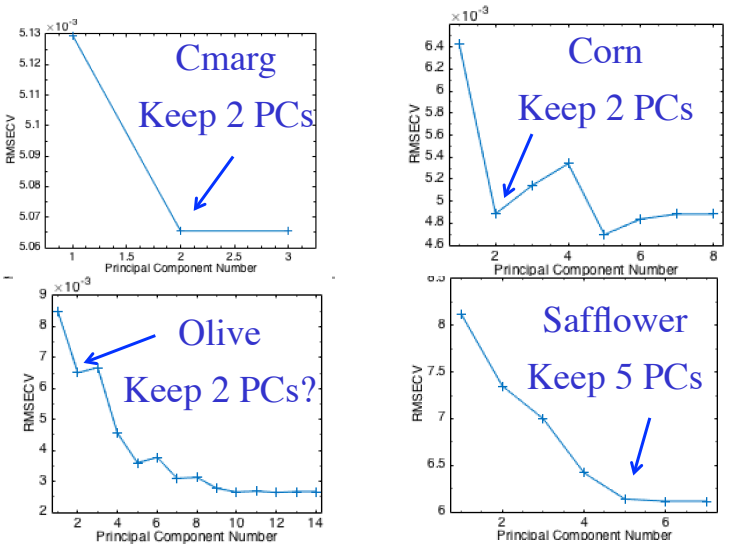
PRESS Plots for Each Class



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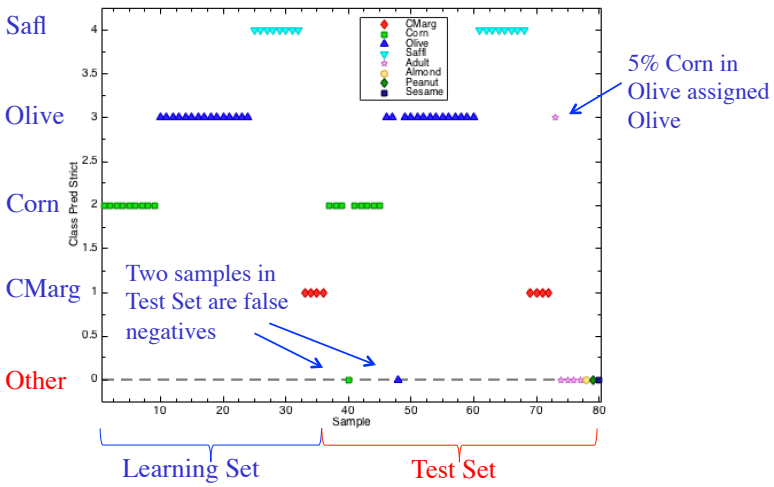
Strict Prediction



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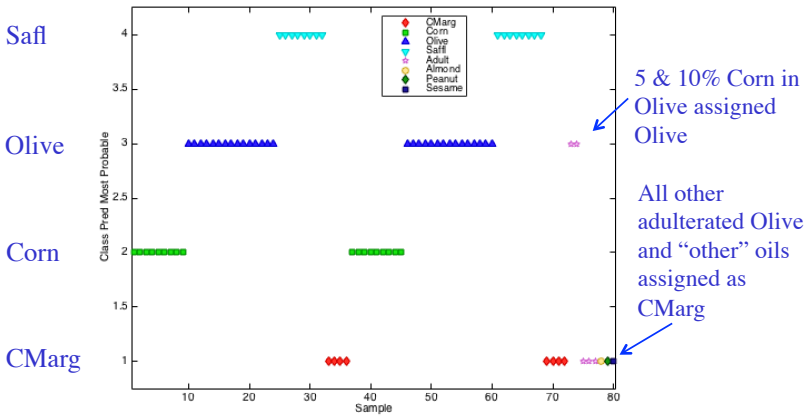
Most Probable



Why is this not useful for this data?

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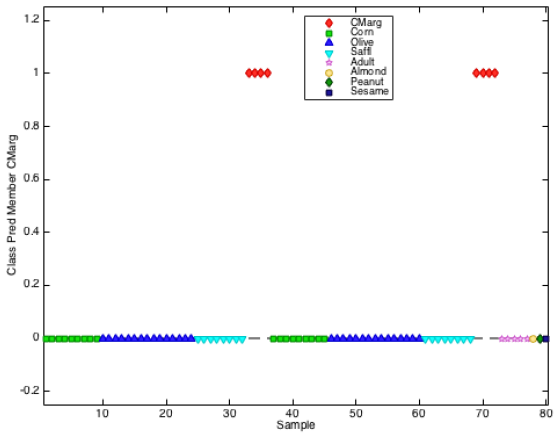
Class Predicted Member CMarg



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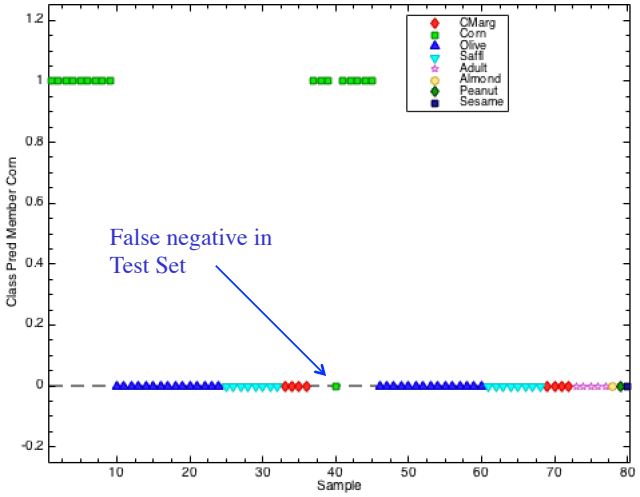
Class Predicted Member Corn



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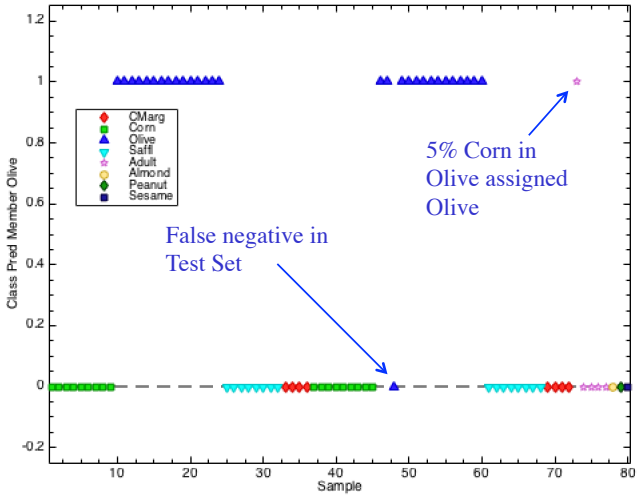
Class Predicted Member Olive



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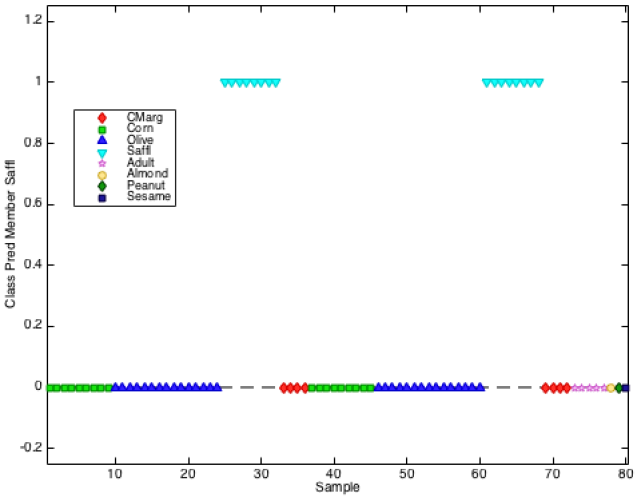
Class Predicted Member Safflower



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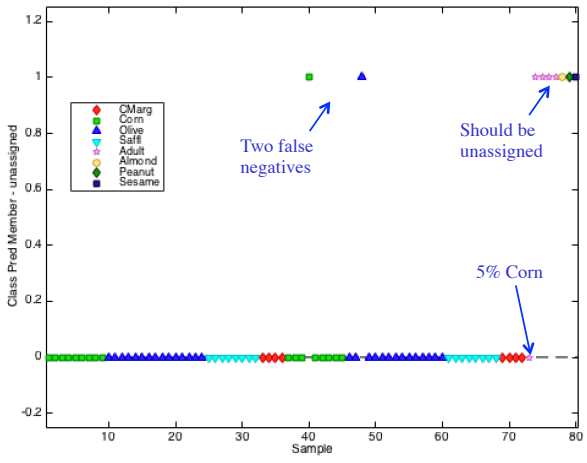
Class Predicted Member Unassigned



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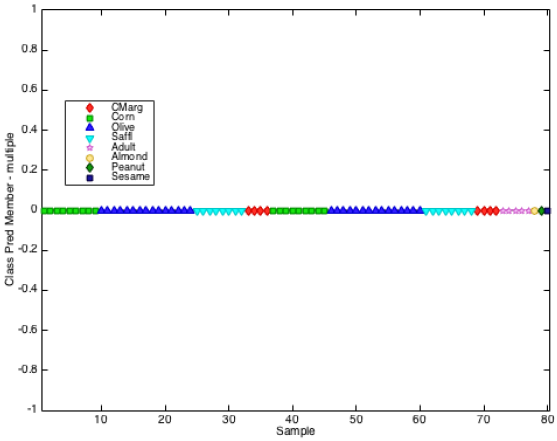
Class Predicted Member Multiple



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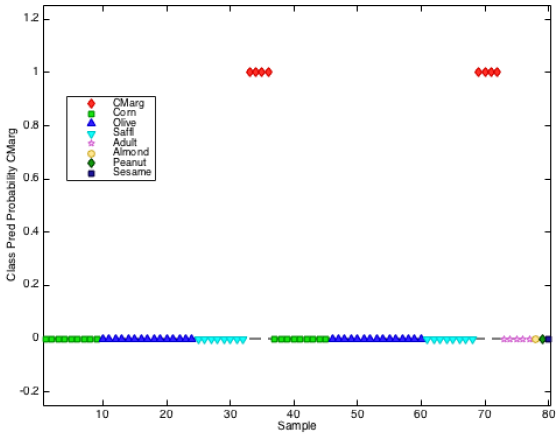
Class Predicted Probability CMarg



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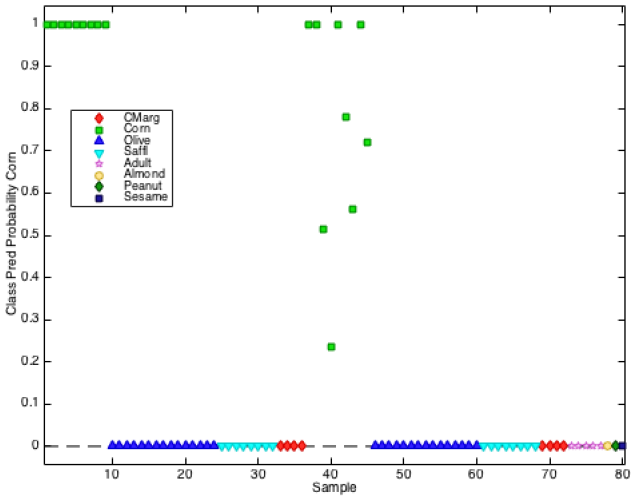
Class Predicted Probability Corn



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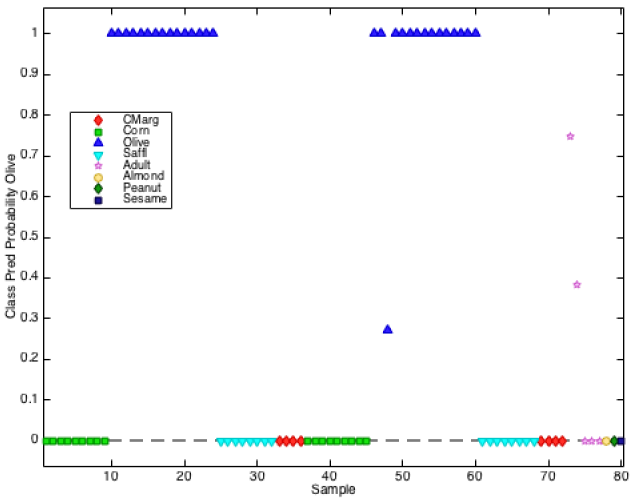
Class Predicted Probability Olive



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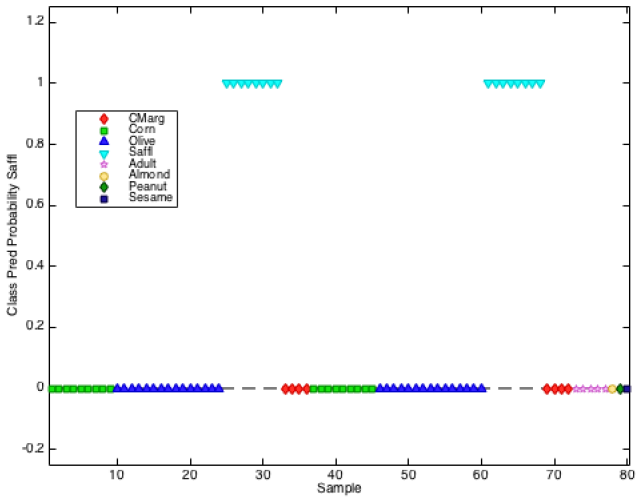
Class Predicted Probability Safflower



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Important Notice: Before Applying SIMCA Models to Real Unknowns

- Play with the number of PCs in the models to see if you can decrease False Negatives without increasing False Positives.
- Determine if you would rather live with false negatives or false positives.
- Validate Thoroughly With a Well Designed, Independent Test Set!
- Models Do Not Last Forever.
- Re-validate Often and Rebuild If Necessary.

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Outline (Part 2)

- Regression Motivation & Rational
  - What is Regression?
  - Why use Regression?
- Classical Least Squares (CLS)
- Multiple Linear Regression (MLR)
- Principal Components Regression (PCR)
- Partial Least Squares Regression (PLS)
- Summary

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We Can't Always Measure What We Want

- Often measurements must be made on something else and the property of interest must be inferred from these measurements.
- This is the idea behind inferential sensing where we measure variables that are available in a timely manner to predict something that is more difficult (or more expensive) to obtain.

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What's Measured:

- Volume of $AgNO_3$ titrant to get moles of chloride in the sample.
- Absorbance at 254 nm to get the concentration of benzene in solution.
- NIR spectrum of gasoline to yield octane number.

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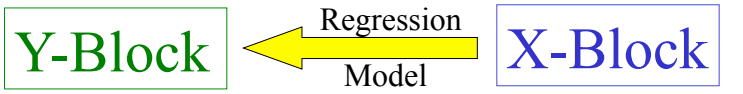
Regression



What's desired

What's measured

Regression analysis identifies the dependency between two blocks of data.

Regression models are often used to obtain estimates (or predictions) for one block of data from the other.

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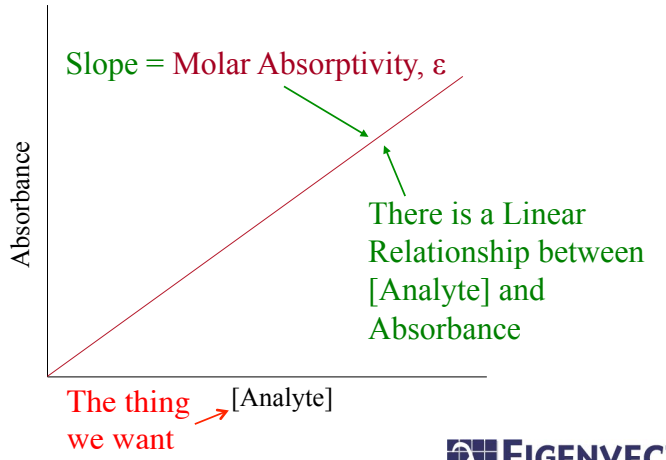
One of the Most Familiar uses of Linear Regression is from the Beer-Lambert Law



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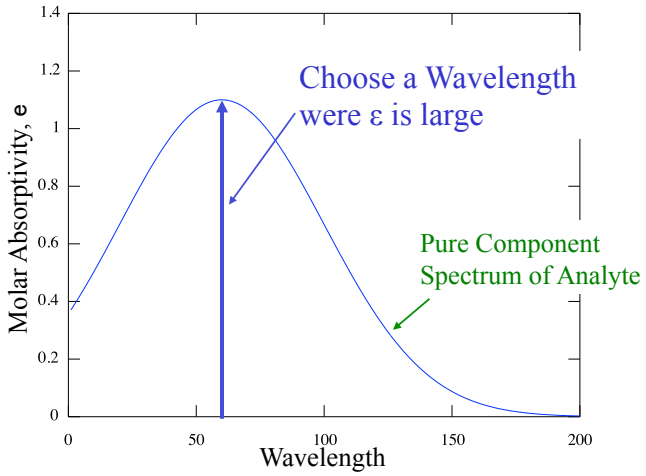
How to Create Beer-Lambert Law Model



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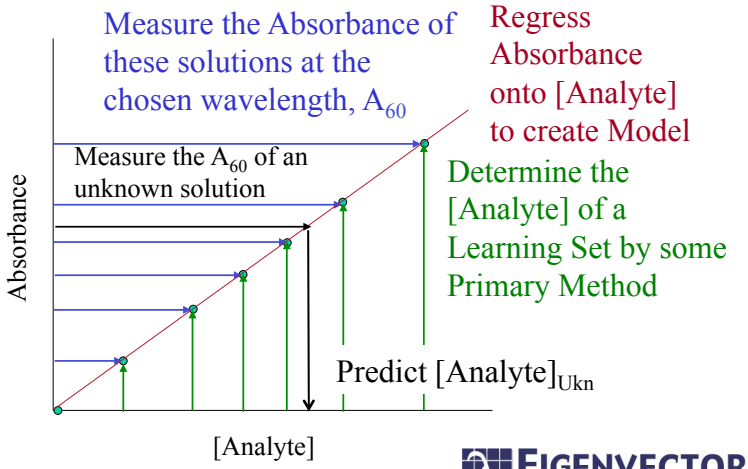
Create Beer-Lambert Law Model



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Outline

- Regression Motivation & Rational
- Classical Least Squares (CLS)
- Multiple Linear Regression (MLR)
- Principal Components Regression (PCR)
- Partial Least Squares Regression (PLS)
- Summary

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Sometimes Must Look at an Equation

Classical Least Squares Model = CLS:

$$A_{60} = [Analyte]\varepsilon + Error$$

Dependent Variable

Independent Variable

Regression Coefficient

Error in measuring A

Notice the thing we ultimately want is on the right side of the equal sign.

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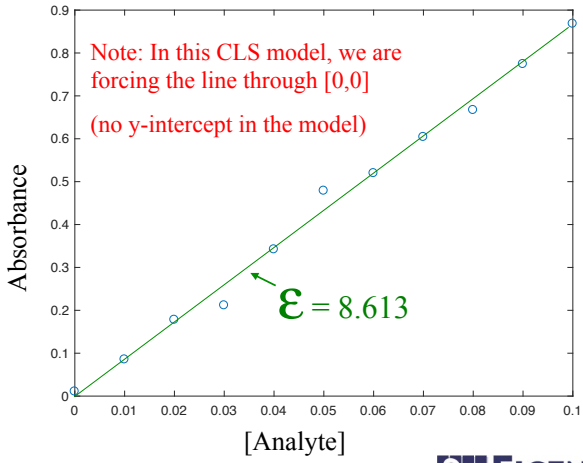
Not All Samples are Equal



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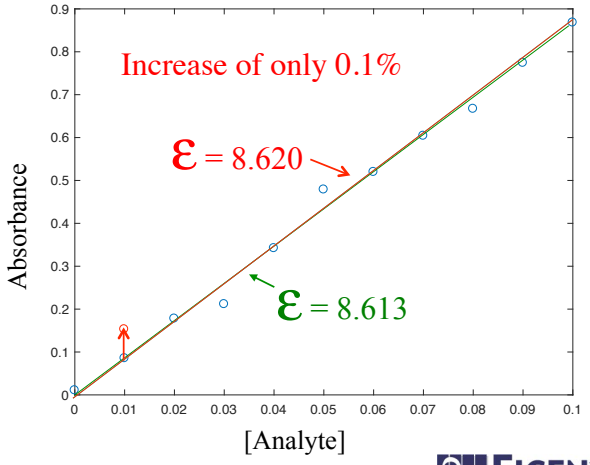
What Happens to the Slope if a Point is Moved Due to Measurement Error?



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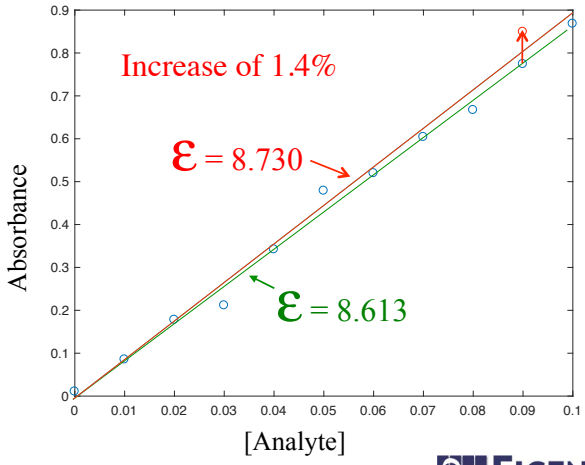
What Happens to the Slope if a Point is Moved Due to Measurement Error?



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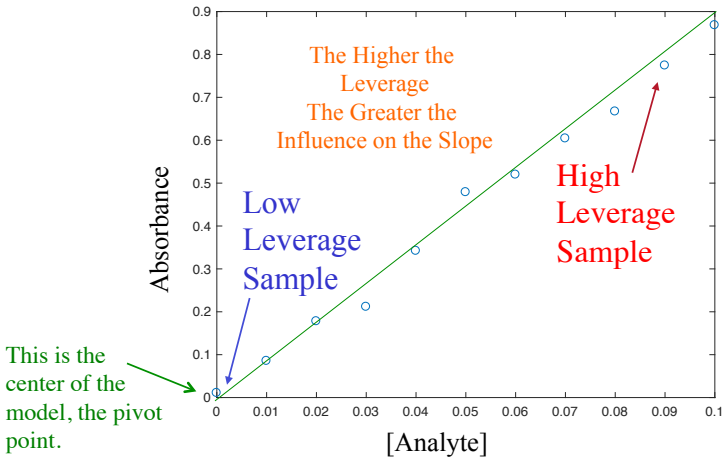
It's About Sample Leverage



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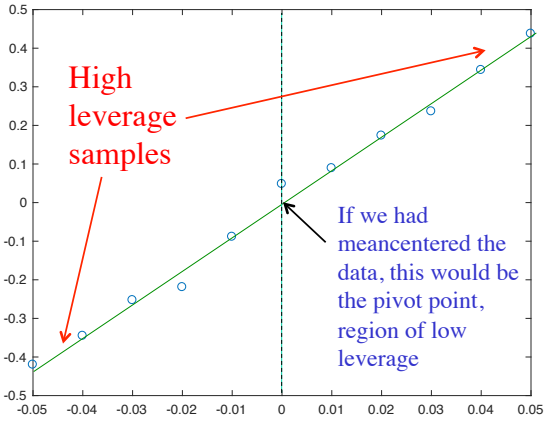
It is Important to Know the Location of the Center of the Model



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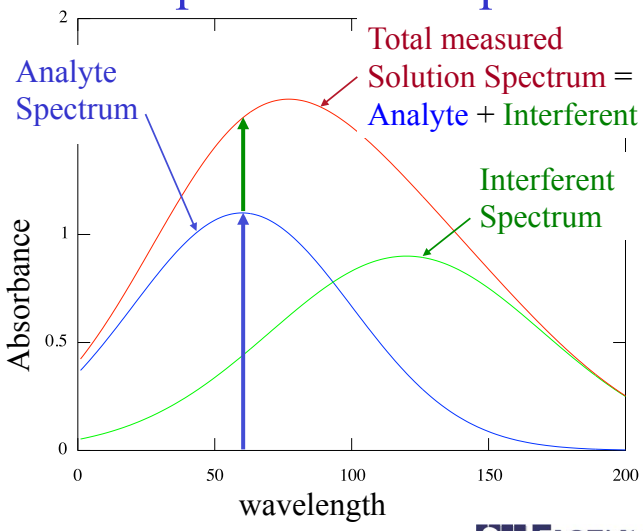
Spectral Overlap



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Beer-Lambert Law Now Looks Like:

$$A_{60} = [\text{Analyte}]\,\varepsilon_{\text{Analyte}} + [\text{Interf.}]\,\varepsilon_{\text{Intrf}}$$

Have two Concentrations to worry about

Have two Molar Absorptivities to Determine

Two Unknown Constants will require at least two equations

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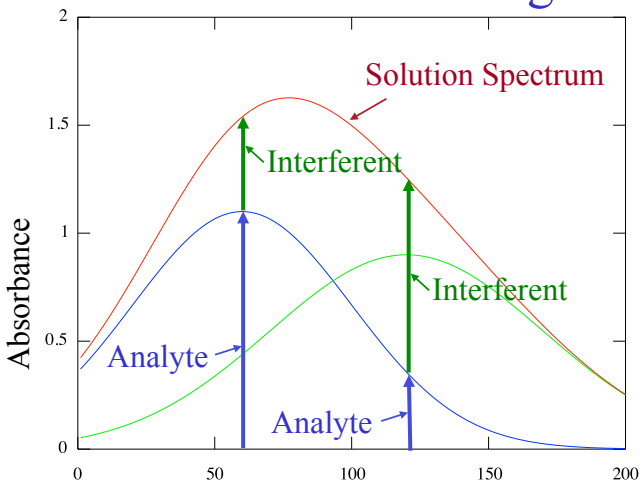
What to Do?
Choose a 2nd Wavelength



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Now Have Two Equations

$$A_{60} = [\text{Analyte}]\,\varepsilon_{\text{Analyte},60} + [\text{Interf.}]\,\varepsilon_{\text{Intrf},60}$$

$$A_{120} = [\text{Analyte}]\,\varepsilon_{\text{Analyte},120} + [\text{Interf.}]\,\varepsilon_{\text{Intrf},120}$$

And FOUR Unknowns to Determine

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What to Do?

$$A_{60,I} = [\text{Analyte}]_I \, \varepsilon_{\text{Analyte},60} + [\text{Interf.}]_I \, \varepsilon_{\text{Intrf},60}$$

$$A_{120,I} = [\text{Analyte}]_I \, \varepsilon_{\text{Analyte},120} + [\text{Interf.}]_I \, \varepsilon_{\text{Intrf},120}$$

Make up a 2nd Solution using different concentrations:

$$A_{60,II} = [\text{Analyte}]_{II} \, \varepsilon_{\text{Analyte},60} + [\text{Interf.}]_{II} \, \varepsilon_{\text{Intrf},60}$$

$$A_{120,II} = [\text{Analyte}]_{II} \, \varepsilon_{\text{Analyte},120} + [\text{Interf.}]_{II} \, \varepsilon_{\text{Intrf},120}$$

Solve the equations simultaneously for the four εs

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Hey, I Thought This Workshop Was without Equations!?

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Don't Worry About It
The computer will do the work

Example:

- Make up two solutions of known [Analyte] & [Interferent] (four known concentrations).
- Measure Absorbance of both solutions at two wavelengths.
- CLS will give you the four Molar Absorptivities.
- Measure Absorbance of unknown solution at two wavelengths.
- CLS will estimate concentrations of both species/analytes in the unknown.

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Which Wavelengths?

- Can use (almost) all of them.
  - don't use wavelengths that can't be trusted
    - *e.g.* out of dynamic range or pure noise
- CLS is a Full Spectrum Technique
  - Multi-channel Advantage: Redundant information of many wavelengths averages out much of the noise
  - Using all channels leads to estimates of "pure component spectra" of all the chromophores
    - No extra charge!

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Which Samples?

- Experimental Design
  - very useful, many books on the topic
- Don't vary concentrations in the same manner
  - CLS won't work if the concentrations are correlated
- Noise can be reduced by using more than the minimum number of samples in the Learning Set
  - perhaps 5 samples per chromophore

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Sixteen Solution Learning Set
Three Component System

Component:	[A]	[B]	[C]
1	0.2845	0.5406	0.1749
2	0.4560	0.3484	0.1956
3	0.3858	0.5355	0.0786
4	0.5847	0.3213	0.0940
5	0.3109	0.2591	0.4299
6	0.2501	0.3805	0.3694
7	0.4772	0.5151	0.0076
8	0.5142	0.3508	0.1350
•	•	•	•
•	•	•	•
•	•	•	•
16	0.3830	0.5563	0.0608

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Sample Dilemma

- You want samples to cover the widest area of sample space in order to have many high leverage samples (far from the mean).
- You do not want to cover too much sample space in order not to deviate from a linear model (*e.g.* deviate from Beer's Law).
- Out of spec samples often have to be produced in the pilot plant or laboratory and may contain artifacts not present in plant samples.

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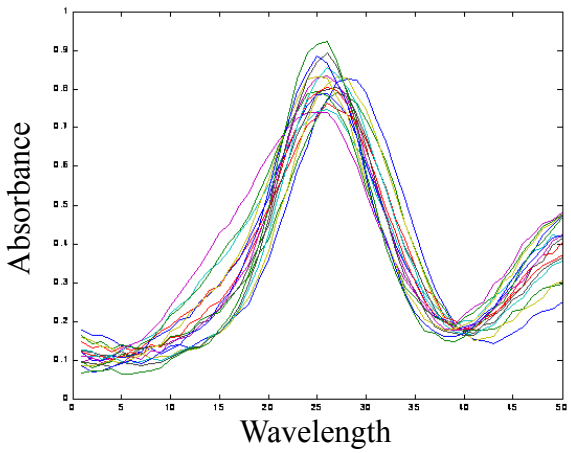
Spectra of 16 Learning Set Spectra



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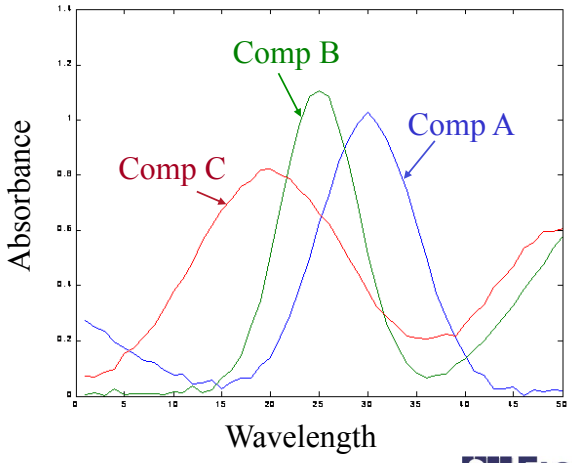
Pure Component Spectra Obtained from CLS



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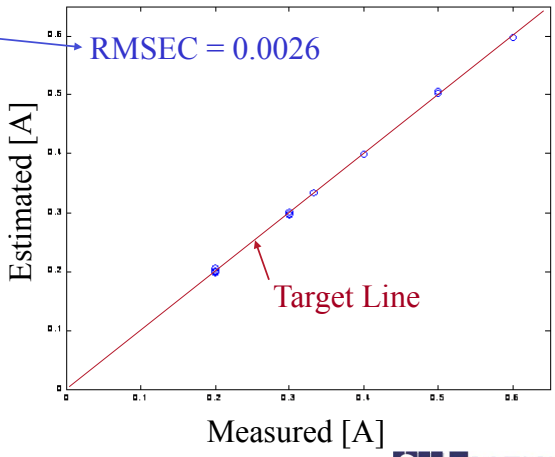
Estimated vs. Measured [A] for Learning Set

Root Mean Square Error of Calibration



Estimated because Model learned from these samples

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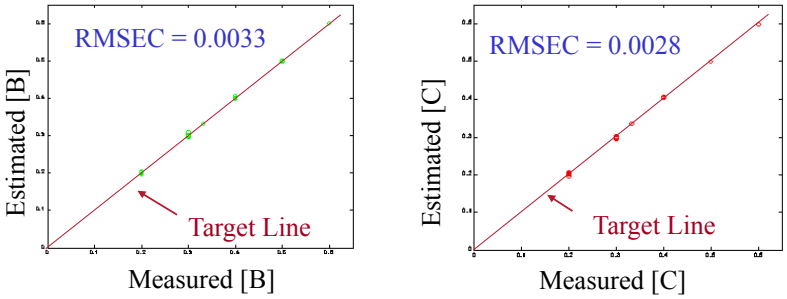
Estimated vs. Measured [B] and [C] for Learning Set



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Estimation versus Prediction

- The previous slides were for the Learning Set samples
  - This provides an estimate of the fit or calibration error
- But the real test of any model is how it does on samples it has **Never Seen Before**

→ Test Set

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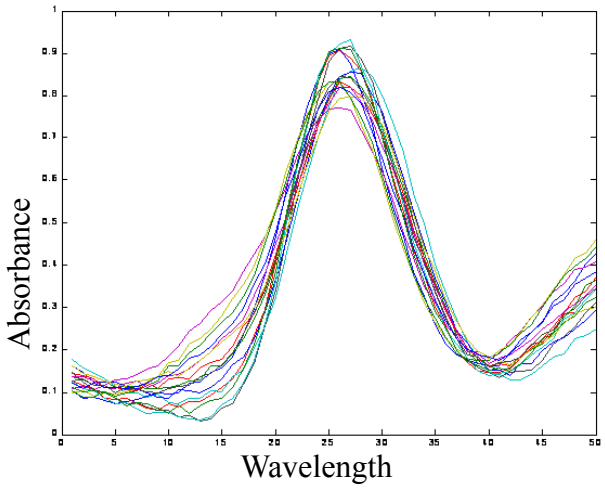
Spectra of 16 Sample Test Set



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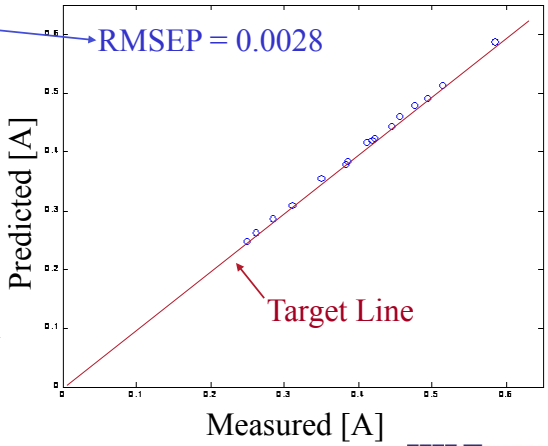
Predicted vs. Measured [A] for Test Set

Root Mean Square Error of Prediction



Predicted because Model has never seen these samples

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Compare RMSEC with RMSEP

Component	RMSEC	RMSEP
A	0.0026	0.0028
B	0.0033	0.0027
C	0.0028	0.0031

For any given component, we want RMSEP ~ RMSEC

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Summary of CLS

Advantages

- Simultaneously determine the concentrations of many analytes in a mixture.
- Multi-channel Advantage.
- Do not need pure component spectra.
- Provides pure component spectra in the sample environment (matrix).

Disadvantages

- Need the concentration of all chromophores in each sample of the learning set.*
- Very sensitive to non-linearity.

*Not necessarily true with more advanced forms of CLS

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Outline

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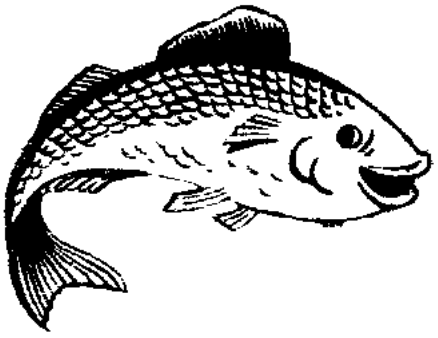
NIR of a Fish



We do not want to know the concentration of all the components in a fish.

What all is in a fish?

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Small Change to the Beer-Lambert Law

Multiple Linear Regression - MLR:

$$[\text{Analyte}] = A_{60}\, b + \text{Error}$$

Dependent Variable

Independent Variable

Regression Coefficient

Error in measuring concentration

Notice the thing we ultimately want is on the left side of the equal sign.

Multiple Linear Regression (MLR) is Sometimes Referred to as Inverse Least Squares (ILS)

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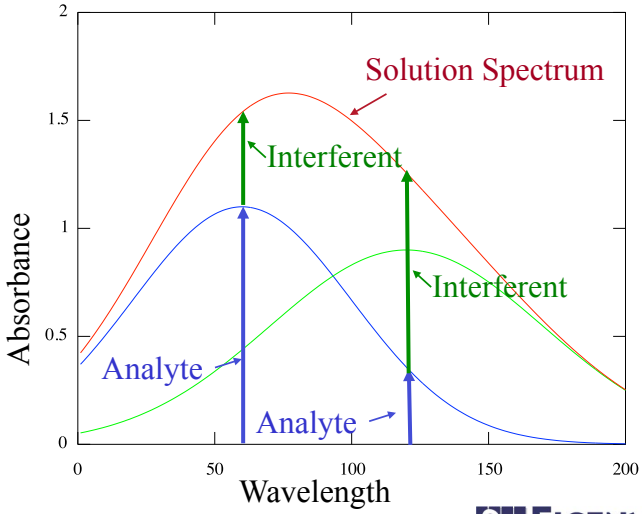
Let's Look at MLR Applied to Our



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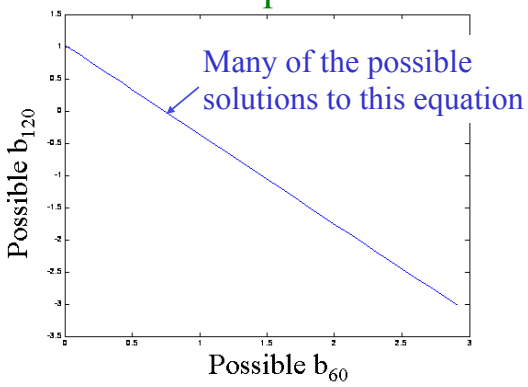
$$[\text{Analyte}] = A_{60}\, b_{60} + A_{120}\, b_{120}$$

One Equation - Two Unknowns
No Unique Solution



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What to Do?

$$[\text{Analyte}]_{I} = A_{60,I}\, b_{60} + A_{120,I}\, b_{120}$$

Make up a 2nd Solution using different concentrations:

$$[\text{Analyte}]_{II} = A_{60,II}\, b_{60} + A_{120,II}\, b_{120}$$

Notice the other interfering chromophores are not in the equation.

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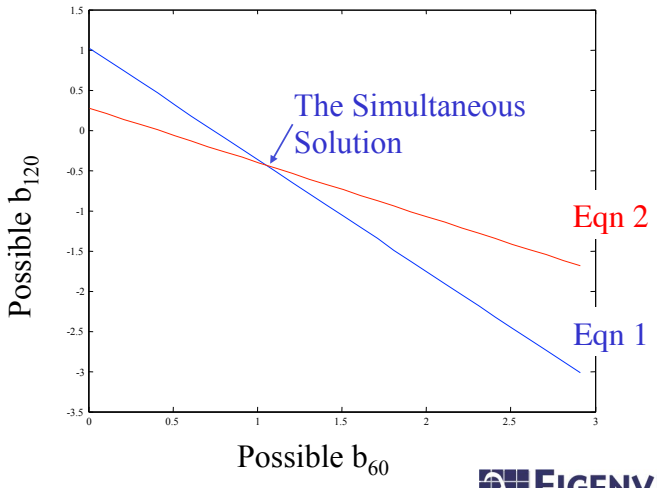
Two Equations - Two Unknowns



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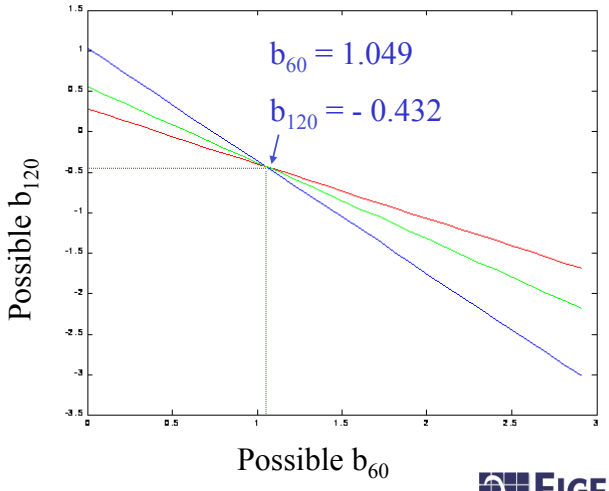
Add a Third Experiment
Overdetermined Solution



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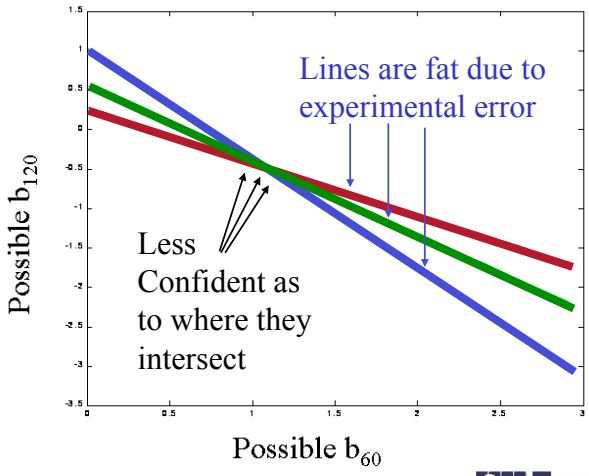
Problem Solved? Not Exactly!



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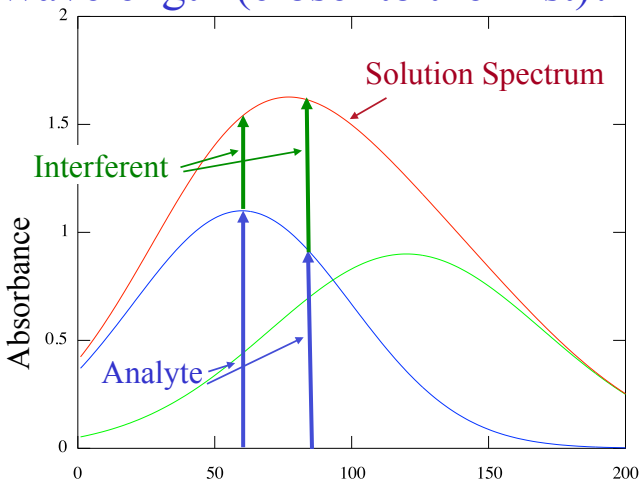
What if We Choose a Different 2nd Wavelength (closer to the first)?



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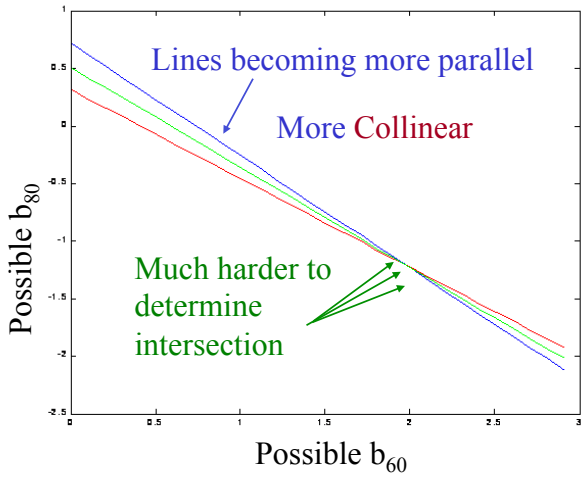
Using Wavelength 60 & 80



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Summary - MLR

Advantages

- Do not need to know the concentration of all chromophores in each sample of the learning set.
- Do not need the pure component spectra.

Disadvantages

- Best noise removal with many more samples than variables.
  - usually variables are cheap but samples are expensive
- Must use carefully chosen independent variables to avoid collinearity.
  - Lose multi-channel advantage
  - Often used just half dozen wavelengths
- Do not get pure component spectra

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Can't I have my cake and eat it too?

- *i.e.* is there a way to do MLR and keep more variables?
  - retain the multi-channel advantage
- How to handle the collinearity?
- Maybe there is a way to use PCA?

Outline

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Problem with MLR

- How do we keep all the variables as in CLS and only have to model the analyte as in MLR?
  - *i.e.* want a full spectrum technique, but don't know the concentration of all the chromophores
- Problem with MLR is: Collinear Variables
  - Also known as: Correlated, Redundant, Parallel
- Opposite of Collinear is Orthogonal
- How do we get Orthogonal Variables?
  - Also known as: Uncorrelated, Independent, Perpendicular

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PCA

- Recall that PCA converted real variables into new orthogonal variables: Principal Components
- Loadings for each PC are orthogonal to the loadings from other PCs
  - Loadings ←→ Variables
- Scores are the positions of the samples in the new PC space.
  - Scores ←→ Samples
- Perhaps MLR on the PCA scores?

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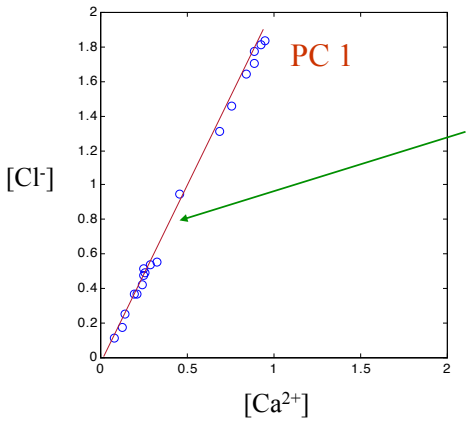
Recall the Solutions Analyzed for Ca^{2+} and Cl^-?



PC 1 was interpreted as related to the $[CaCl_2]$

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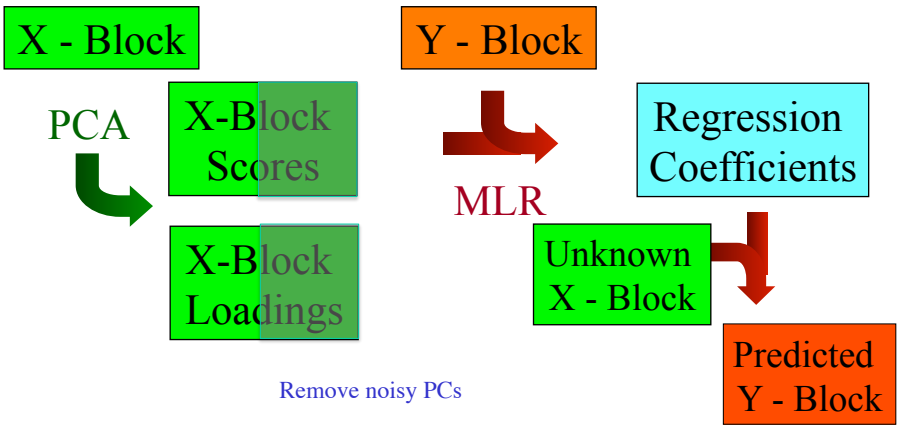
Principal Components Regression

PCR



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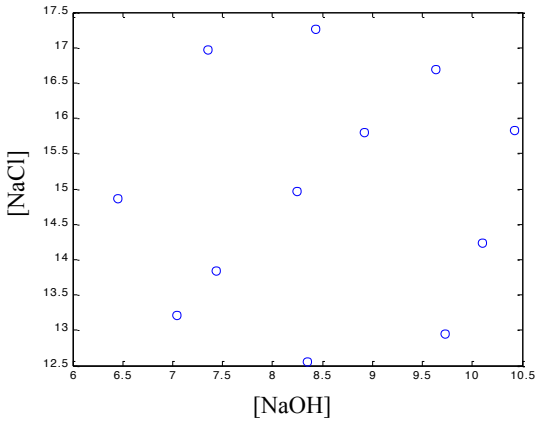
Want to determine the concentration of NaOH in aqueous caustic brine solutions using SW-NIR

Have 12 Solutions of NaOH and NaCl in Water



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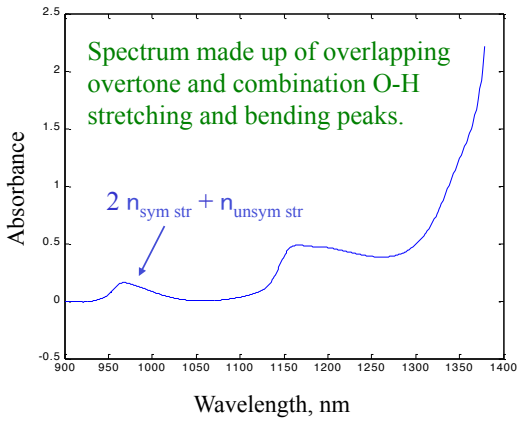
Typical SW-NIR Spectrum of Caustic Brine Solution



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Peak Shift

- The NIR water peaks shift with changes in
  - NaCl and NaOH, and
  - temperature
- Since the temperature will vary in the application, temperature variation must be included in the Learning Set
  - although temperature need not be known to calibrate for NaOH, it must vary in the Learning Set for the model to be robust to changes in temperature.
- Interferents (e.g. NaCl & temperature) must either be kept constant forever, or varied in the learning set.

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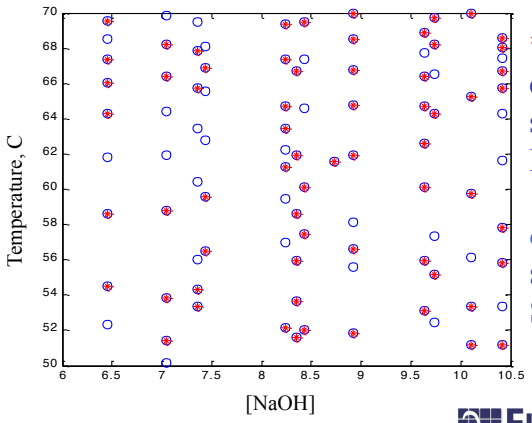
Data: 95 Spectra SW-NIR of 12 Caustic Brine Solutions from 50-70° C



* Randomly choose 65 samples as Learning Set

o Remaining 30 samples are Test Set

Seasholtz MB., *Chemom. Intell.Lab. Syst.* 1999; **45**: 55–63.

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PCR on Learning Set

- Should we mean center or autoscale?
  - Usually just mean center spectra
  - recall that this means that we're allowing for an intercept other than zero
- How many PCs to keep in the model?
  - RMSEC vs #PC's
  - RMCECV vs #PC's Cross-validation

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Cross Validation

- Works just like it did for PCA
- Except we have two types of errors (Residuals):
  - X-block Error – same as for PCA
  - Y-block Error = $Y_{pred} - Y_{meas}$

Used to calculate RMSECV

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Cross-Validation Rules of Thumb

- Only use Leave-One-Out CV for fewer than 16 samples.
- Best results are obtained if you take out Square Root of Number of Samples at a time.
- "Genuine Replicates" can be split between the Learning and Test Sets
  - "'genuine replicates' are repetitions which are subject to ALL the sources of error that affect runs made at different experimental conditions"*
- If simple repeat measurements, keep them together, *i.e.* have all in either the Learning Set or Test Set.

*Box, Hunter, and Hunter, "Statistics for Experimenters", Wiley (1978)

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Cross-Validation for PCR Example

- Venetian Blind
- 8 Splits (approx. ~65 samples)
- Calculate RMSECV for up to 20 PCs
  - expect the number to be <20

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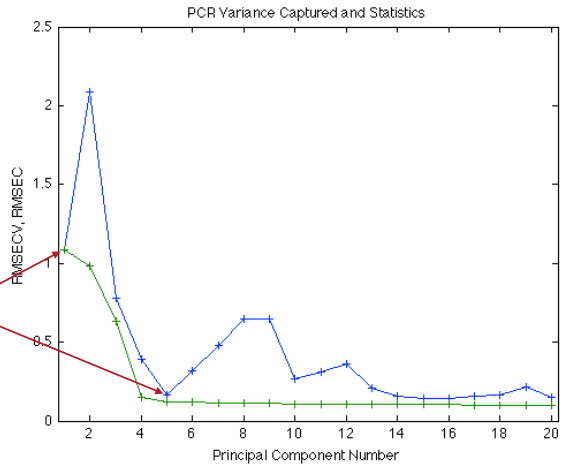
PRESS Plot

Double Minimum often suggests Outliers



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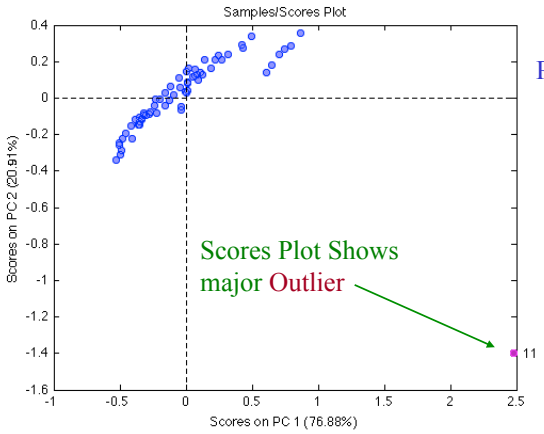
Keep 5 PCs
just to see what is going on



PC 1+2 ~97.79%
of the variance
in the X-block

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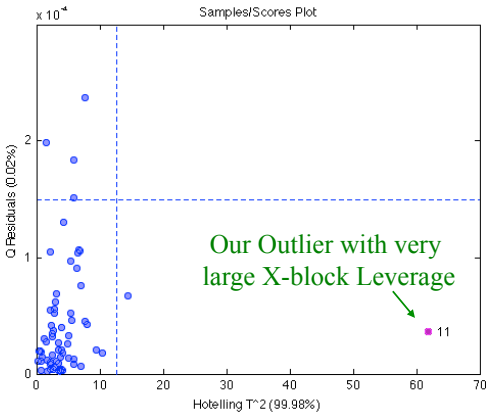
Outliers Can Also be Found by Examining the X-Block Influence Plot

X-block Residual
- How much each
spectrum differs
from the PCA
Model (retained
PCs) of spectra.



It appears to be
within the model,
because it pulls
the model to
itself

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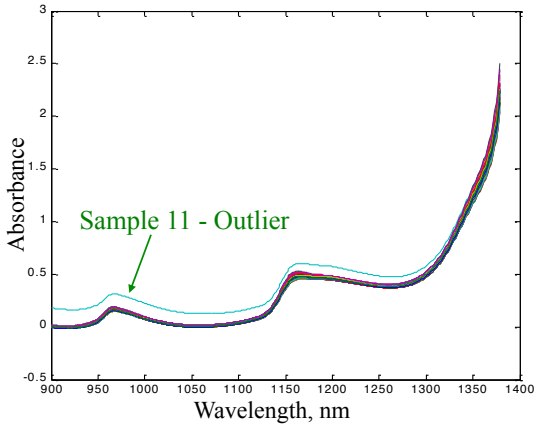
Spectra of Learning Set



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Important Lessons

- Always plot spectra to see if there are any obvious outliers.
  - Plot your data
- Do PCA on your X-Block and examine the scores, Q, and T^2 for Outliers before building calibration model
  - Exploratory data analysis

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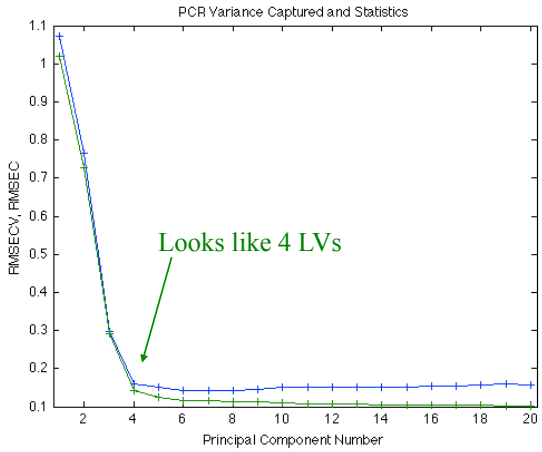
Delete Outlier Sample 11, Rebuild Model and Examine PRESS Plot



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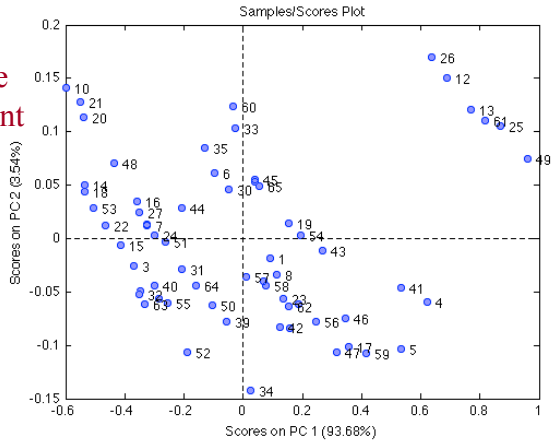
Scores Plot

additional outliers are not apparent



Identical to PCA Scores Plot

Lots of Structure to examine when we are done finding outliers.

PC 1+2 ~97.22%

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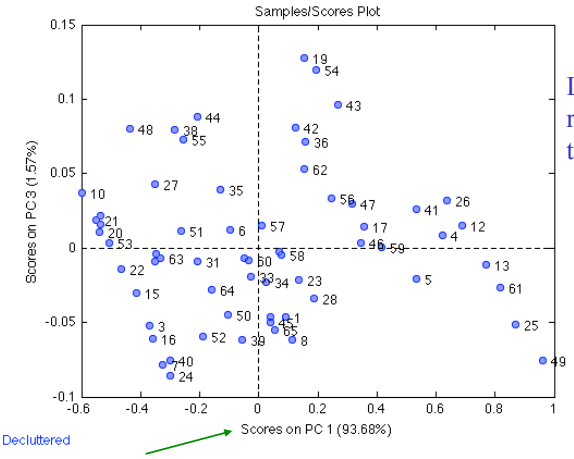
Examine All the PC Scores



Look for samples removed from the rest.

PC 1+3
~95.25%

Keep PC 1 as the x-axis

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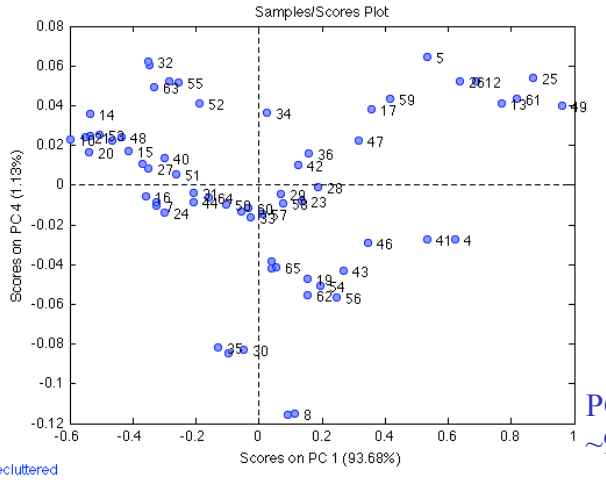
Scores PC 4 vs PC 1



PC 1+4
~94.81%

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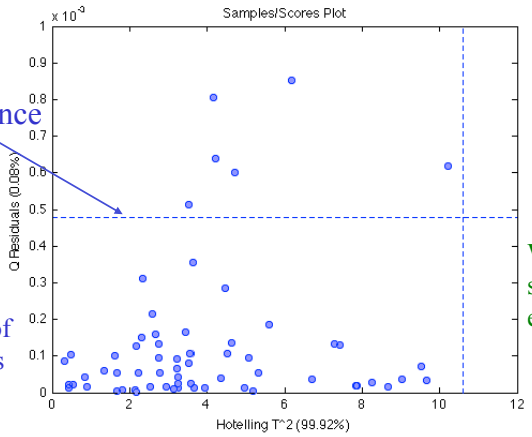
X-Block Influence Plot

95% Confidence Level



We have no samples with excessive T^2

At 95% CL we would expect 1 of every 20 samples (about 3) above line.

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Looking for X-block Outliers

- Look for isolated sample(s) in Scores Plots.
- Look for samples with excessive Q Residual or Hotelling's T^2.
- An Outlier is not necessarily a bad sample, just unique. Examine it:
  - If good, add more like it to Learning Set.
  - If bad, fix it or remove it.

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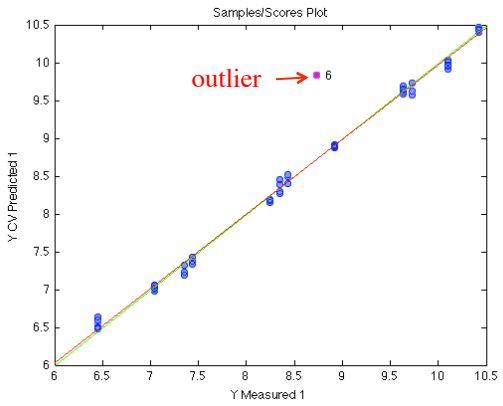
How Did We Do?
Estimated [NaOH] vs. Measured [NaOH]



This plot is for the Learning Set, but since we are doing CV, it is a "Predicted" Y values.

Deviations from the diagonal are used to calculate RMSECV.

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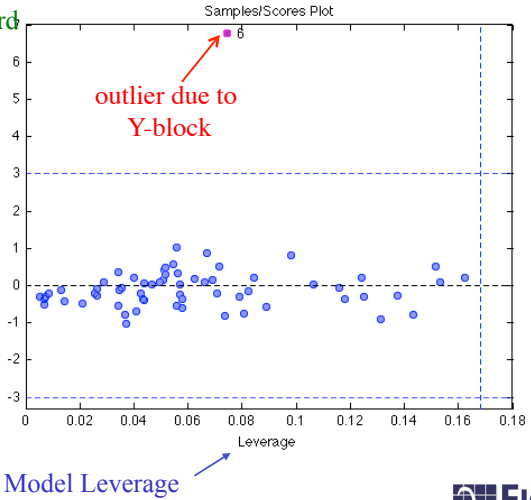
Model or Y-Block Influence Plot

Residual expressed in units of Standard Deviation



Y Residual

Large Leverage ⟶ implies error in **X**-block

Model Leverage

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Looking for Outliers

- Samples with excessive Y-block Residuals of Estimation
  - Look for problem in **Y**-block
- Samples with excessive X-block Leverage or Q (X-Block) Residual
  - Look for problem in **X**-block
- Remove or fix samples with problems.
- If sample OK, then add more like it.

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Remove or Fix Sample #6

- It turns out that there was an "entry error" for the "known" [NaOH] for Sample #6
  - The measured was 9.7325
  - Entered in the data was 8.7325
- Once corrected, we can make another model
- Removing or correcting outliers and rebuilding the model often reveals new outliers not seen before.
- Check for outlier again using Influence & Scores Plots
- Modeling tends to be iterative
  - Use what is learned at each step to help in subsequent models

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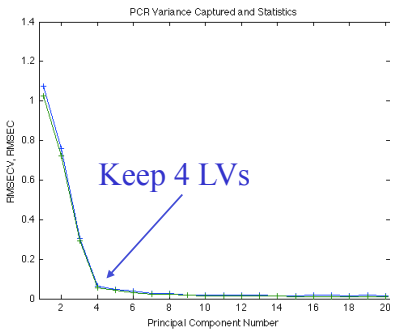
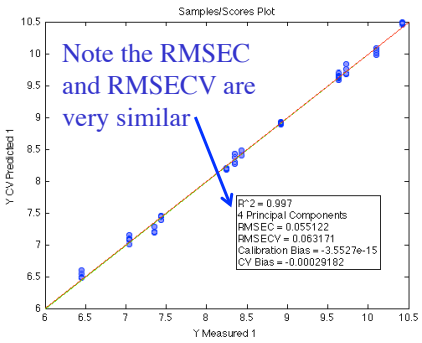
PRESS and Prediction Plot Plots





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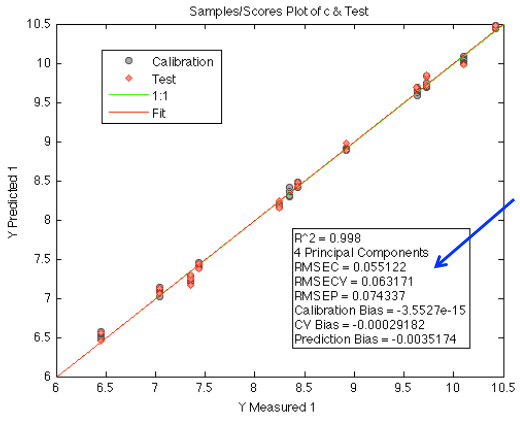
Apply Model to Test Set



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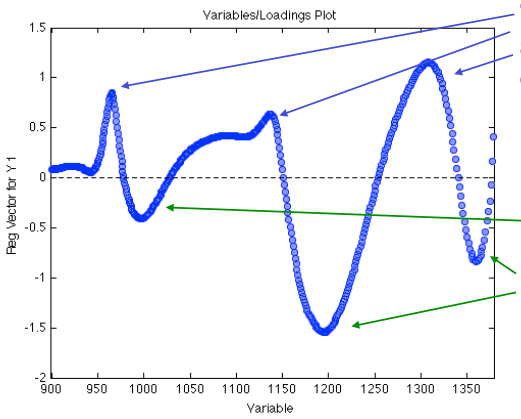
This Is the Model Regression Coefficients

Danger:

Be careful interpreting Regression Coefficients from PCR (and PLS). They are ***not*** pure component spectra.



These variables are positively correlated to [NaOH]

These variables are negatively correlated to [NaOH], correcting for an overlapping interferent

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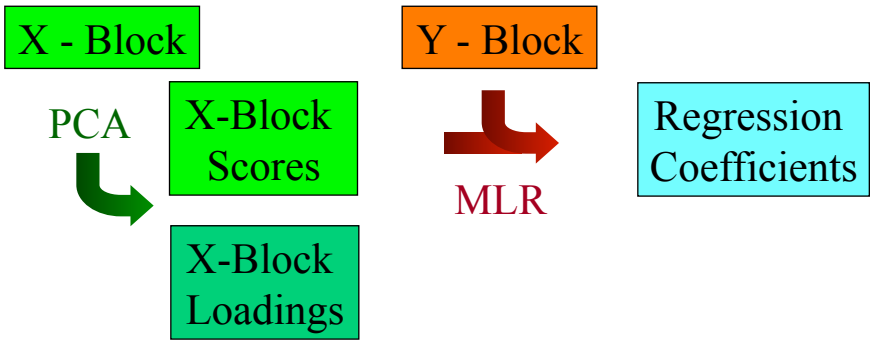
Recall How PCR Works



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Problem with PCR

- The PCs were created from the **X**-Block without any help from the **Y**-Block
  - and sorted in order of variance captured
- The PCs useful for predicting the **Y**-Block may be deep down in the pile, … with all the noise
- Shouldn't we bring in the **Y**-Block earlier in the process, at least as a consultant on the construction of the Principal Components ←→Latent Variables?

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Outline

- Regression Motivation & Rational
- Classical Least Squares (CLS)
- Multiple Linear Regression (MLR)
- Principal Components Regression (PCR)
- Partial Least Squares Regression (PLS)
  - PLS-1
  - PLS-2
- Summary

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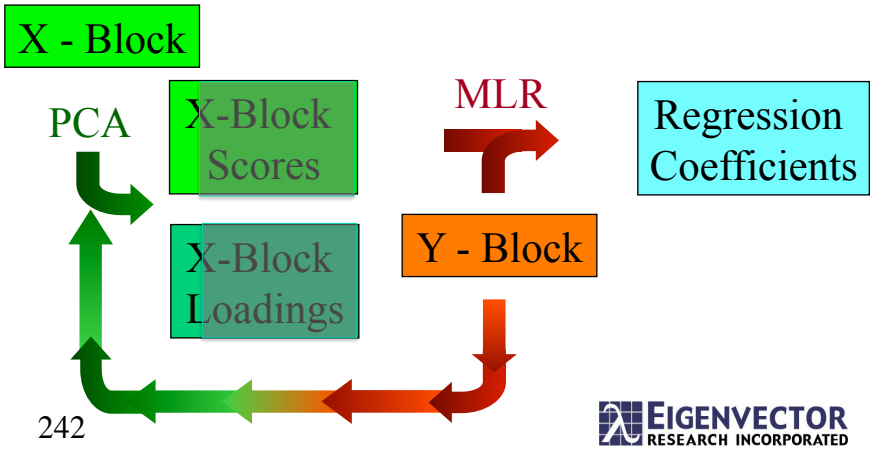
Modification to PCR
Partial Least Squares, PLS-1



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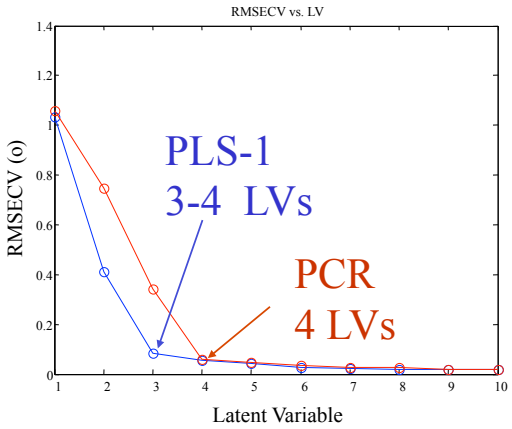
Compare PRESS Plots



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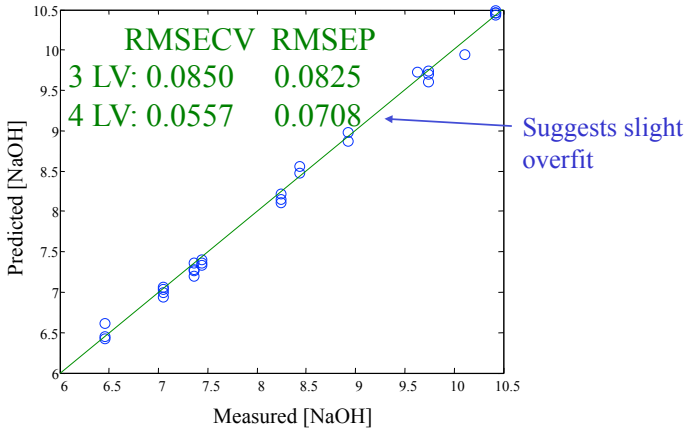
PLS-1 Prediction of Test Set with 3 LVs



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PLS-1 Compared to PCR

- The hope is that PLS-1 will require fewer latent variables than PCR requires principal components which suggests that there will be less noise
- The two methods often provide very similar results
- PLS usually brings more useful information into the earlier LVs for easier interpretation

Our example:	RMSECV	RMSEP
PLS-1 (3 LV)	0.0850	0.0825
PLS-1 (4 LV)	0.0557	0.0708
PCR (4 PC)	0.0592	0.0743

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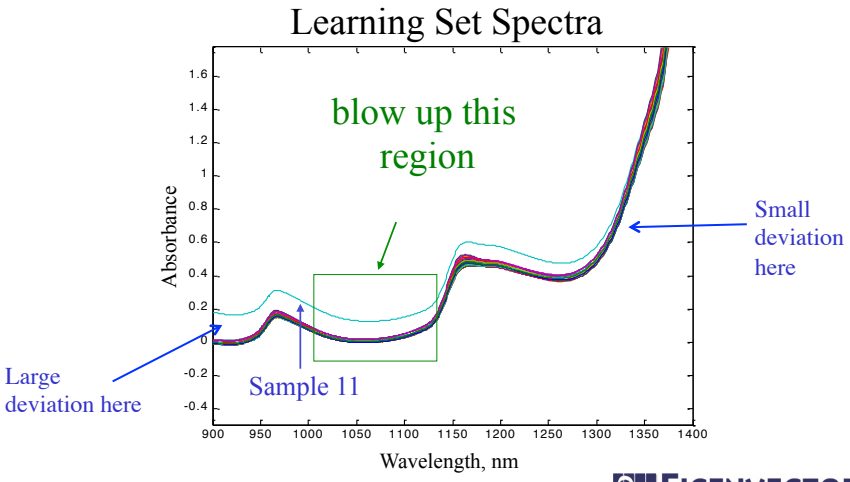
Baseline Ramp?



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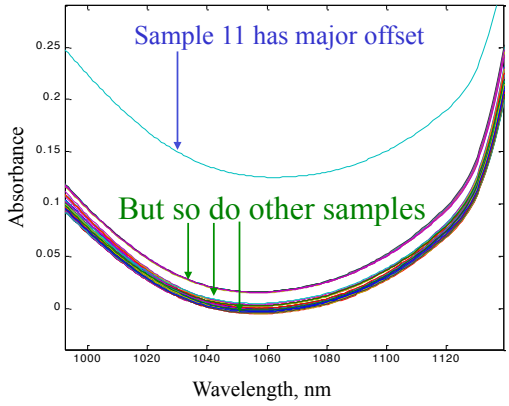
Yes



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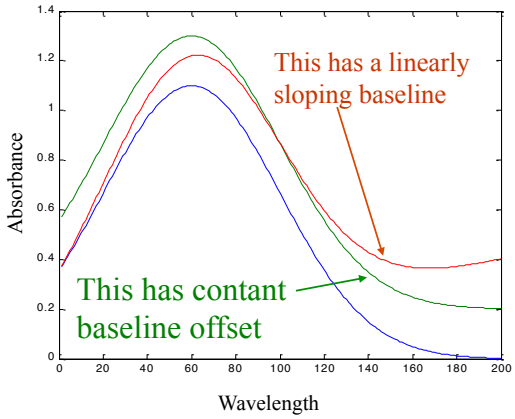
Baseline Problems Example

Here are three identical spectra, except:



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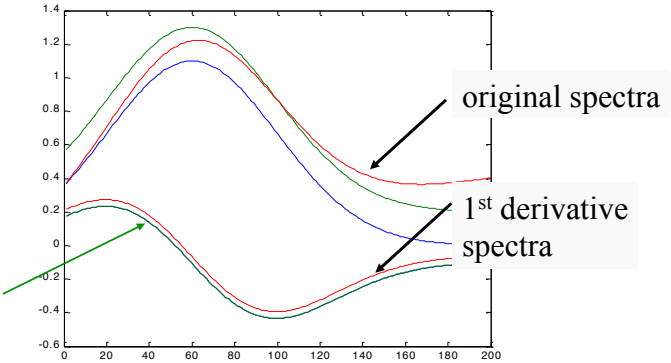
Using Derivative Spectra

Take 1st derivative of the Three Spectra



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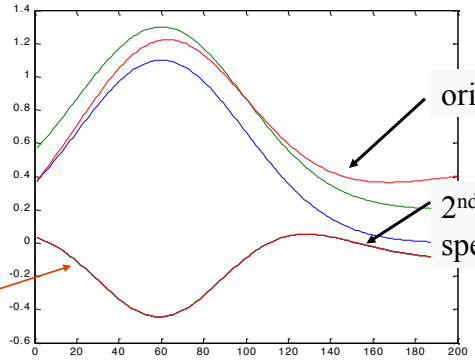
Using Derivative Spectra

Take 2nd derivative of the Three Spectra



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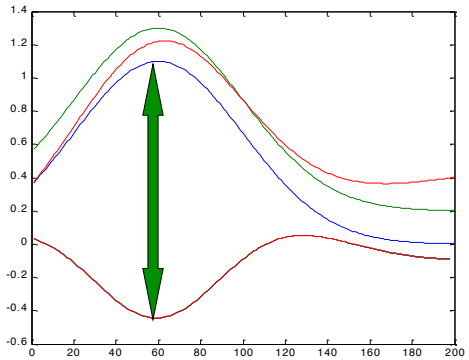
Comments on 2nd Derivative

2nd derivative spectra look different



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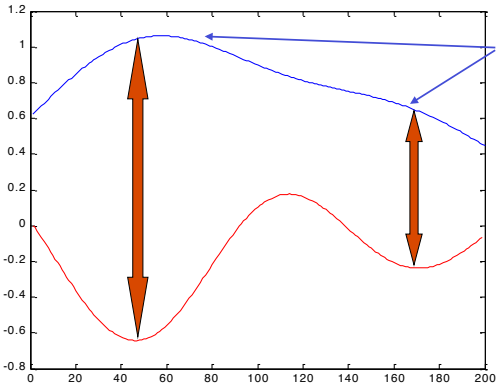
Comments on 2nd Derivative

2nd derivative increase resolution



Peak with possible shoulder

2nd derivative resolves the peak and shoulder

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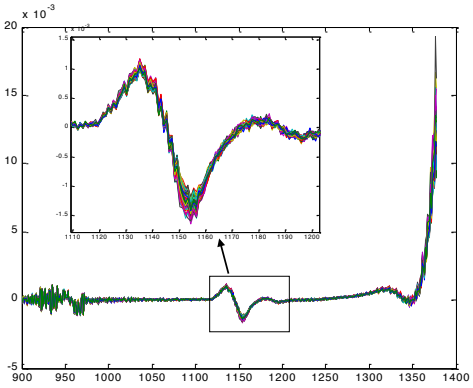
2nd Derivative of Learning Set Spectra



Derivatives can also increase the effect of Noise!

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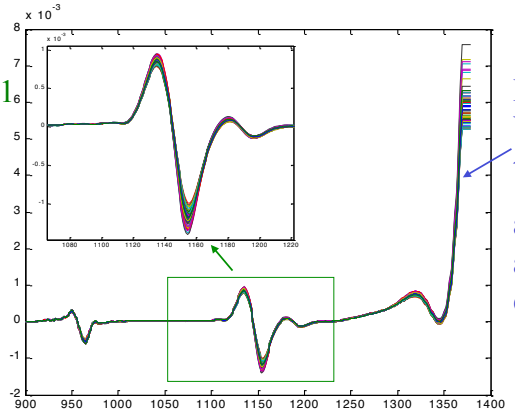
Filter the Data Before Taking 2nd Derivative: Savitzky-Golay

Even Sample 11 looks good!



Must remove Variables in this steep area

and be careful about "end effects"

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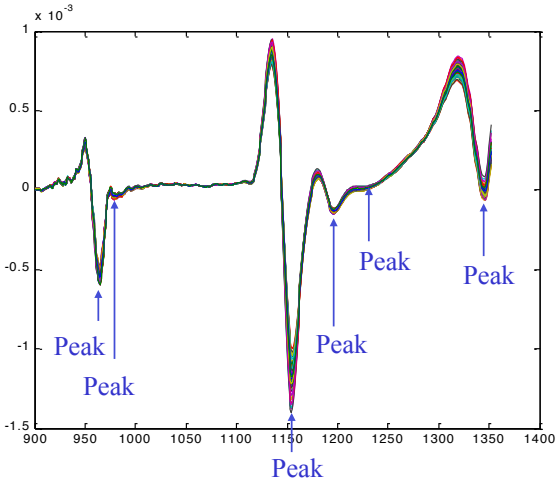
Notice the Resolution



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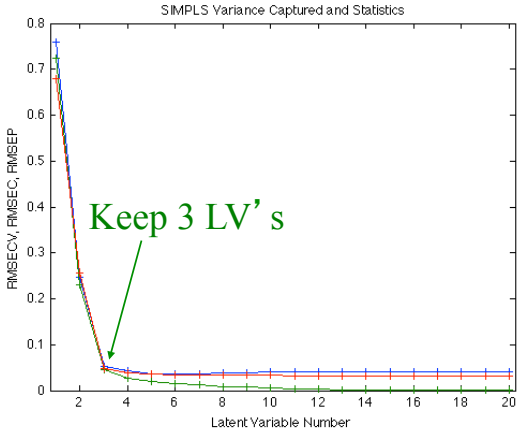
PLS-1 of 2nd Derivative Spectra for the Learning Set



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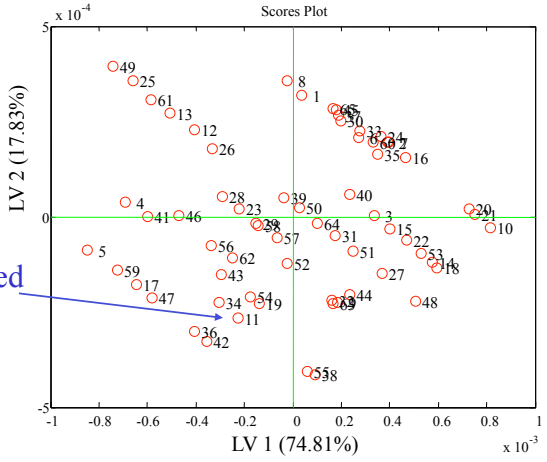
Scores Plot

Sample 11 has returned to the fold



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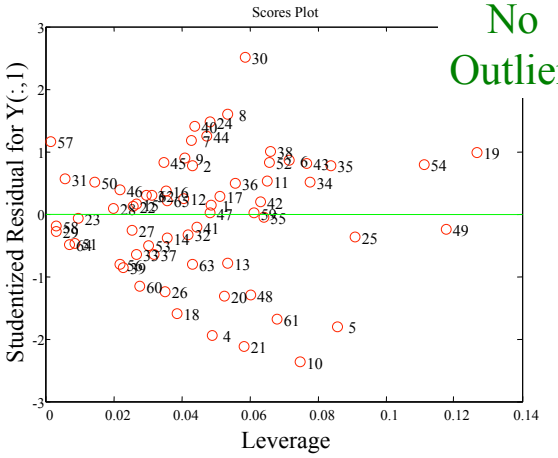
Model Influence Plot



No Outliers

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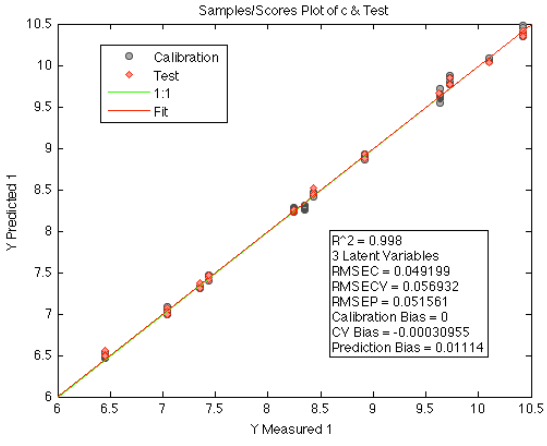
Prediction of [NaOH]



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Comparison of Regression Methods and Pretreatments

	RMSECV	RMSEP	
PLS-1 2nd Derivative (3 LV)	0.0569	0.0516	Including sample 11
PLS-1 (3 LV)	0.0850	0.0825	
PLS-1 (4 LV)	0.0557	0.0708	
PCR-1 (4 PC)	0.0592	0.0743	

Need to ask: is the difference statistically significant with respect to error in the reference method.

Note: It is possible to obtain a RMSEP somewhat smaller than that from the reference method.

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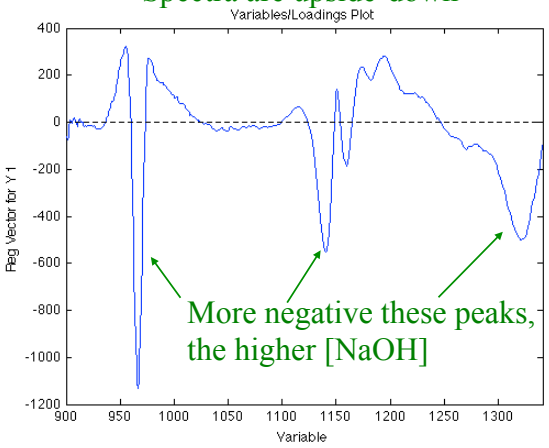
Regression Coefficients

Remember 2nd Derivative
Spectra are upside-down



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PLS-2

- PLS-2 is just like PLS-1 except the **Y**-Block has more than one variable (multivariate Y).
- Note that MLR and PCR can also be performed for multivariate Y, however since the **Y**-block is not used to identify the PCs it is the same as multiple univariate Y models.
- PLS uses the **Y**-block, therefore PLS-2 can provide different results than multiple PLS-1 (univariate Y) models.
- An important use of PLS-2 is PLS-DA, a classification technique

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We Wish to Identify Geographic Origin of Romano-British Pottery

We have samples of pottery from:

Gloucester	22
Wales	16
New Forest	10

X-Block: 8 metal concentrations as determined by Atomic Absorption Spectroscopy:

Al Ba Ca Fe K Mg Na Ti

A. Tubb, et. al., *Archacometry*, **22**, 153 (1980)

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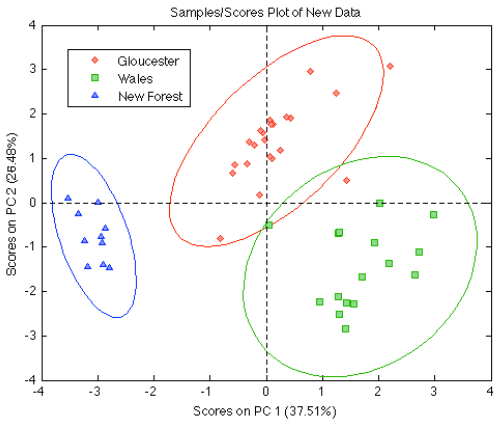
PCA (autoscaled) Scores Plot for **X**-Block of All 48 Samples

Not very good discrimination.



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PLS-DA
Y-Block coding of Class Membership

Sample	Gloucester	Wales	New Forest
1	1	0	0
2	0	1	0
3	0	0	1

Not Glouchester

Not Wales

Yes New Forest

Create and assign classes to samples and software will code samples

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Split Samples into a Learning Set (32 samples) and Test Set (16 samples)

Learning Set:	Gloucester	15
	Wales	11
	New Forest	10
Test Set:	Gloucester	7
	Wales	5
	New Forest	3

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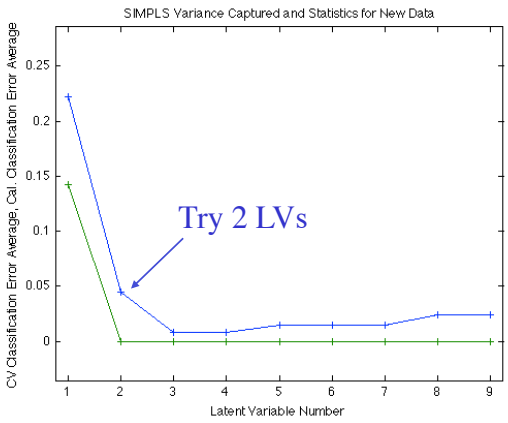
RMSECV (5 Split Venetian Blind) vs. Number of Latent Variables



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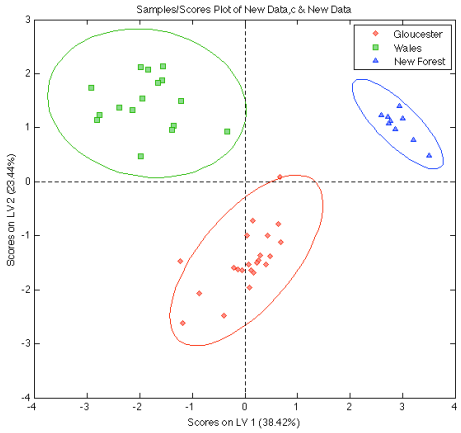
PLS-DA Scores PV2 vs LV1

Bringing in the Y-block during the creation of the LVs improved class discrimination



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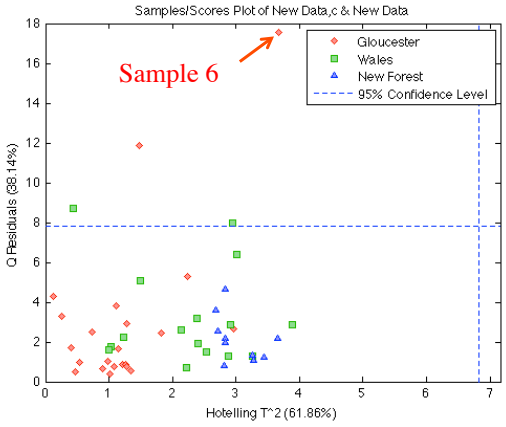
Always Check X-block Influence Plot



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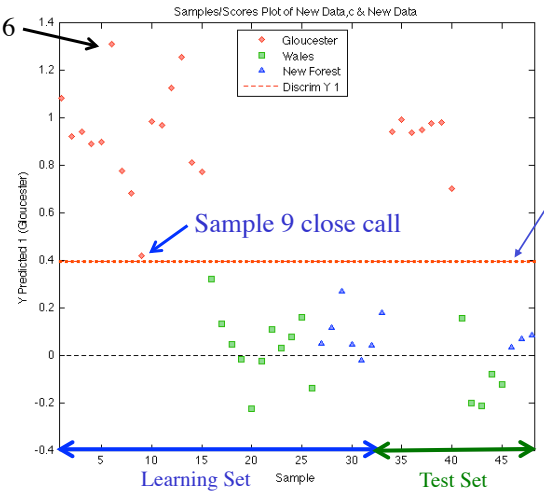
Predictions for Class Gloucester



Sample 6

No false positives or negatives

Sample 9 close call

Decision Line

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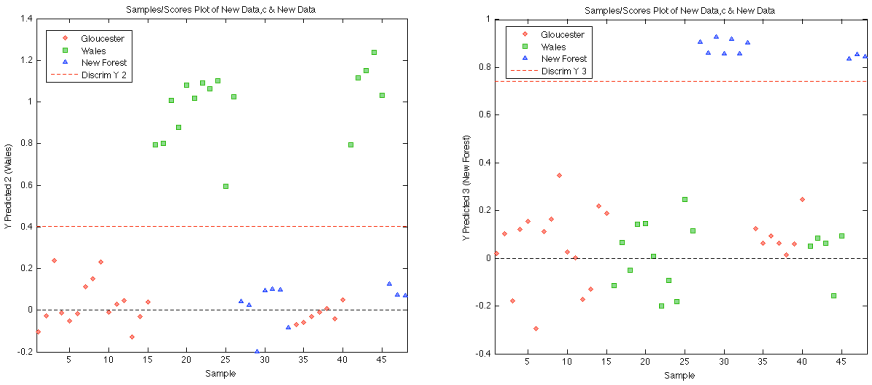
Predictions for Class Wales



No false positives or negatives

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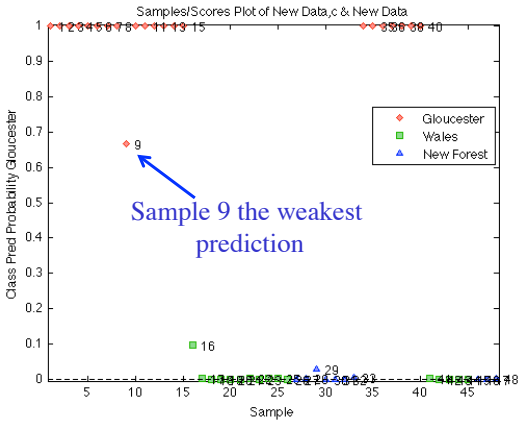
Class Prediction Probability for Gloucester



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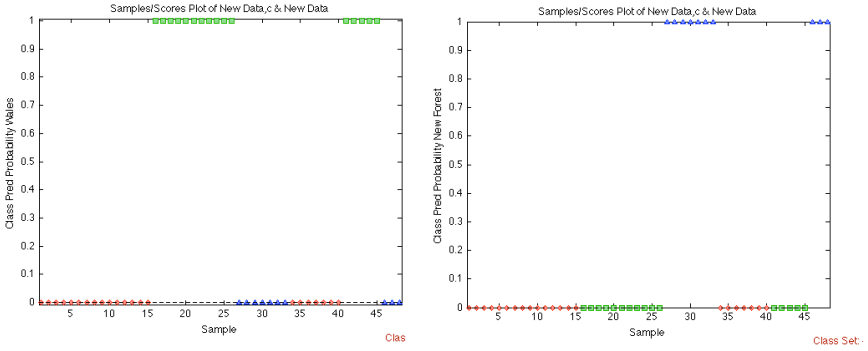
Class Prediction Probability for Wales and New Forest



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Dangers in using PLS-DA

Create a Learning Set X-Block of 100 samples with 100 random variables

Assign samples to two classes at random – Y-Block

Create a similar Test Set of 100 samples

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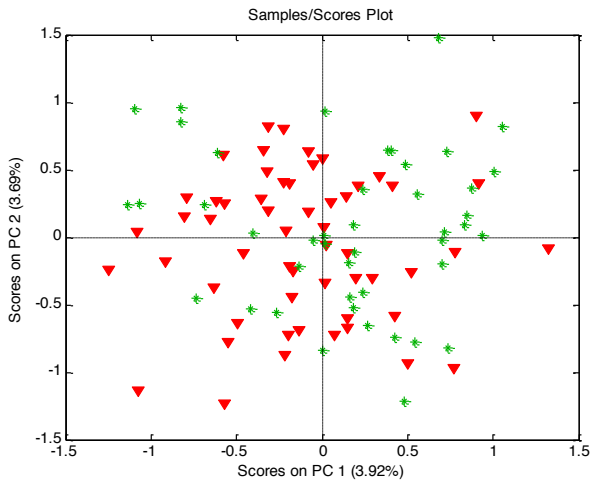
PCA Scores Plot of Learning Set



Totally Random distribution of samples and classes

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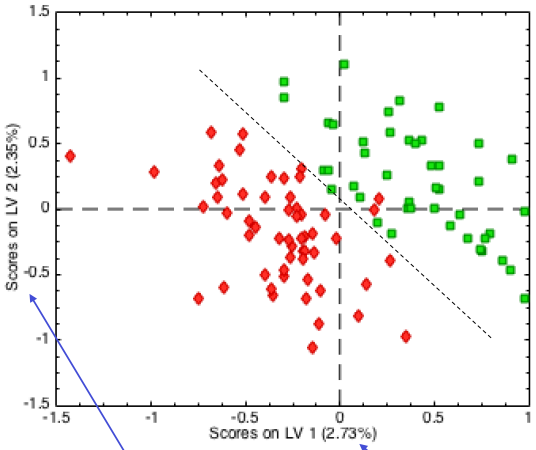
PLS-DA Scores Plot of Learning Set



PLSDA has found differences between the two classes in the Learning Set

Notice small amount of variance captured.

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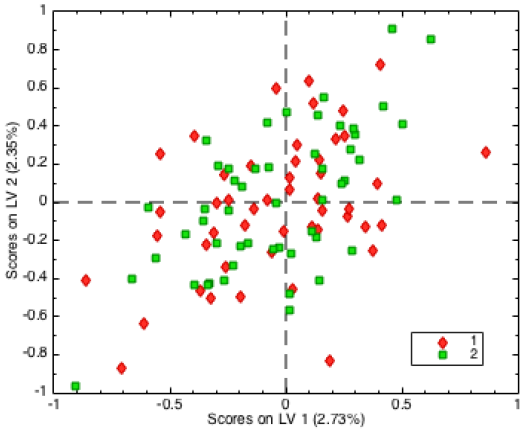
The Test Set Projected onto the Model Scores Space



The PLSDA Model fails to distinguish the samples in the Test Set

This is important. Validation can indicate performance problems – do not kid yourself!

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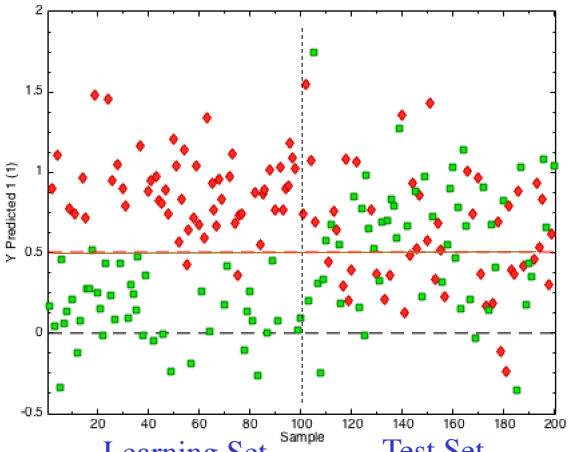
Classification of Learning Set and Test Set



Learning Set Test Set

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Always Use an Independent Test Set to Validate Your Model(s)!!!

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Even the Most Expensive Cars Require Regular Maintenance

- Periodically Revalidate Model to make sure conditions have not changed over time
- Update Model if necessary
  - many new tools to assist in this process.

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Also Remember

Rome Was Not Built in a Day

It took days, months or even years to take the data.

It may take days or weeks to build a proper model. Initial models may indicate that new measurements are necessary.

Be Patient and Think about What You Are Doing

Math, Physics and Chemistry are your guides.

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LVC

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