Food Screener™: Innovative NMR-tools for Food Quality and Safety Screening

食品质量和安全性筛选的NMR创新解决方案

Bruker China
Xu Wenxin(徐雯欣), Ph.D
NMR Application Specialist

Would you like to learn more ? Contact a customer service representative to discuss about Wine Profiler, Honey Profiler, Juice Profiler.

Overview 概要

- What can Nuclear Magnetic Resonance (**NMR**) accomplish in food analysis?
  NMR为什么可以运用于食品筛选？
- NMR-based screening features
  基于NMR的食品筛选具有什么特征？
- What information can we obtain from Food screener?
  Food Screener可以为用户提供什么信息？

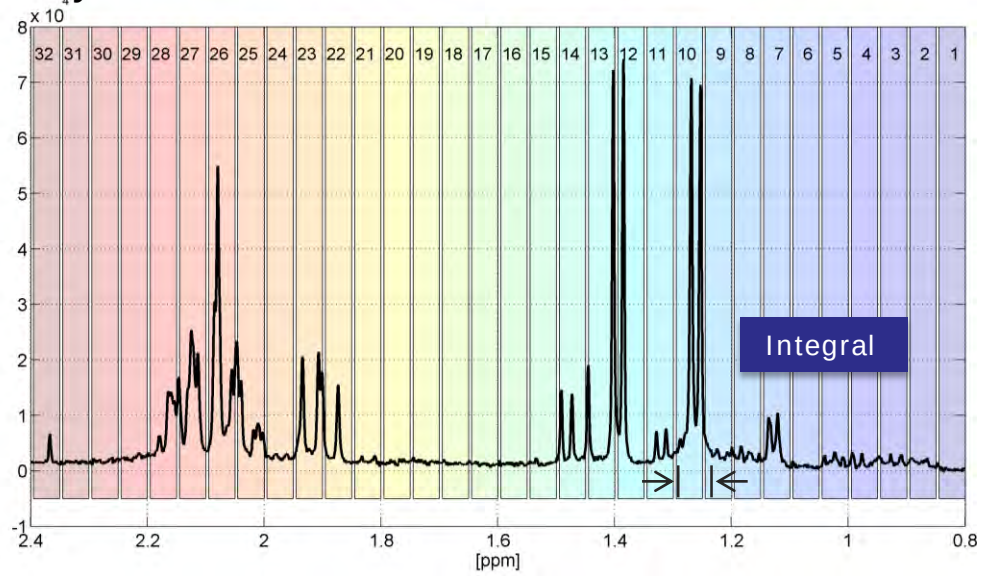
Why NMR is able to accomplish in food analysis

- Sample Materials: Liquid, Solid & Semi-Solid samples
- Unbiased detection

- Integral area of singal is proportional to its molar concentration (except active 1H)
- The combination of hundreds of detective molecules, with thousands of signals shows a highly specific `fingerprint´-- Phenotype.
- This `fingerprint´ is a clear multidimensional marker, which is able to make conclusions about origin, quality, classification, concentration, and so on.

1D 1H spectra

- Organic compound
- 1H sensitivity



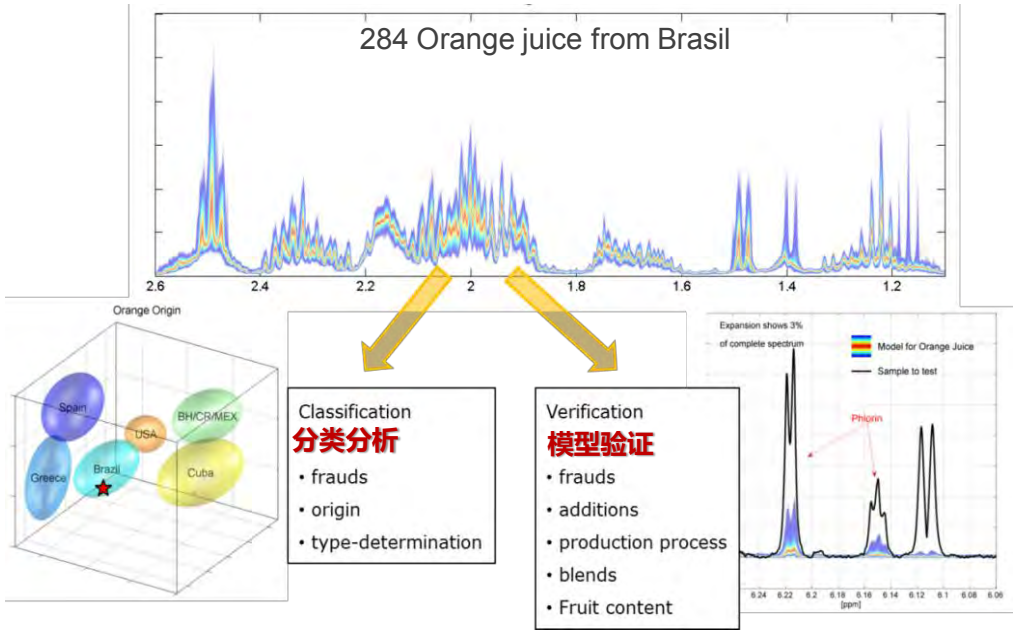
What can NMR accomplish in Food Analysis?

- All conventional food tests are targeted. What is not directly checked for, will most often be overlooked!

- Unbiased detection for ALL proton atoms in sample (**Holistic**)

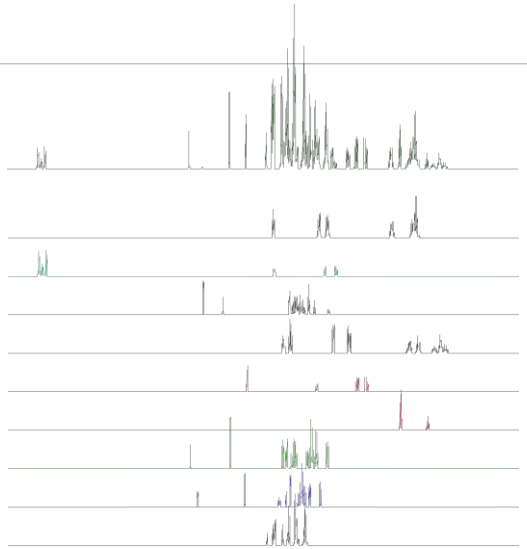
- Nontargeted Screening enables the detection and analysis of unexpected and unknown parameters, that targeted analysis methods can not detect.

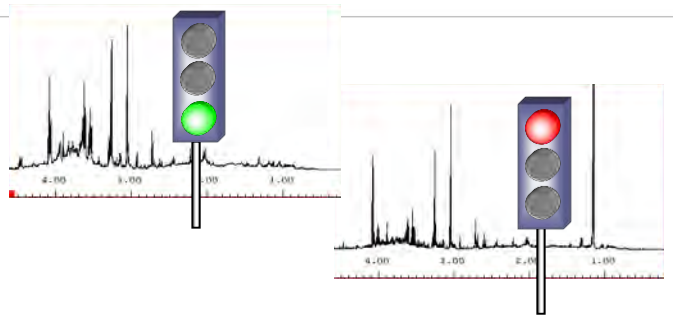
Multivariate statistics
多变量统计分析

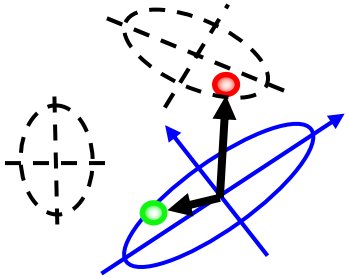


What can NMR accomplish in Food Analysis?

Targeted- / Nontargeted- screening within a set of experiments! (~15min)







Deconvolution and LC-SPE-NMR Targeted!

Metabolic profiling

Identification / quantification

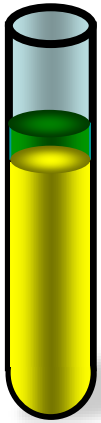
Statistics Untargeted!

Metabolic fingerprinting

Classification / discrimination

NMR-based Screening Features

Minimal sample preparation

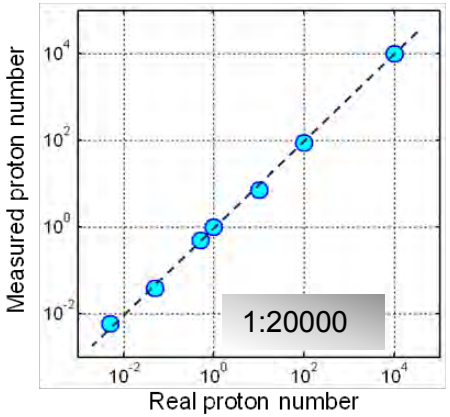


10% buffer addition
Might need centrifugation

Intact mixtures

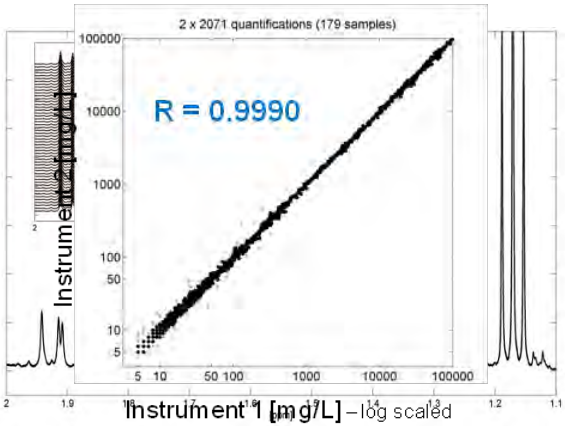
High throughput

Inherently quantitative linearity and huge dynamic range



Targeted analysis
Quantification of compounds

Highest Reproducibility and Transferability



Non-targeted /statistical analysis
Metabolic Fingerprinting
(Classification/ Verification)

FoodScreener: 400MHz Standard Platform for multiple Bruker Solutions

SGF-Profiling™ module

Wine-Profiling™ module

Honey-Profiling™ module

TBD
Module offered by external provider

BBIO
FoodScreener™ 400MHz platform
ISO 9001

Juice Screener™ / SGF-Profiling™

SGF: SURE-GLOBAL-FAIR

- Non-profit organization dedicated to Juice Safty & Quality Control
- Supported and financed by over 650 members from nearly 60 countries
- Inspectors all over the world taking samples on site
- No analysis in house
- Suspicious samples are sent to external laboratories

Fruit Juice Analysis

- Minimal sample preparation
- Only one measurement (~ 15 minutes)
- Targeted analysis (Quantification of >30 compounds)
- Non-Targeted analysis, up to 10 results (Classification and Verification)
  - Origin, Product Type, Untargeted Analysis, Fruit content, Mixture Dectection …
- Database of more than **16000** juices (Version 3.0)
- PDF report of all results

INDUSTRIE PREIS 2008

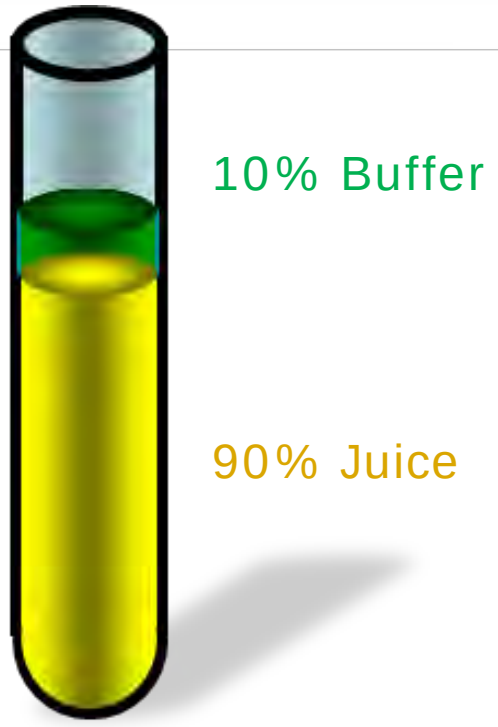
German Industry Award 2008
Category: Automation

Greentech Asia Shanghai 2010: Award for most innovative Food Analysis System

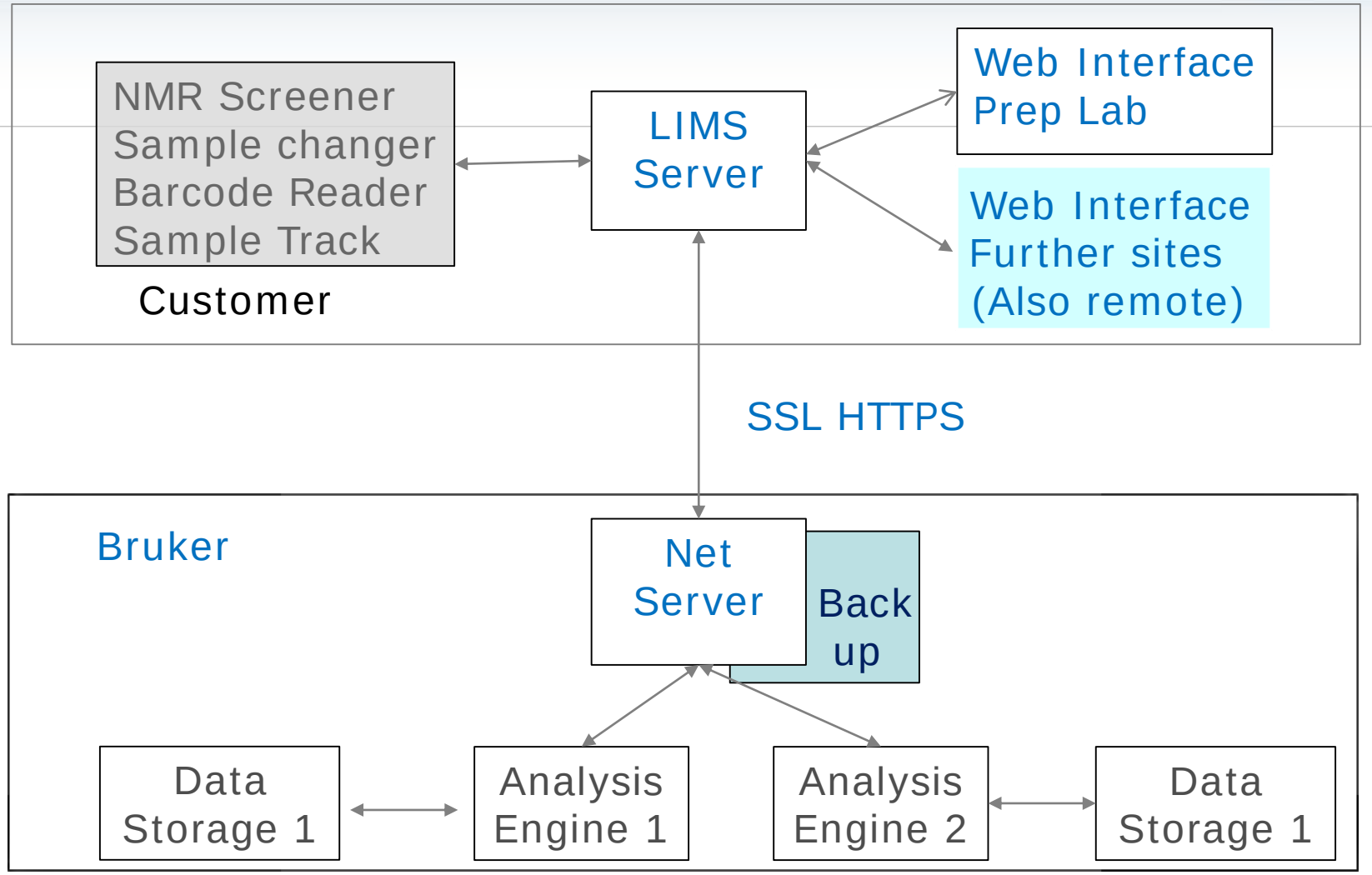
Minimal Sample Preparation

- Clear juice (e.g. apple juice):
  - Addition of 10 % buffer

    (KH_2PO_4, D_2O, NaN_3, TSP)
- Cloudy sample (e.g. orange juice)
  - Centrifugation 10 minutes(6000 rpm)
  - Addition of 10 % buffer

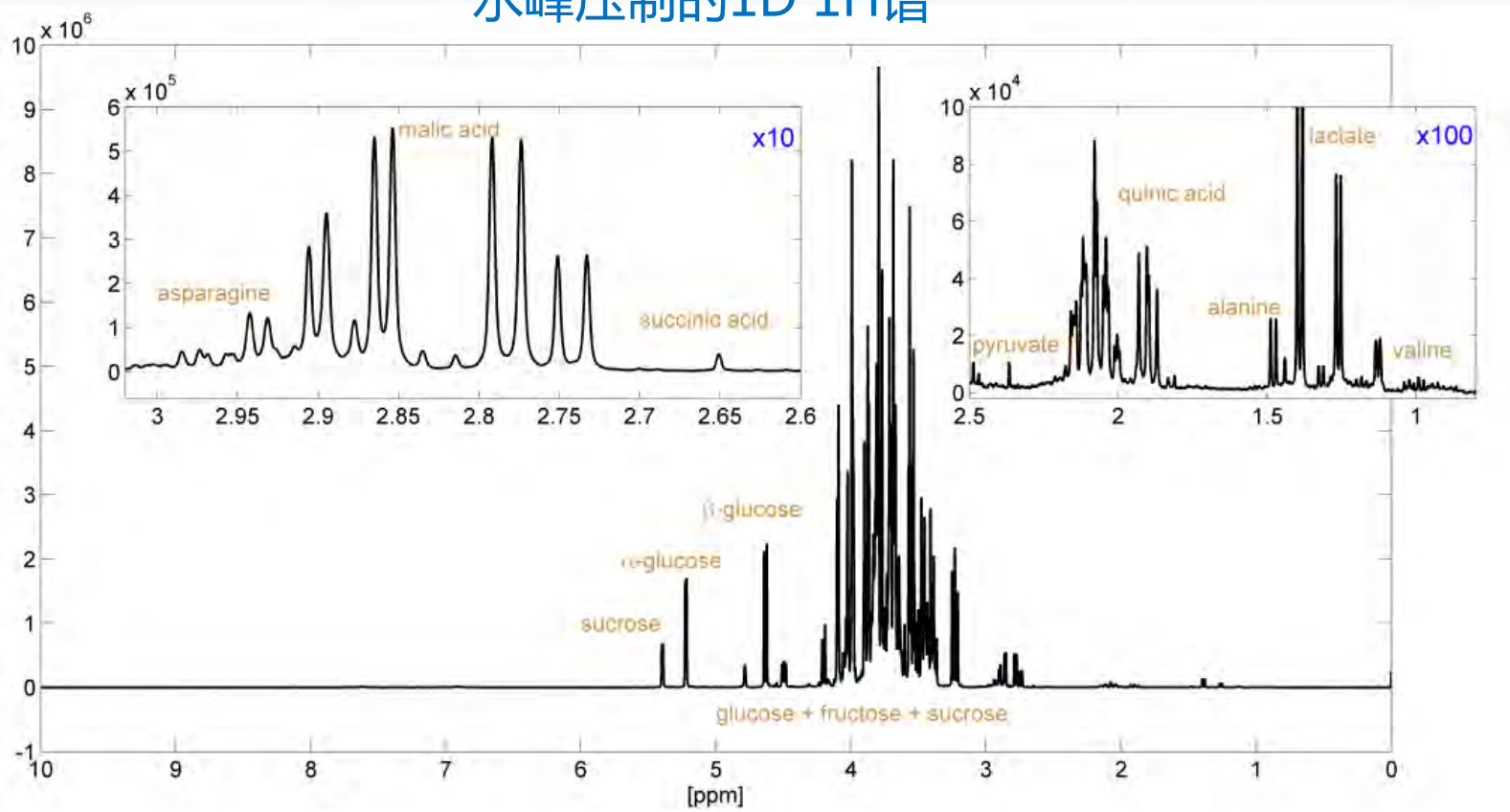


Procedure for secure data transfer
Food Screener



NMR–Spectrum of an Apple Juice

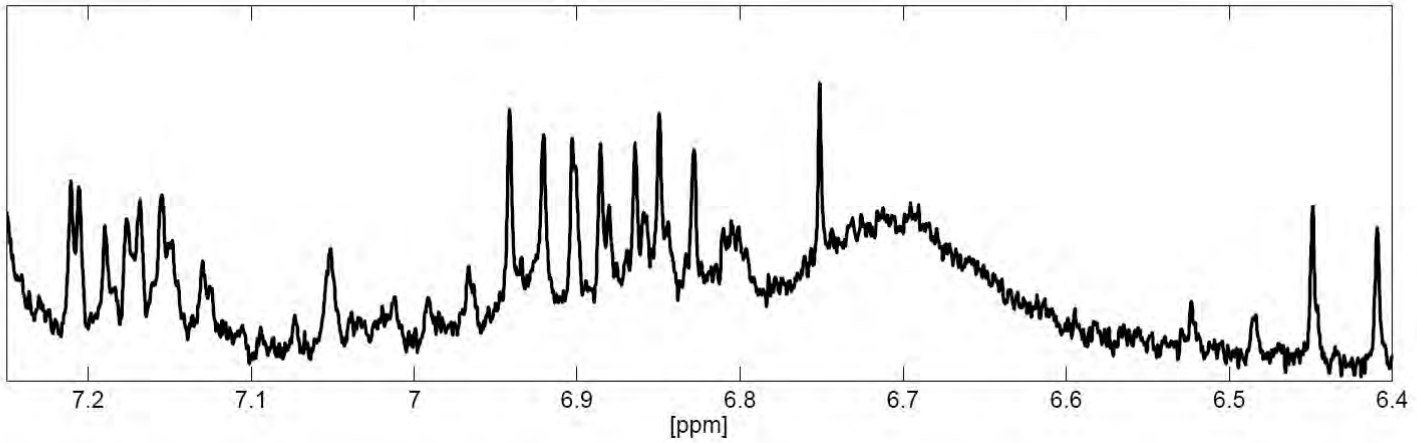
水峰压制的1D 1H谱



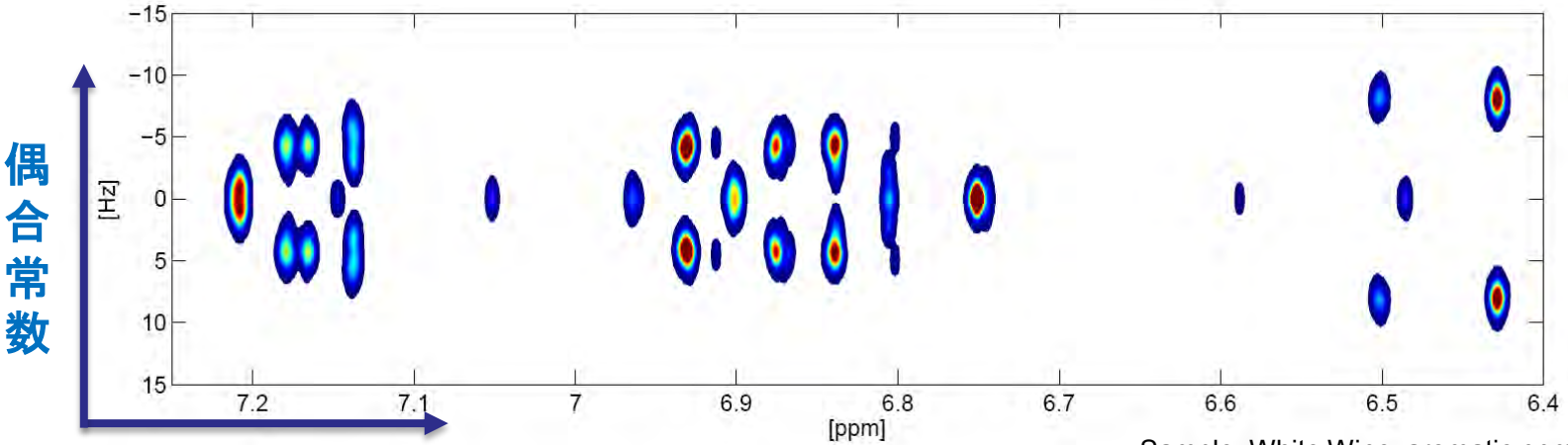
Dynamic range: **1:20,000** (lowest concentration to highest concentration)
With a throughput of 4 samples/hour
Very high reproducibility (spectrometer comprehensive)

Comparison 1D-NOESY and 2D-JRES (for safe identification)

1D 水峰压制氢谱

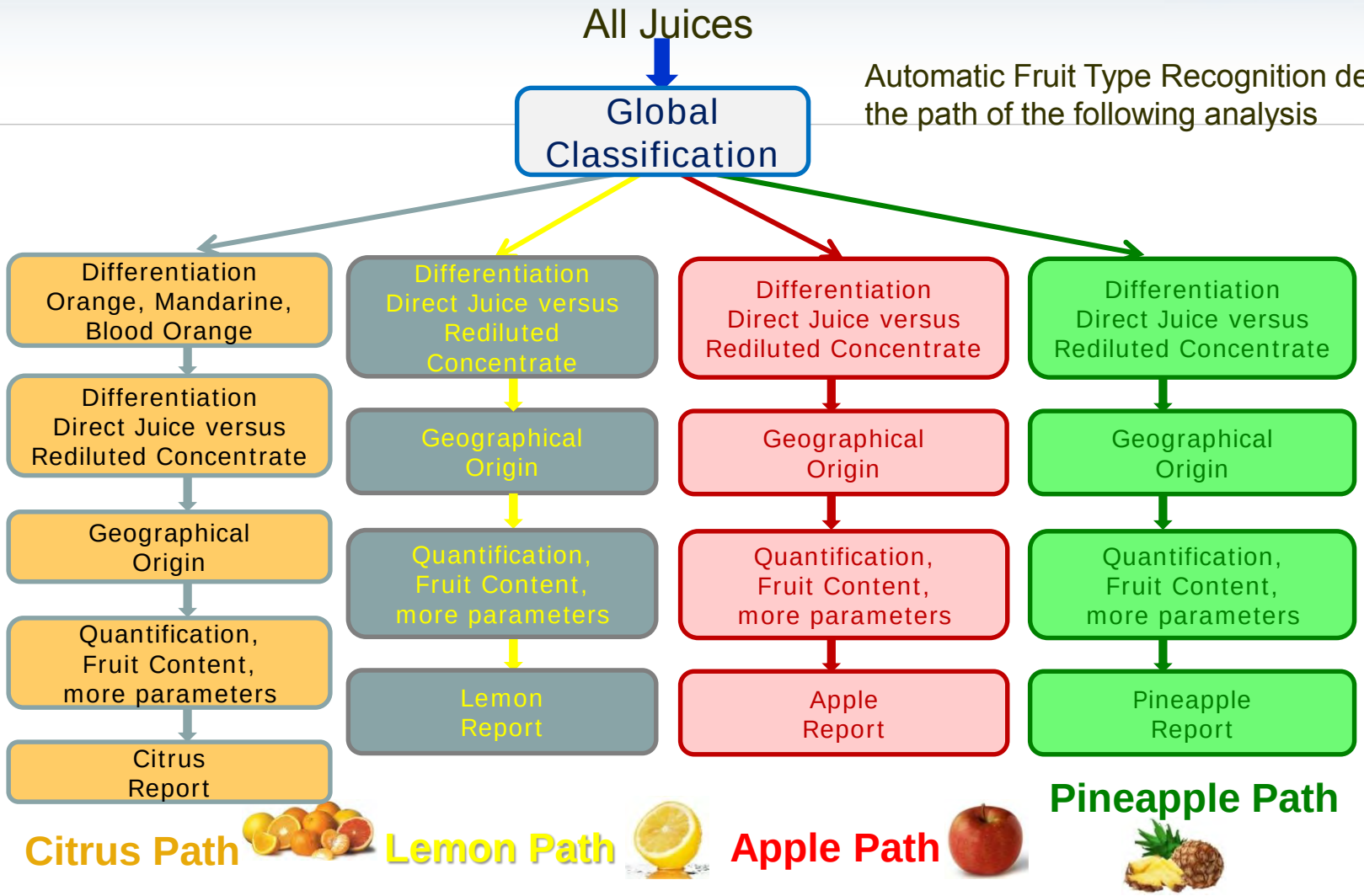


2D J-Resolved谱图（保证化合物信号峰的识别）



Sample: White Wine, aromatic ppm-region

Flowchart of decision making during post processing using cascading models in NMR based juice analysis

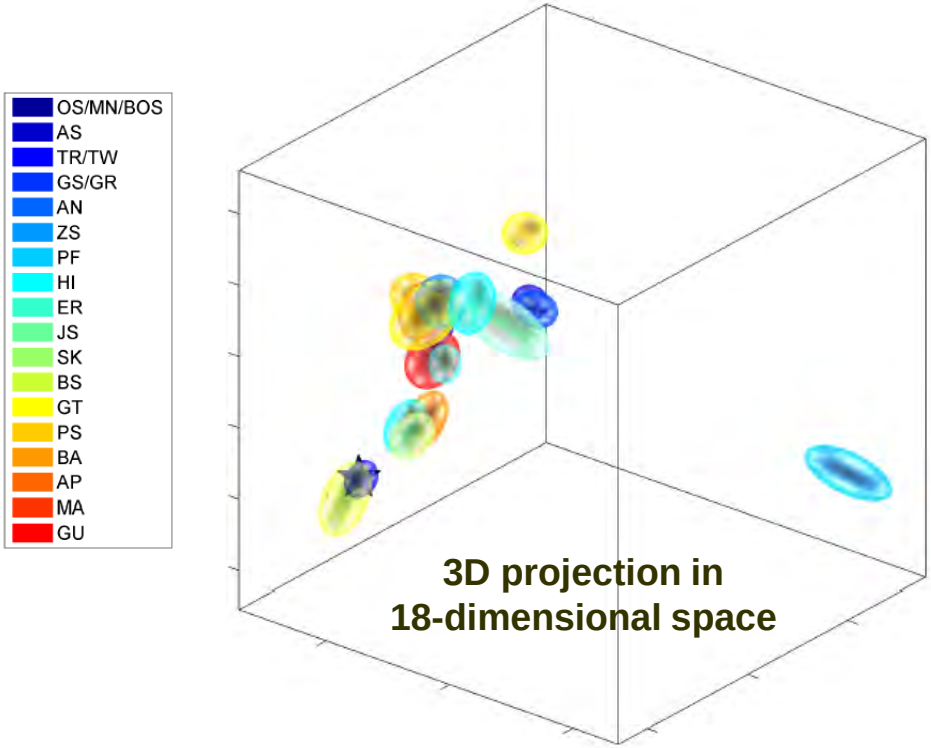


Report Example - Classifications

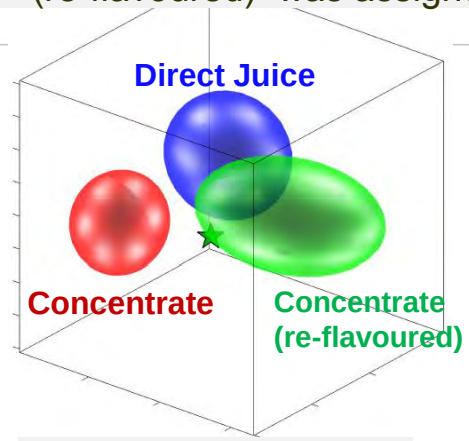
Apple juice(re-flavoured concentrate) from market in China.

Type of Fruit "Apple" was assigned

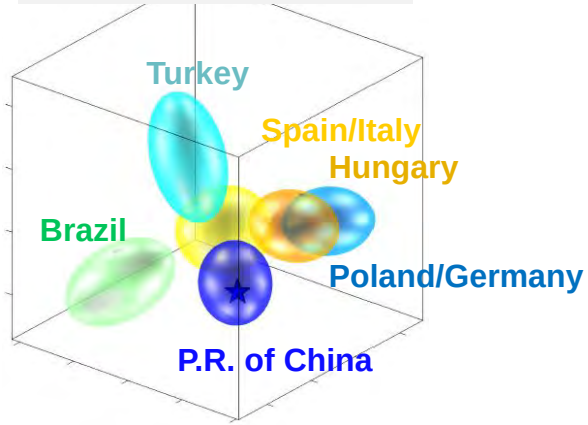
Orange/Mandarin/Blood-Orange, Apple, Grape, Grapefruit, Pineapple, Lemon, Peach, Raspberry, Strawberry, Black Currant, Sour Cherry, Pear, Pomegranate, Passion Fruit, Banana, Apricot, Mango, Guava



Type of Product "Concentrate (re-flavoured)" was assigned



Origin "P.R. of China" was assigned



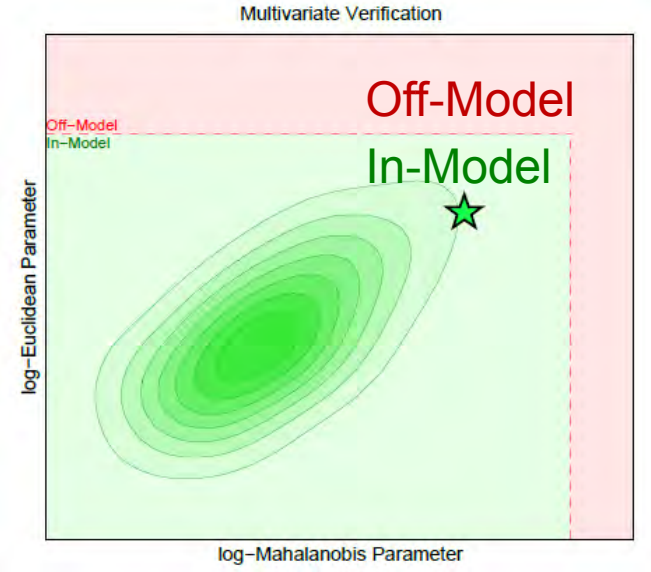
Version 3.0

Report Example - Verification Models

Apple juice(re-flavoured concentrate) from market in China.

- Applied Model: Apple from P.R. of China
- Univariate Verification:
  - Result: No deviation was detected in univariate verication (In-Model).
- Multivariate Verification
  - Result: No deviation was detected in multivariate verication (In-Model).
- Fruit Content
  - Applied Model: Apple

Origin of Fruit	Consistent with 100%
P.R.of China	Yes



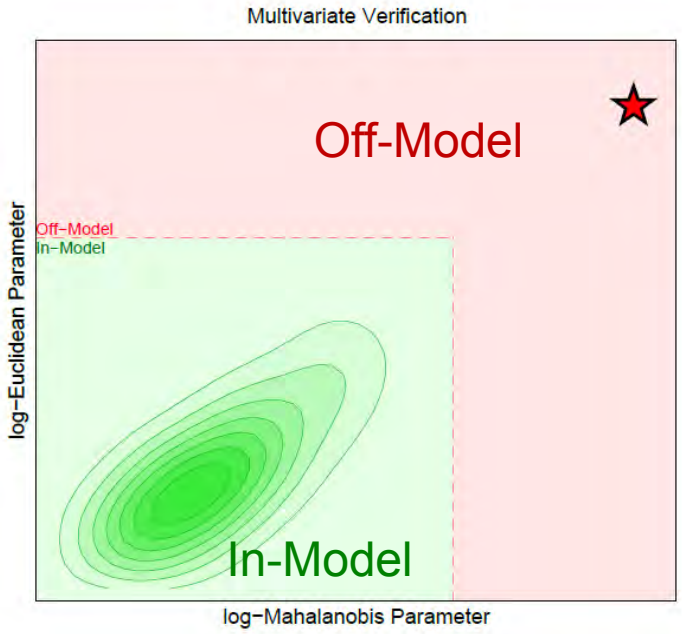
Report Example - Verification Models

Grape juice(re-flavoured concentrate) from market in China.

- Applied Model: Grape
- Univariate Verification:
  - Result: Deviating signals were found at following chemical shifts:

1.393up 1.410up 5.391up 5.396up 5.402up 3.218low 3.242low 3.371low 3.383low 3.395low 3.406low 3.418low 3.442low 3.465low 3.512low 3.524low 3.694low 3.712low 3.729low

- Multivariate Verification
  - Result: Sample was classified as Off-Model in multivariate verification.



Report Example - Targeted Analysis (Quantification)

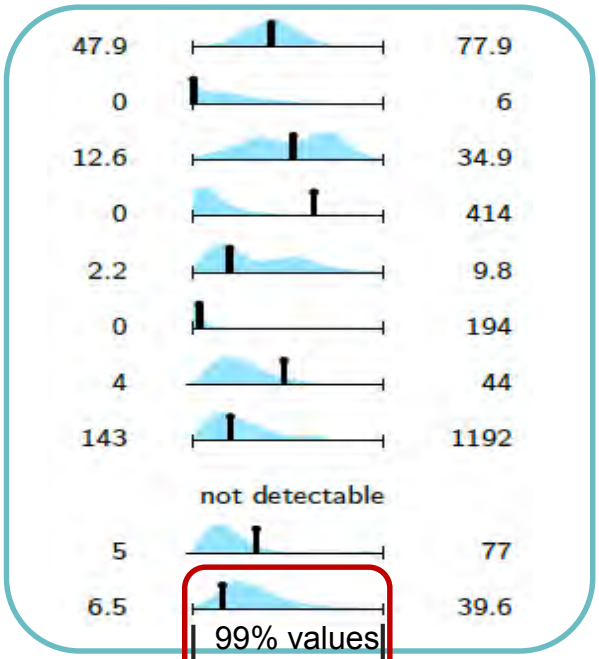
A.I.J.N.: 欧盟果蔬汁协会公布的相应化合物浓度参考范围

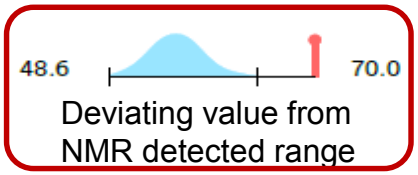
与NMR模型中相应化合物浓度分布进行比较

Quantification Results:

Compound	Value	Unit	LOQ	A.I.J.N. (Apple) Flag	min	max	SGF-Profiling Database n = 1413
ethanol	138	mg/L	10		-	3000	<10 263
lactic acid	63	mg/L	10		-	500	<10 319
5-hydroxymethylfurfural	<5	mg/L	5		-	20	<5 14
Titr. Acidity pH 7*	51	meq/l	-		-	-	21 74
Titr. Acidity pH 8.1*	51	meq/l	-		35	117	22 76
Titr. Acidity (pH 7, tartaric acid)*	3.7	g/l	-		-	-	1.5 5.6
Titr. Acidity (pH 7, malic acid)*	3.2	g/l	-		-	-	1.4 5.0
Titr. Acidity (pH 8.1, citric acid)*	3.2	g/l	-		2.2	7.5	1.4 4.9
citric acid	<0.5	g/L	0.5		-	0.1	<0.5 g/L in reference set
malic acid	4.4	g/L	0.5		3.0	-	2.4 6.8
fumaric acid	<5	mg/L	5		-	5	<5 mg/L in reference set
potassium*	1147	mg/l	-		900	1500	869 1218
magnesium*	47	mg/l	-		40	75	30 53
glucose	30.1	g/L	0.5		15.0	35.0	22.3 34.9
fructose	61.5	g/L	0.5		45.0	85.(	49.1 68.8
glucose/fructose ratio**	0.49	-	-		0.30	0.5(	0.38 0.62
sucrose	13.8	g/L	0.2		5.0	30.(	6.8 22.7
% sucrose**	13	%	1		-		7 22
total sugar**	105.4	g/L	2.0		-		88.5 119.2
alanine	35	mg/L	5		1	5(	8 41
proline	<50	mg/L	50		-		<50 mg/L in reference set
arbutin	<10	mg/L	10		-		<10 mg/L in reference set
benzaldehyde	<5	mg/L	5		-		<5 mg/L in reference set
benzoic acid	<10	mg/L	10		-		<10 mg/L in reference set
chlorogenic acid	<20	mg/L	20		-		<20 185
citramalic acid	40	mg/L	10		-	-	<10 86
formic acid	<5	mg/L	5		-	-	<5 13
galacturonic acid	782	mg/L	100		-	-	<100 1721
malic/quinic ratio**	11.3	-	-		-	-	4.3 30.6
methanol	27	mg/L	10		-	-	<10 43
pyruvic acid	<10	mg/L	10		-	-	<10 10
quinic acid	391	mg/L	50		-	-	201 893
sorbic acid	<10	mg/L	10		-	-	<10 mg/L in reference set
succinic acid	24	mg/L	10		-	-	<10 47
xylose	341	mg/L	300		-	-	<300 721

Distribution curves get better representative with every sample measured

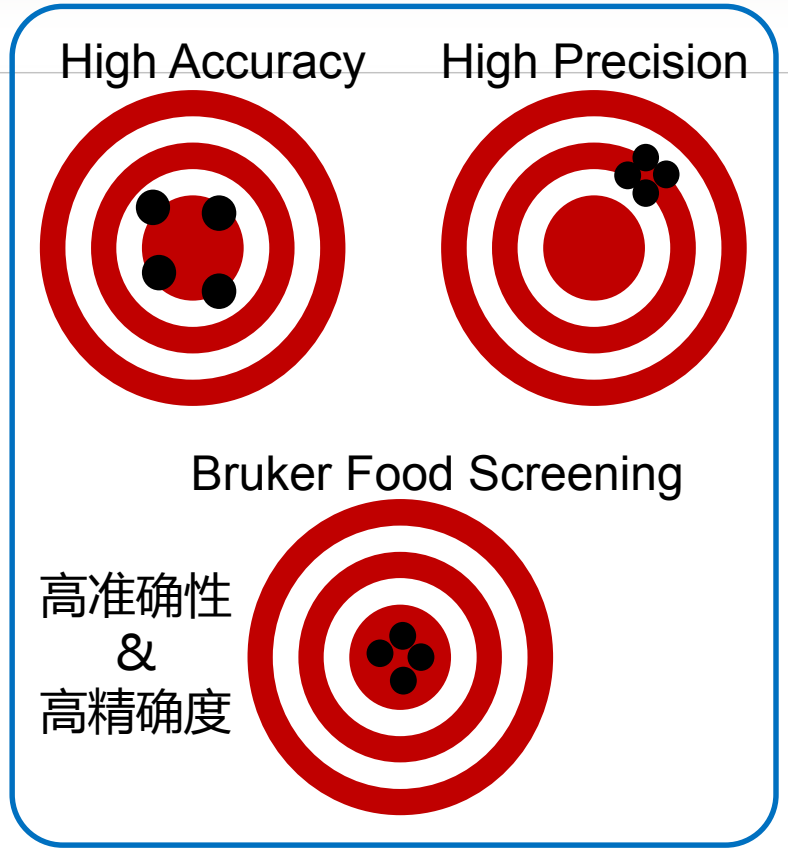




Quality validation of Bruker NMR-based food screening
基于NMR的食品筛选结果的验证—确定分析结果的可靠性

For Food screening the following validation methods are continuously applied:

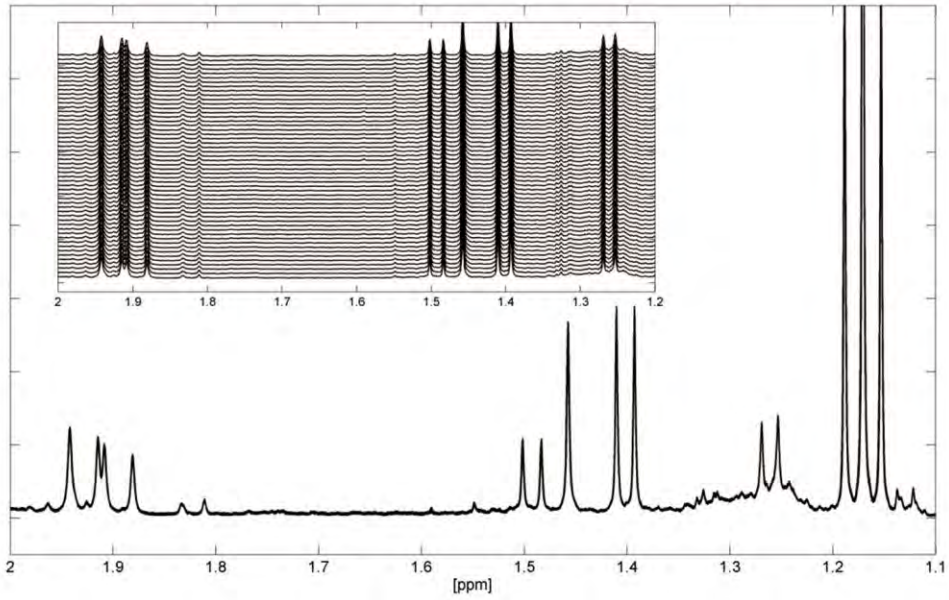
- Proof of highest-level reproducibility
- Proof of inter-instrumental identity of results
- Comparison with official reference methods (e.g. IFU)
- Participation in Proficiency Tests by FAPAS®



These tests prove the high precision and high accuracy of Bruker SGF/Wine-Profiling™

Highest Level of Reproducibility
最高水平的重现性

One main advantage of our NMR-based screening systems is the highest level of reproducibility of the complete 1H-NMR fingerprint, and therefore of all the extracted results - quantification as well as results from statistical models.

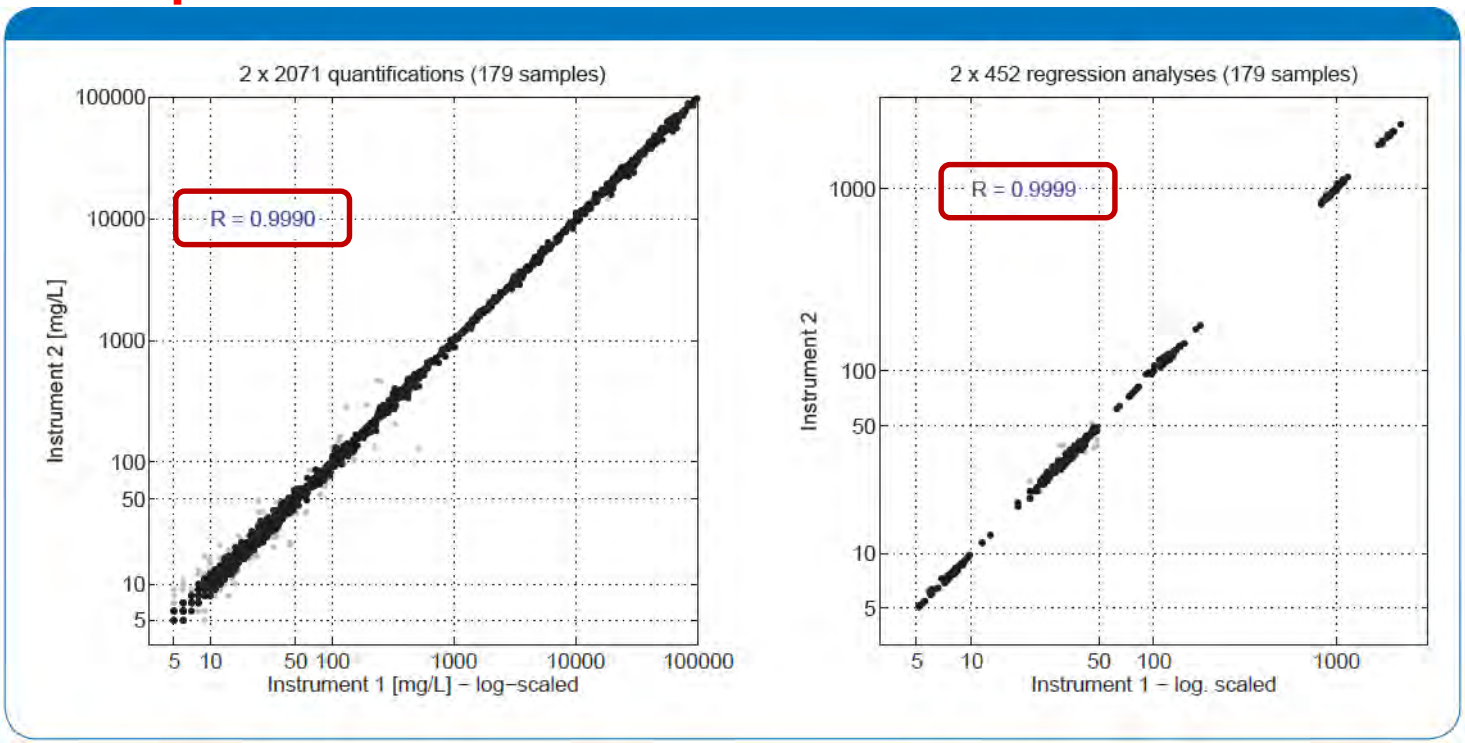


Highest precision is proven

50 NMR-spectra of replicate preparations and measurements of one apple juice

Inter-Instrumental Comparison at 400 MHz
不同的2台400MHz的定量结果比较

The **same sample** measured on **two equivalent machines** (same field-strength, same hardware) **generates the same fingerprint** and therefore **equivalent quantification and statistical results**.



This is a fundamental requirement to develop a reliable analysis across the border of one laboratory.

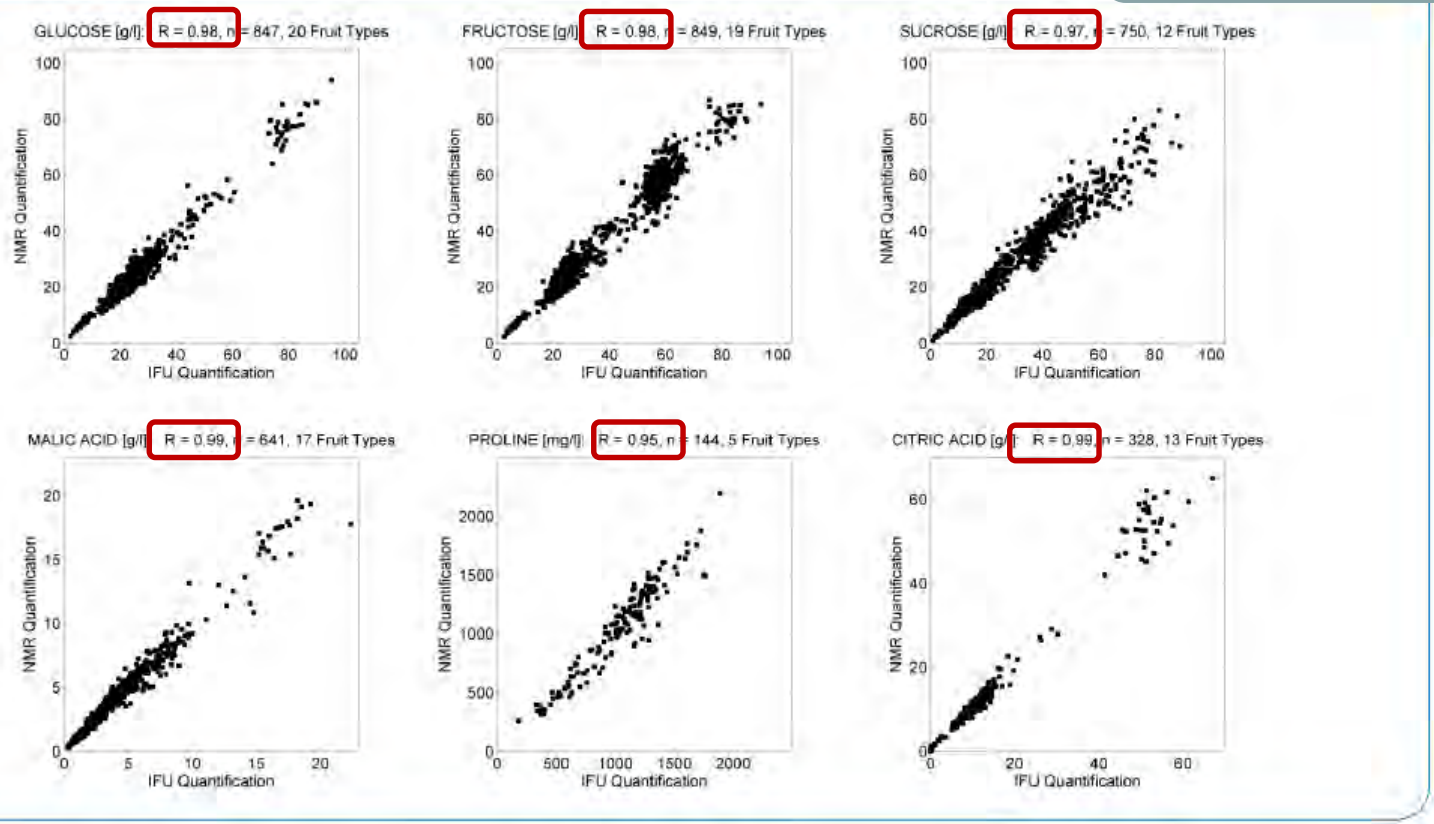
Comparison with Official Reference Methods
与官方参考方法的结果比较

Very good correlations with reference methods

More than 3500 comparisons of 20 different types of fruit

High accuracy is proven

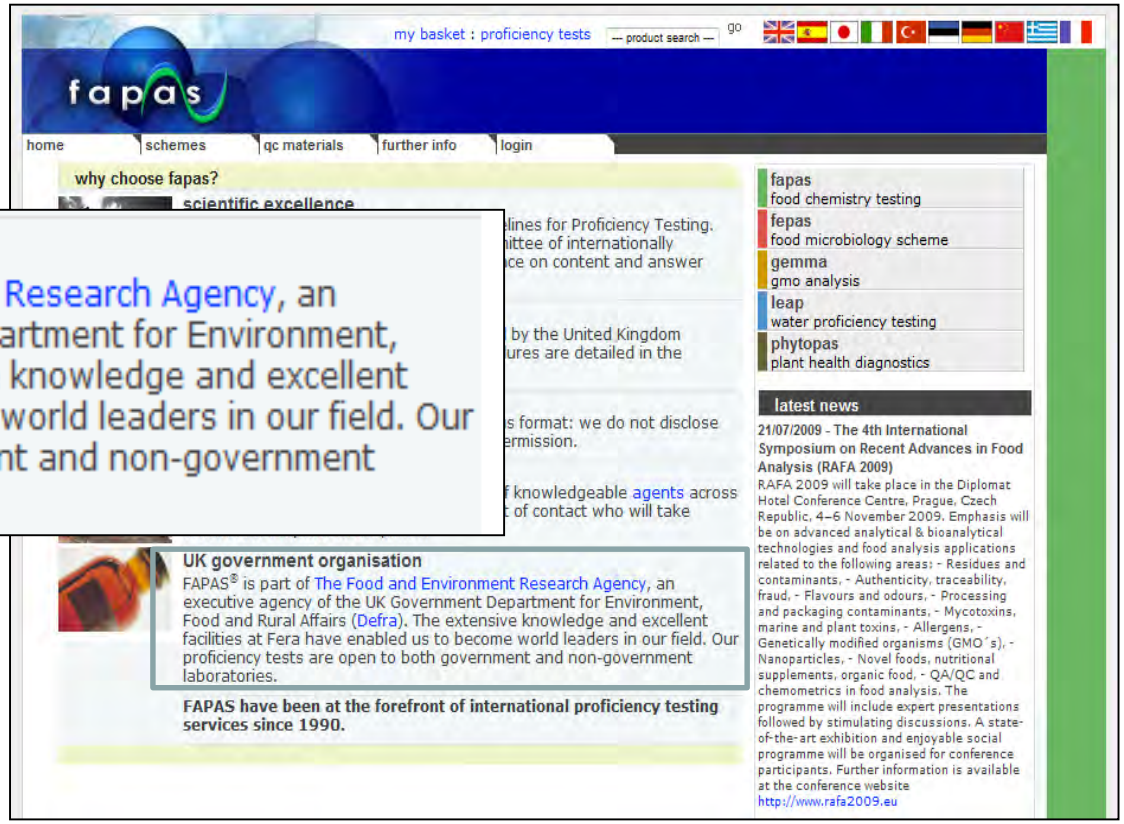
Validation of NMR results
Against IFU standard methods



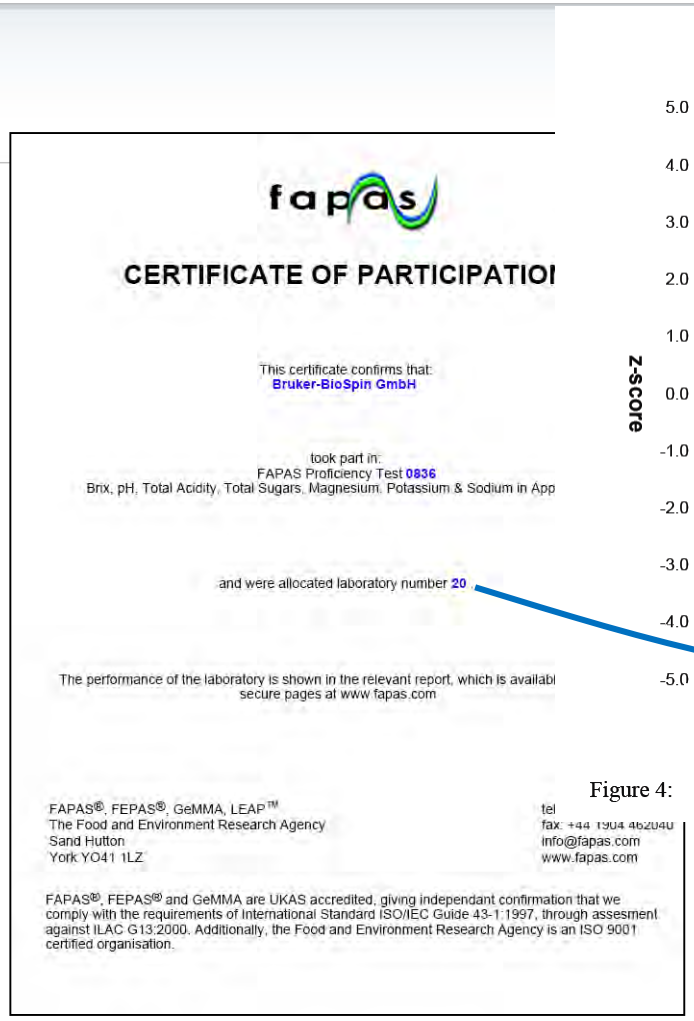
FAPAS®:Food Analysis Performance Assessment Scheme
FAPAS®:食品分析能力评估测试

- Inter-labor comparison

  Constant testing of analysis quality by participation in official and certified ring tests, is of high priority for Bruker BioSpin NMR-based juice and beverage screening.



FAPAS® Results of the JuiceScreener


fapas

CERTIFICATE OF PARTICIPATIOI

This certificate confirms that:
Bruker-BioSpin GmbH

took part in:
FAPAS Proficiency Test 0836
Brix, pH, Total Acidity, Total Sugars, Magnesium, Potassium & Sodium in App

and were allocated laboratory number 20

The performance of the laboratory is shown in the relevant report, which is availabl
secure pages at www.fapas.com

FAPAS®, FEPAS®, GeMMA, LEAP™
The Food and Environment Research Agency
Sand Hutton
York YO41 1LZ

tel
fax: +44 1904 462040
info@fapas.com
www.fapas.com

FAPAS®, FEPAS® and GeMMA are UKAS accredited, giving independant confirmation that we comply with the requirements of International Standard ISO/IEC Guide 43-1:1997, through assesment against ILAC G13:2000. Additionally, the Food and Environment Research Agency is an ISO 9001 certified organisation.

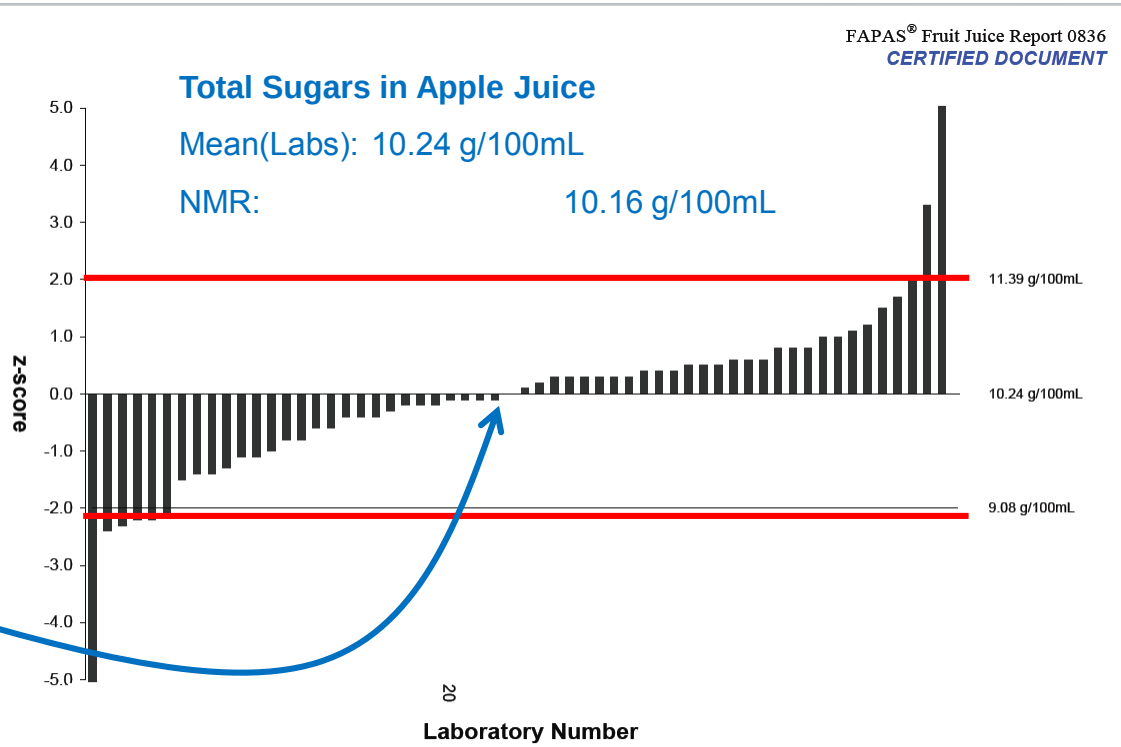


Figure 4: z-Scores for Total Sugars (10.24 g/100mL) in Apple Juice Test Material

z-Score:
Value = *Median* + Z * *StdDev*
Values with a z-score
greater than 2 are rejected
(no successful participation)

Some other Examples on FAPAS® Results

Material	Compound	Unit	Min-Conf	Max-Conf	Z-Score Chart	NMR-Value
Apple Juice	Potassium	mg/L	930.0	1166.0	-0.4	1024.0
Apple Juice	Magnesium	mg/L	39.9	57.3	1.0	53.0
Apple Juice	Total Sugars	g/100mL	9.1	11.4	-0.1	10.2
Apple Juice	Total Acidity	g/100g	0.4	0.5	-0.7	0.4
Tonic Water	quinine	mg/L	59.5	83.6	0.1	72.0
Tonic Water	Benzoic Acid	mg/L	102.0	139.0	-1.0	111.0
Tonic Water	aspartame	mg/L	86.0	119.0	0.1	103.0
Tonic Water	acesulfame-K	mg/L	84.0	117.0	-0.6	96.0
Soft Drink	Saccharin	mg/L	12.8	19.6	-1.3	14.0
Soft Drink	Cyclamate	mg/L	165.0	221.0	1.4	213.0
Soft Drink	Citric Acid	mg/L	2826.0	3422.0	-0.2	3100.0
Cola Soft Drink	Benzoic Acid	mg/L	122.0	165.0	0.0	144.0
Cola Soft Drink	Caffeine	mg/L	89.0	122.0	0.7	111.0
Orange Juice	Potassium	mg/L	1576.0	1942.0	1.2	1870.0
Orange Juice	Total Sugars	g/100mL	8.1	9.1	-0.2	8.6
Apple Juice	Potassium	mg/L	912.0	1143.0	0.9	1082.0
Apple Juice	Magnesium	mg/L	38.7	55.6	0.4	49.0
Orange Juice	Total Sugar	g/100mL	8.2	9.2	1.2	9.0
Orange Juice	Sucrose	g/100mL	2.9	3.3	1.2	3.2
Orange Juice	Glucose	g/100mL	2.5	2.9	1.6	2.8
Orange Juice	Fructose	g/100mL	2.7	3.1	0.9	3.0

Z-Scores in the range of -2 and 2 cover the range of minimum and maximum confidence (green region).

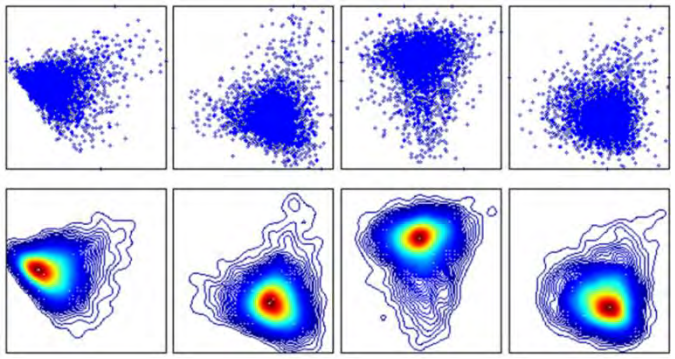
Correspog Z-Score of SGF-Profiling

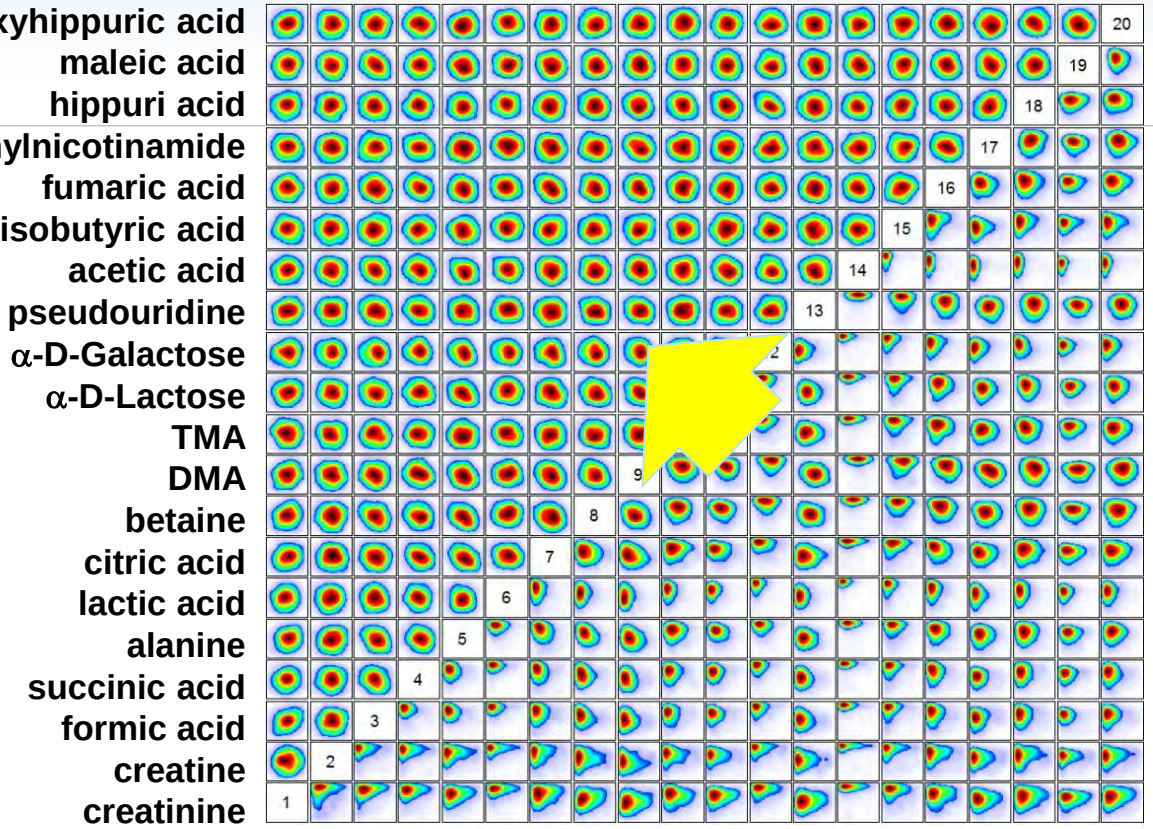
Regression Results (based in Proton-Spectrum)

Reconstitution of gaussian distribution for secure outlier determination
高斯分布的重建—— 确保多变量统计分析结果

no solutions offered in commercial programs !

Multivariate distributions are not Gaussian!!





Conversion part of PHD B.Schütz

Fruit Content in Red Fruits
– Case Study: Raspberry Puree

BRUKER

Conventional parameter (Calculated at Brix 8.5°)

Potassium	2161 ppm	√
Phosphate	636 ppm	√
Magnesium	135 ppm	√
Isocitric acid	106 ppm	√
Formol number	18.9 ml/100ml	√
Citric/isocitric ratio	151	√
Glucose/Fructose ratio	0.82	√

Fruit Content Estimation

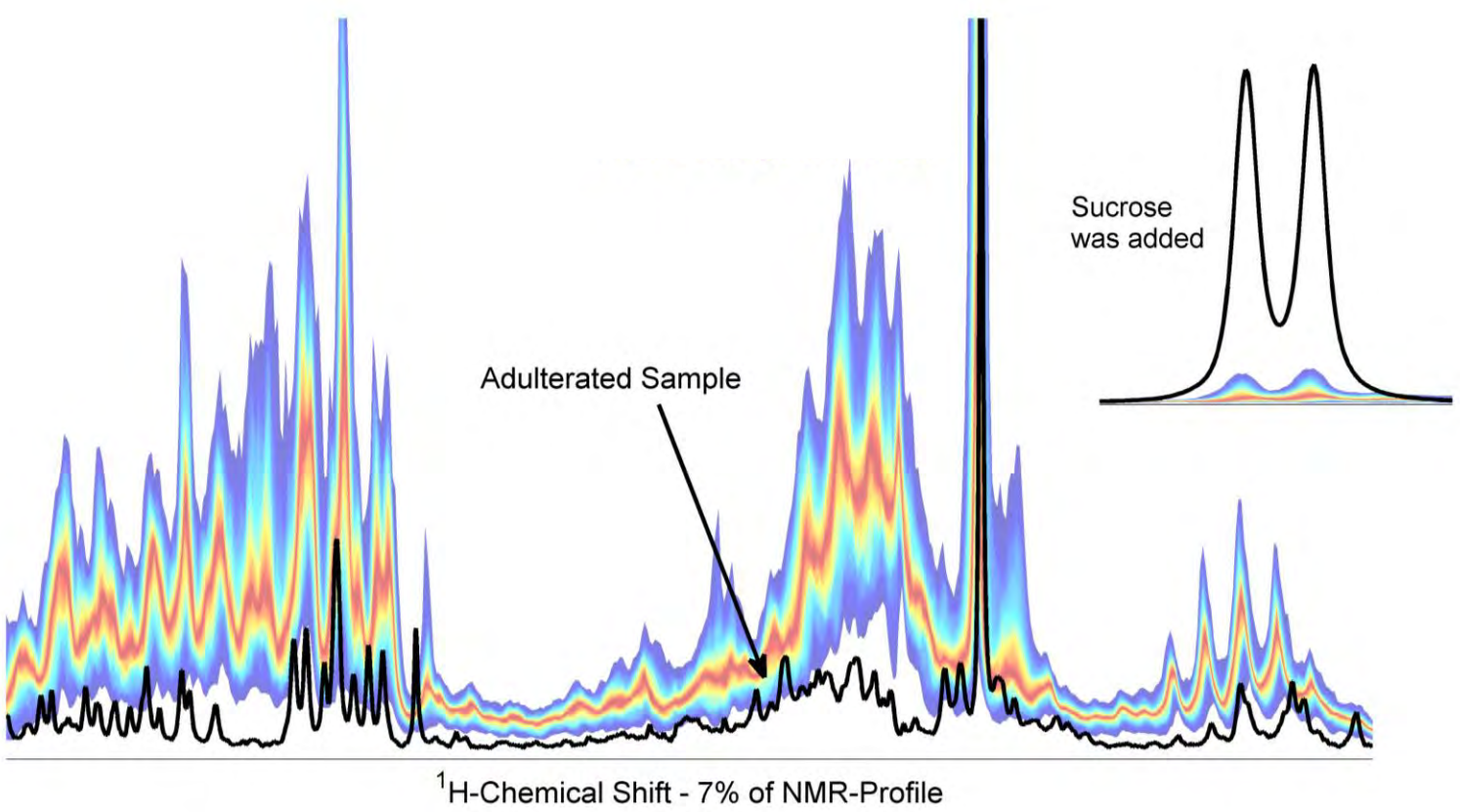
105 % √

Isotopic profile

		Difference
δ13C- Sugar	-24.7 ‰ V-PDB √	-0.2 ‰ V-PDB √
δ13C- Acids	-24.9 ‰ V-PDB √	
δ13C- Pulp	-23.9 ‰ V-PDB √	-0.85 ‰ V-PDB **Fail !**

Added sugar: 20% at least

Fruit Content in Red Fruit Juice



- Spectrums are normed on the total sugars
- Substrate reflects the fruit content
- **Black** sample doesn’t seem to have a 100% fruit content

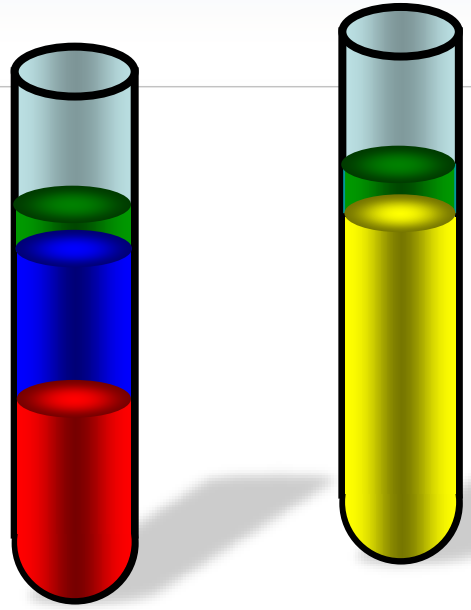
Wine Analysis and Requests

Wine by NMR:

- Replaces conventional, targeted wine analysis
  - Quantification of 56 parameters
- Determination of grape variety (22 types)
- Mix of grape varieties, 15% foreign grape variety allowed (Europe)
- Country classification (Red wine)
- Selected region classification for the major European wine-producing countries (Red wine)
- Verification of Vintage (White wine)
- Indication for addition of water
- Detects irregularities of any kind (untargeted analysis)

- SOP`s for preparation, measurement and Quantification established
- Statistical models established

Very Simple Preparation
- can be done by robotic system



Buffer 10%

D2O 40%

Plasma 50%

10%Buffer

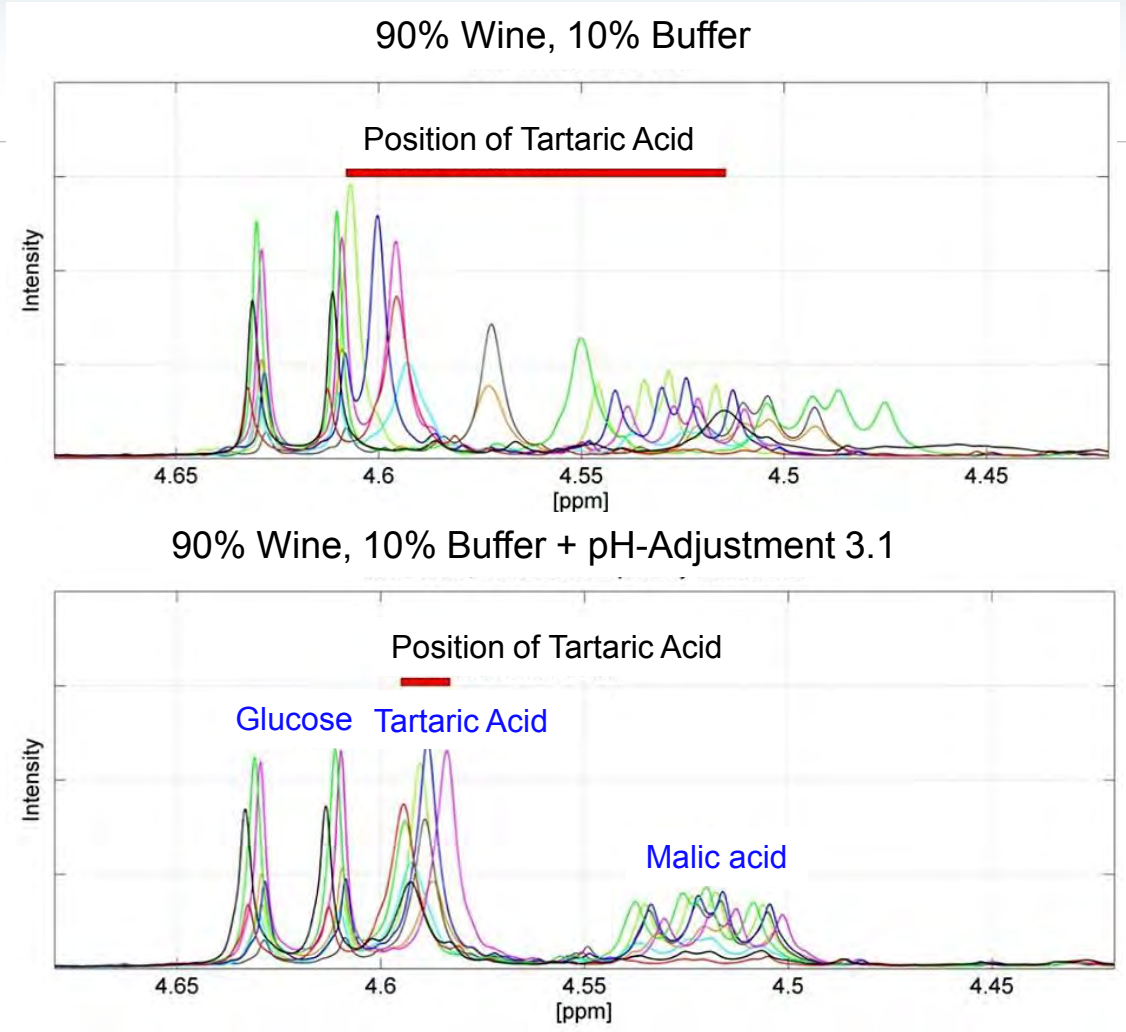
90%Urine/ Juice/ Wine
Might need centrifugation
For Wine precise pH Adjustment

Total sample volume typically 600µL
in 5mm diameter tubes

pH-Effect on NMR signal positions of Wine

Without pH adjustment

With pH adjustment



Preparation Tool for Wine Analysis

Automatic pH-adjustment unit for small volumes ~1ml (Bruker patent)

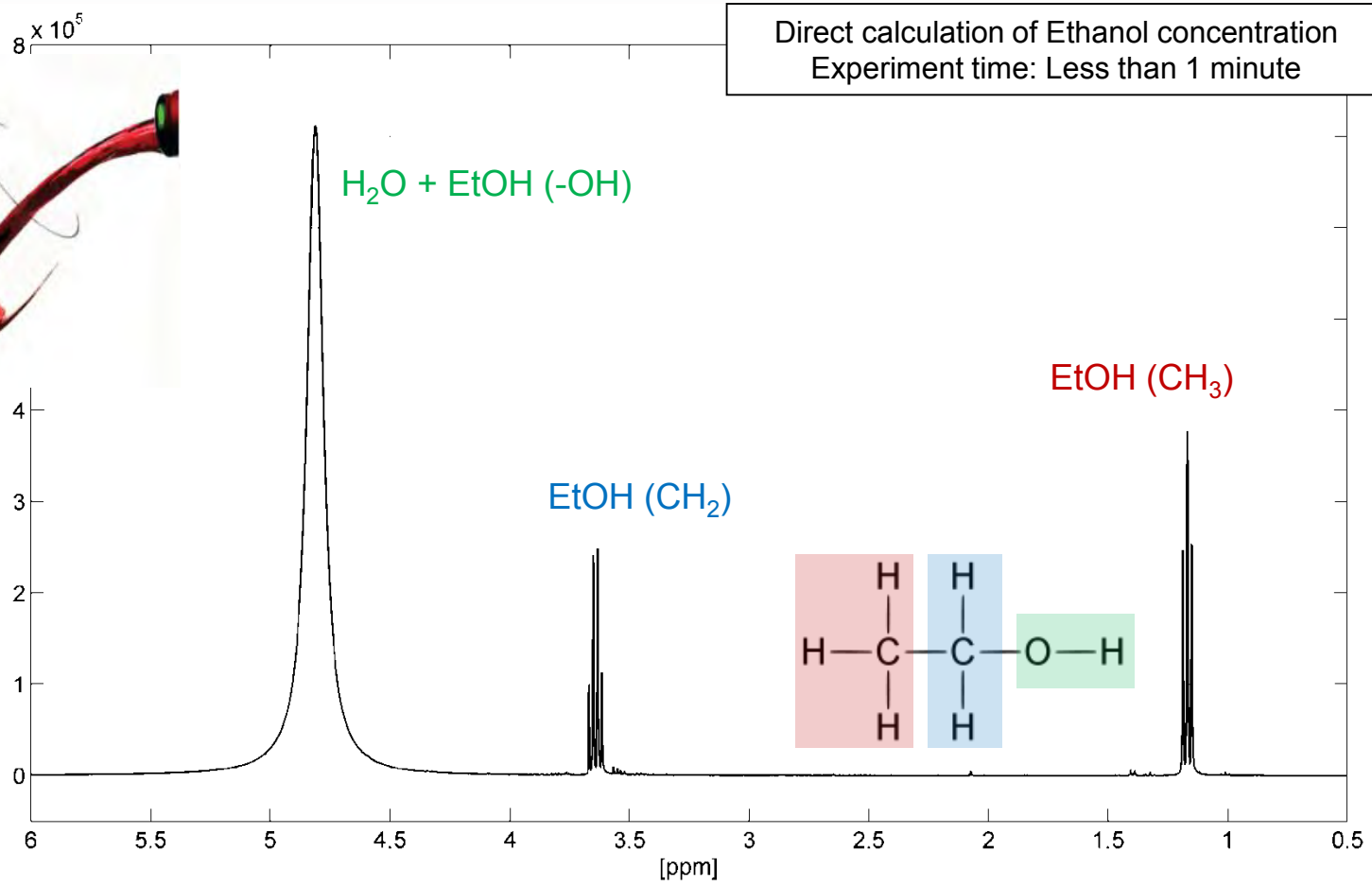
pH Titration Unit with high precision pumps, syringes with minimized void volume, vial shaker and touch panel for control

Special
pH Electrode

Wine-Screener™

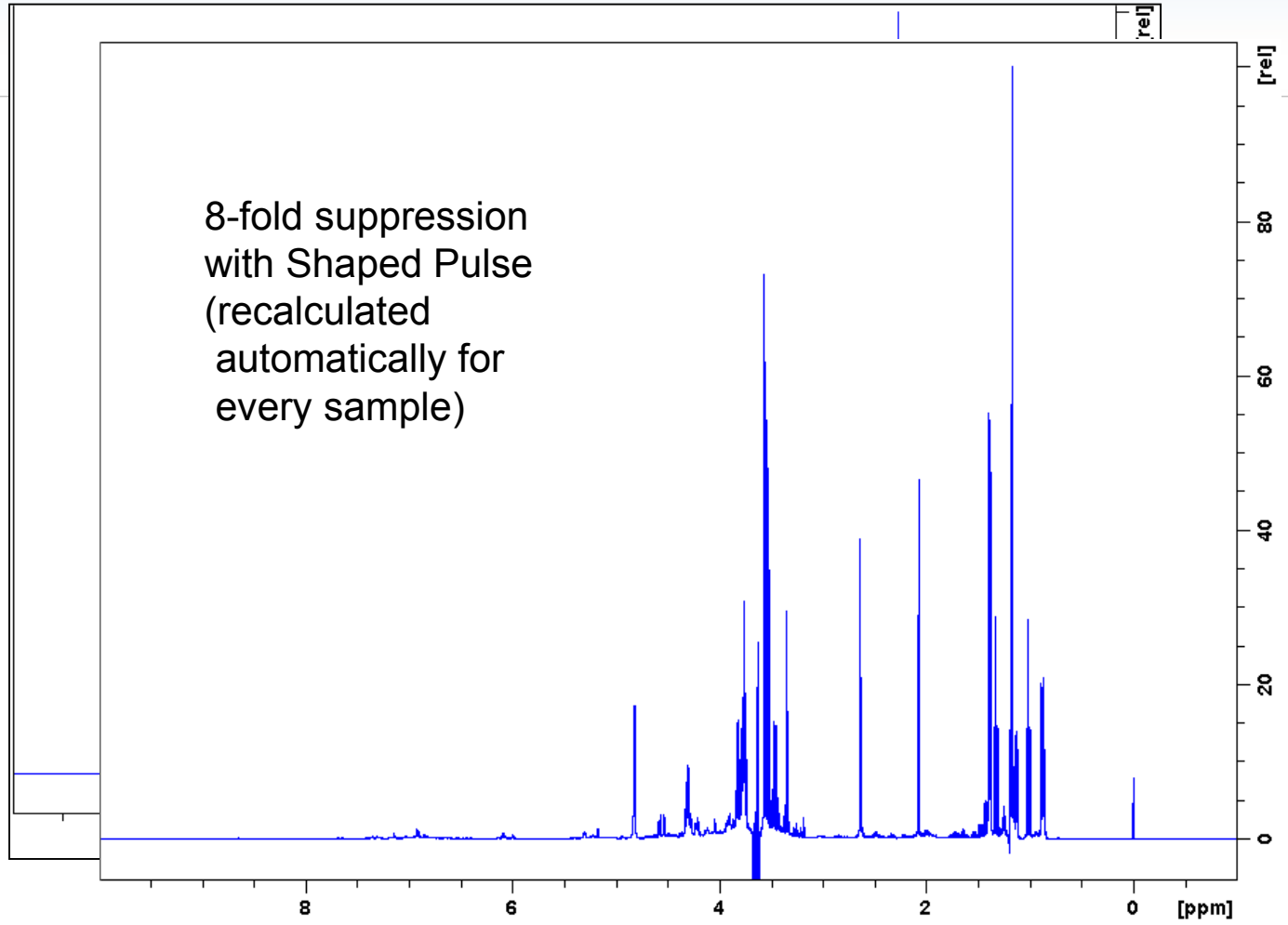
Direct measurement of a wine

Direct calculation of Ethanol concentration
Experiment time: Less than 1 minute

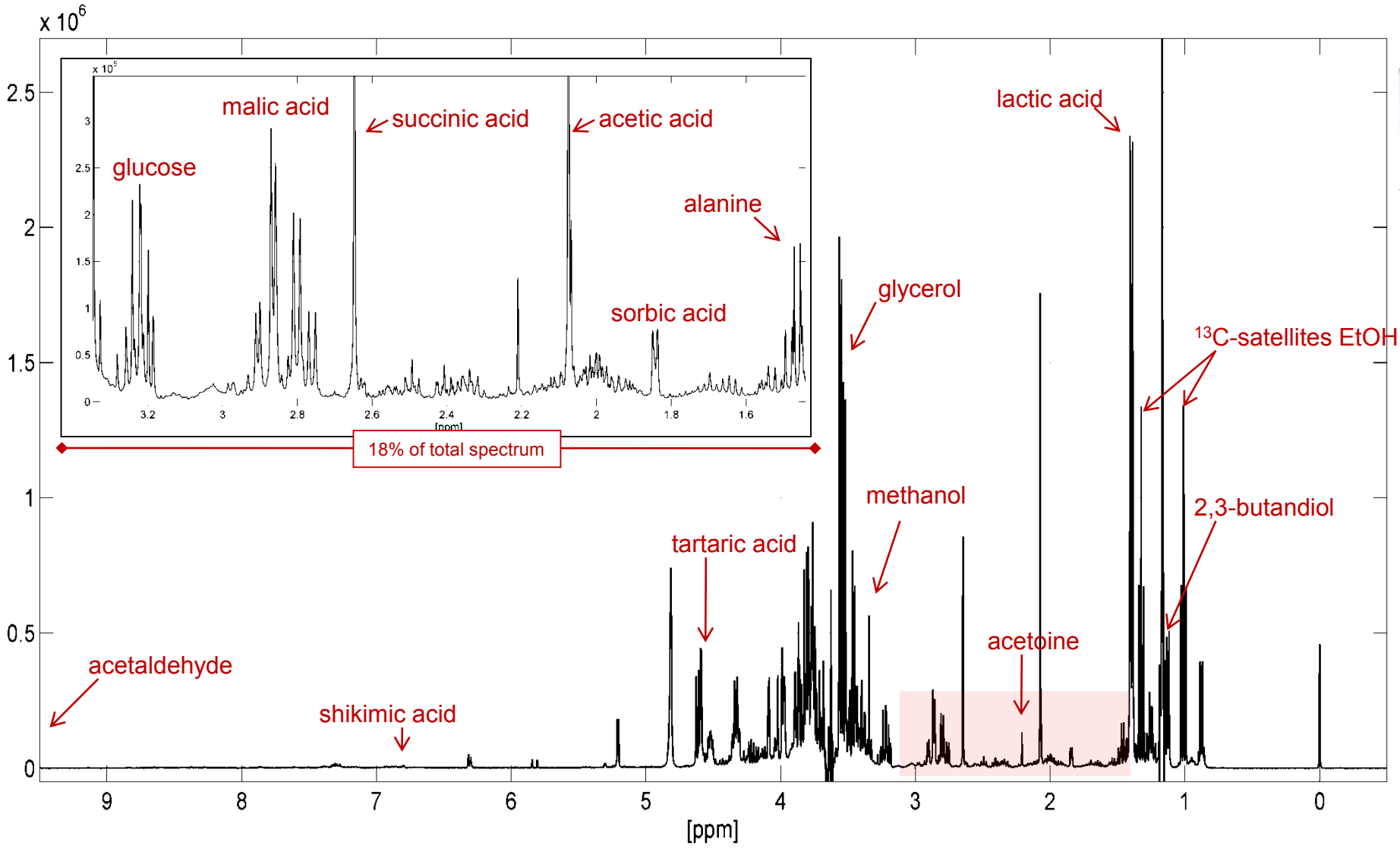


Solvent suppression

—— Important for NMR based quantification in mixtures and to detect low concentrated compounds



1H-NMR spectrum of wine (with suppression)
Currently ~60 compounds quantified

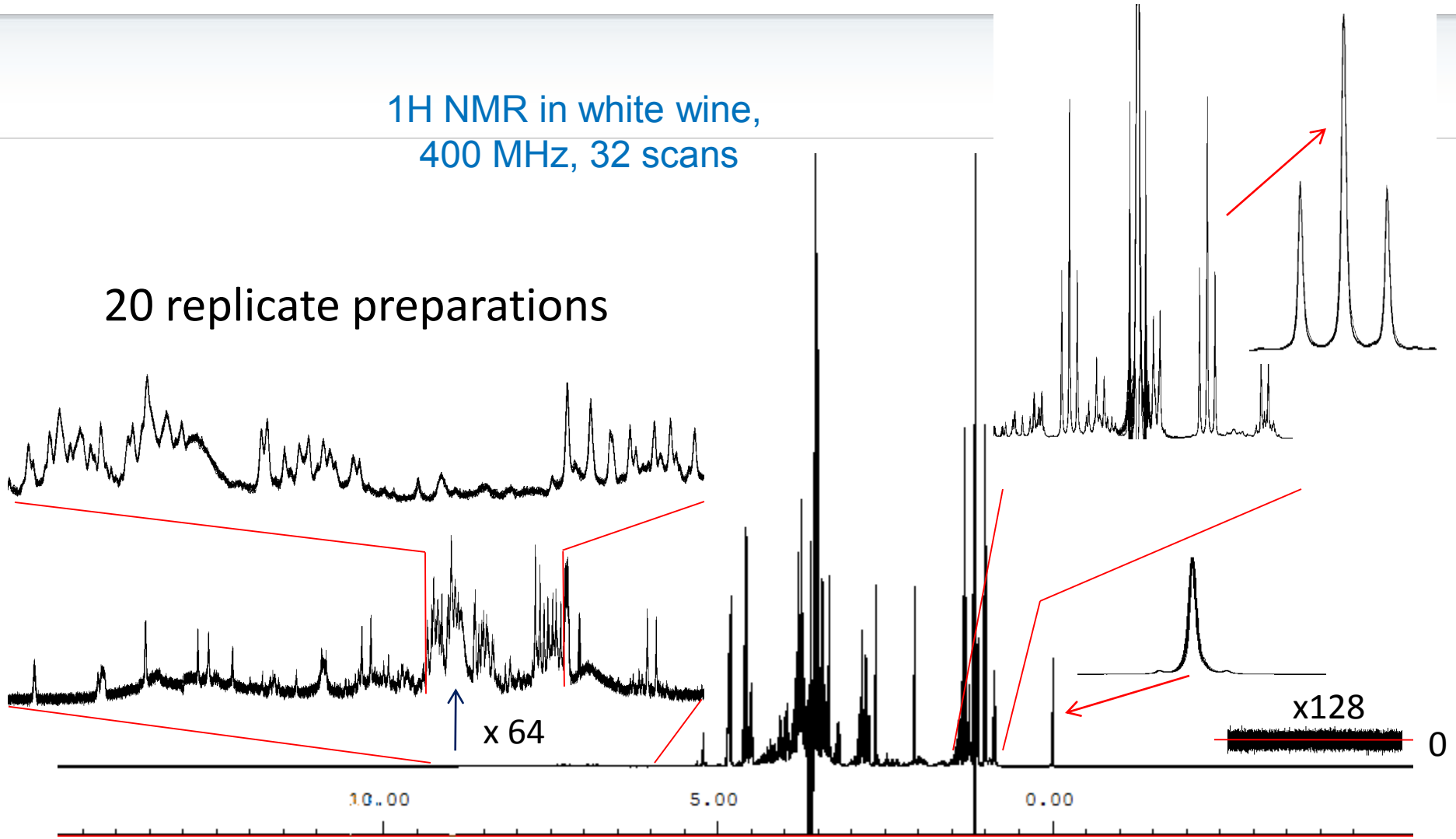


Additional compounds: HMF, trigonelline, sucrose, fructose, citric acid, fumaric acid, proline, …

Reproducibility in Wine NMR

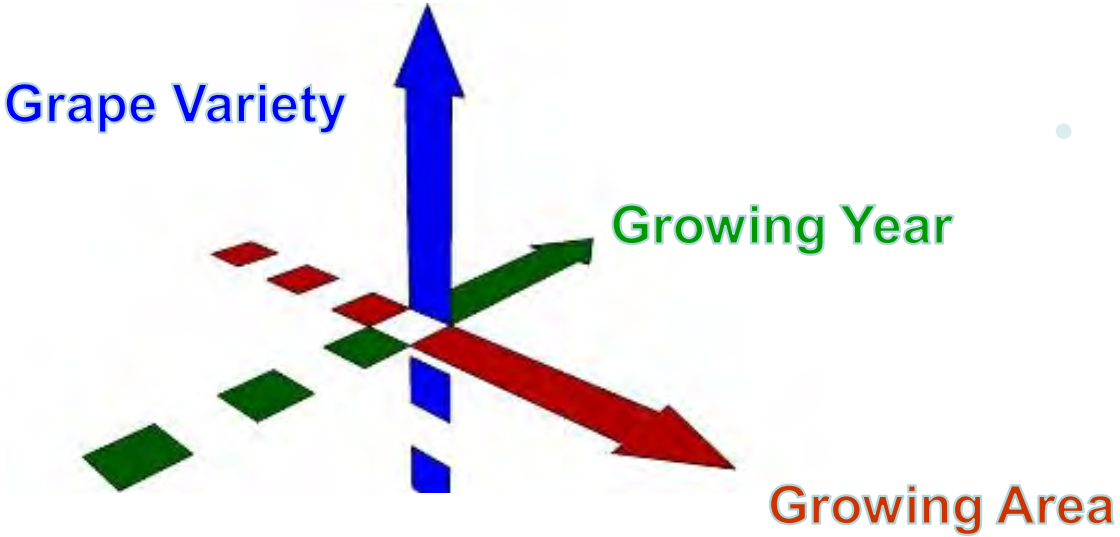
1H NMR in white wine,
400 MHz, 32 scans

20 replicate preparations



Statistical Modelling with authentic wine

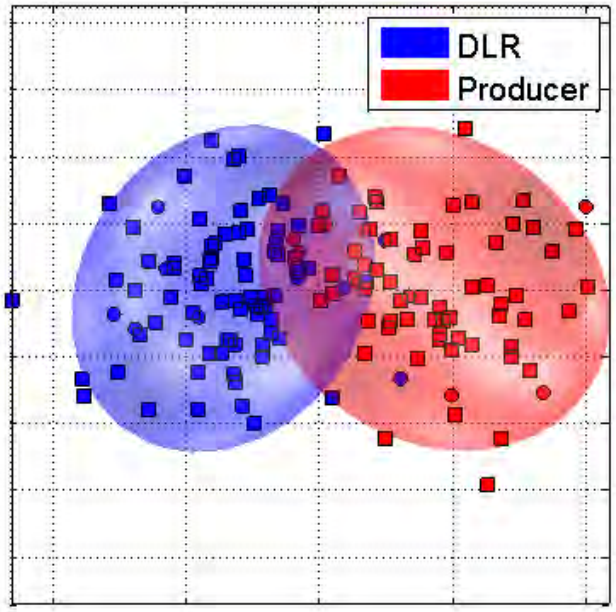
- In cooperation with several wine laboratories more than 13,000 wine samples have been collected and measured at 400 MHz
- NMR, once trained, can predict parameters, that are not related to a special molecule
- NMR can deliver statistical results beyond quantification.

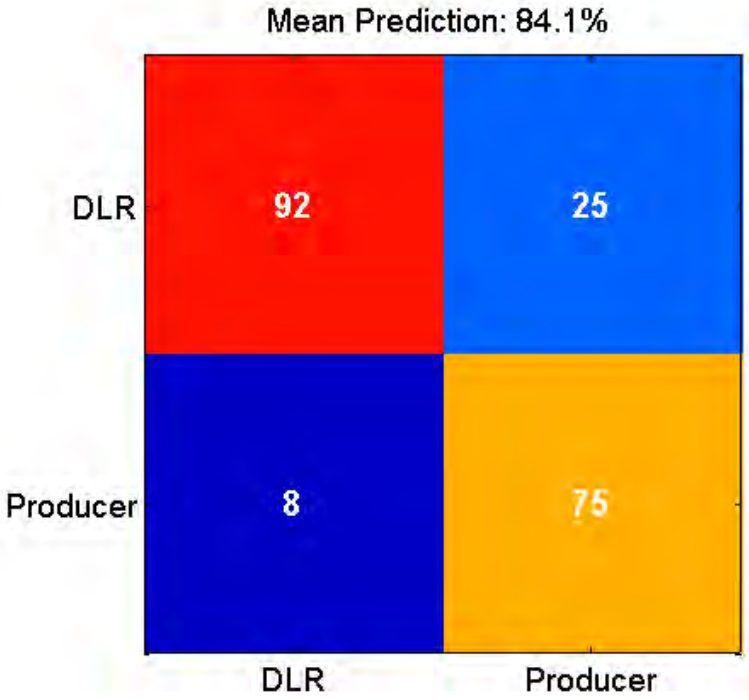


- In statistical modelling, we have to deal with orthogonal parameters that influence the spectra considerably

Vinification versus Micro-Vinification

Cooperation with Prof. Dr. Fischer, DLR Rheinland-Pfalz and CVUA



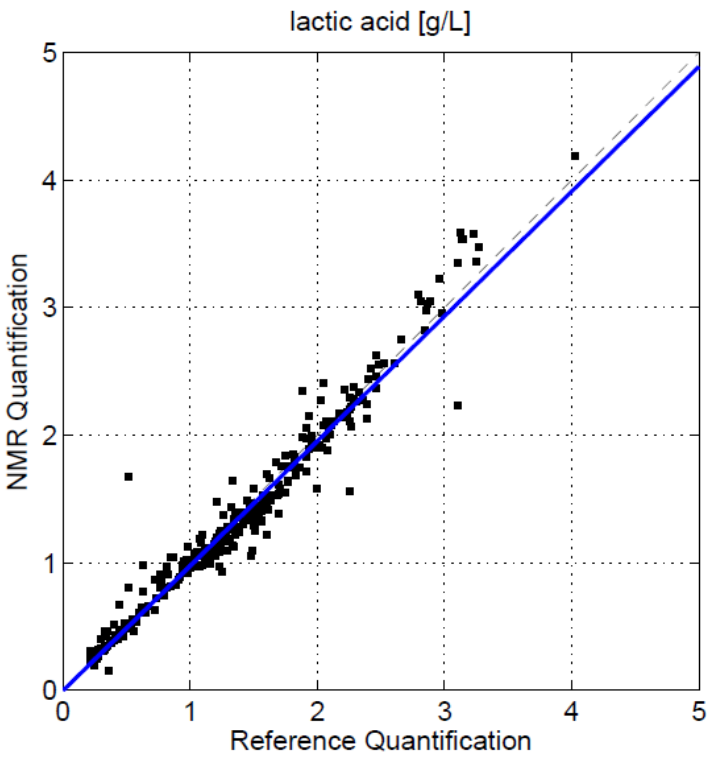


Consequences: We have removed all spectra from microvinification in the authentic database

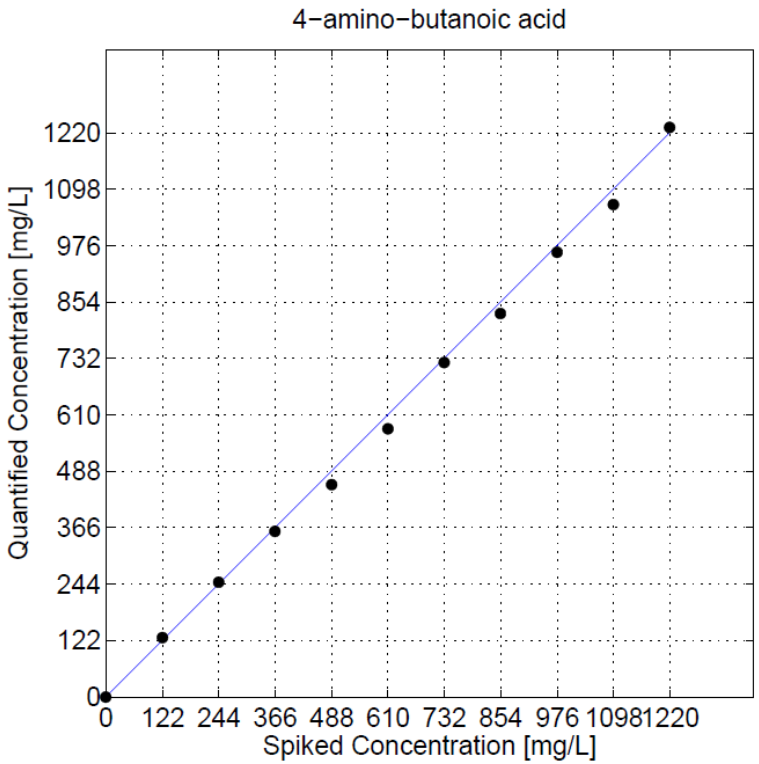
Validation Quantification
NMR定量结果的验证

The Quantification methods are validated and optimized by Spiking-Studies and by comparison with conventional analyses

Comparison to conventional analysis(FT-IR)

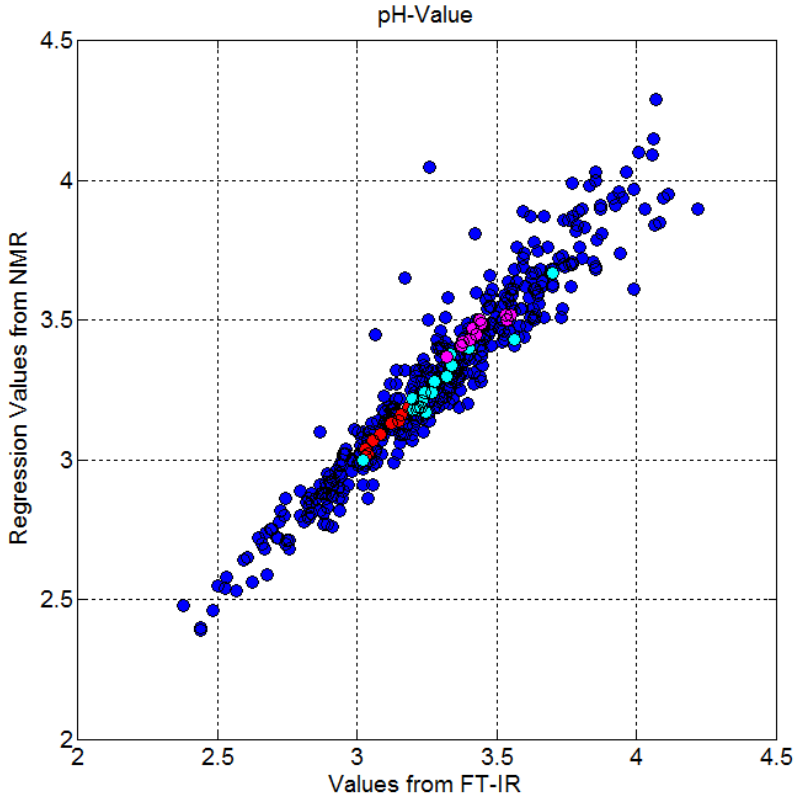


Validation by manual Spiking-studies according to DIN 32645 (LOD/LOQ)

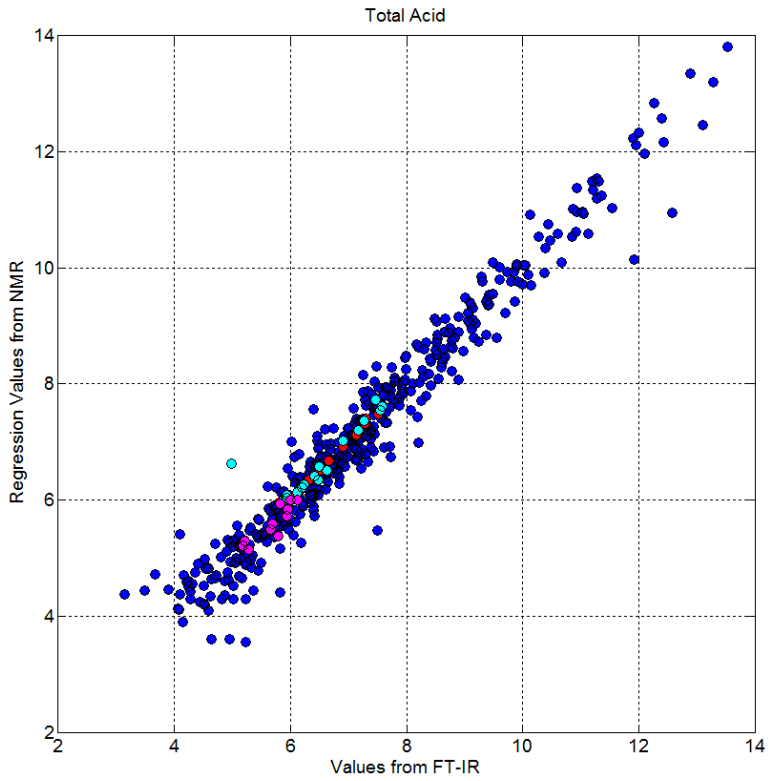


pH and total acids via Regression
NMR回归分析的结果验证

pH value



Total acid

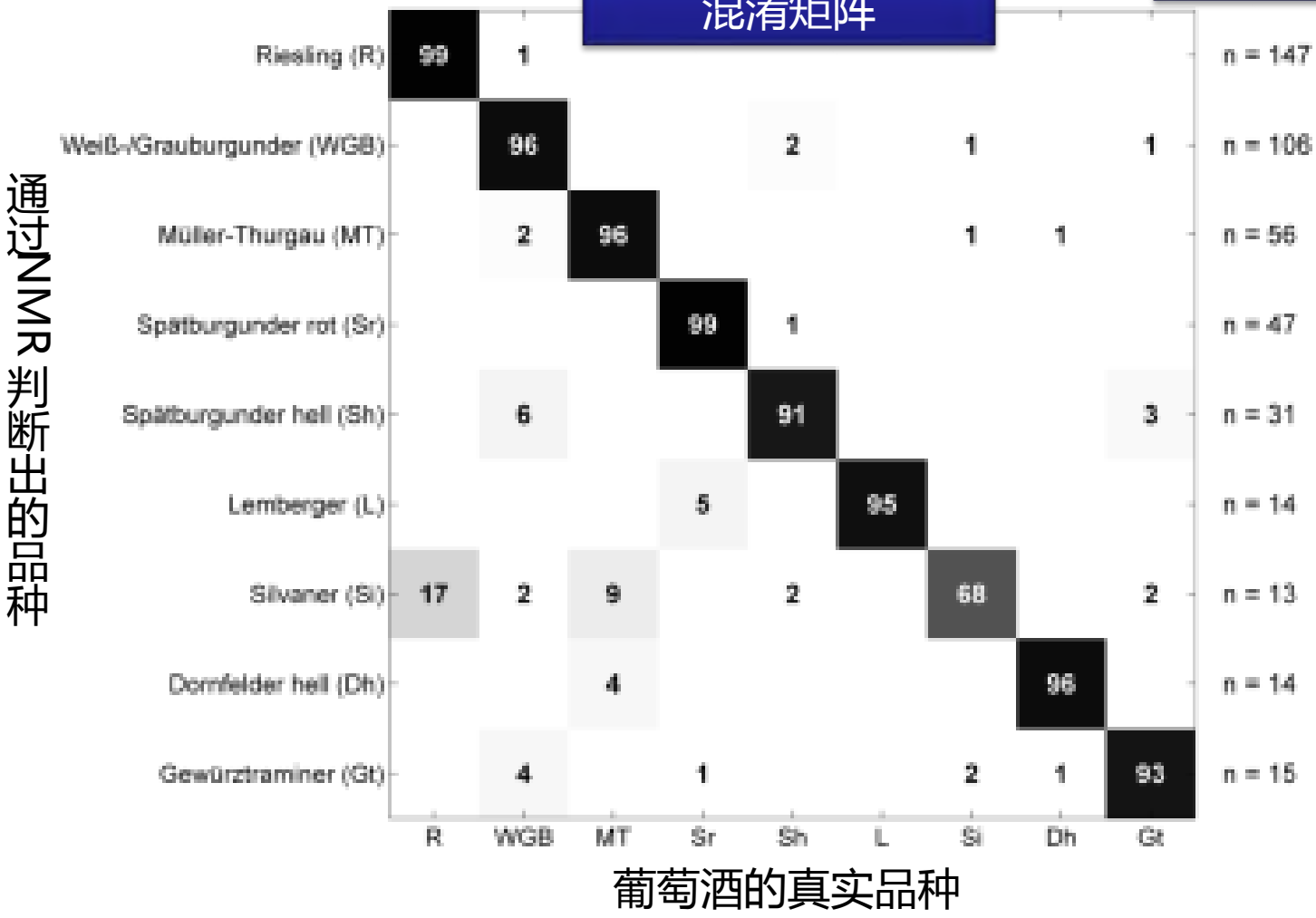


Classification of Grape Varieties
葡萄品种分类结果的验证

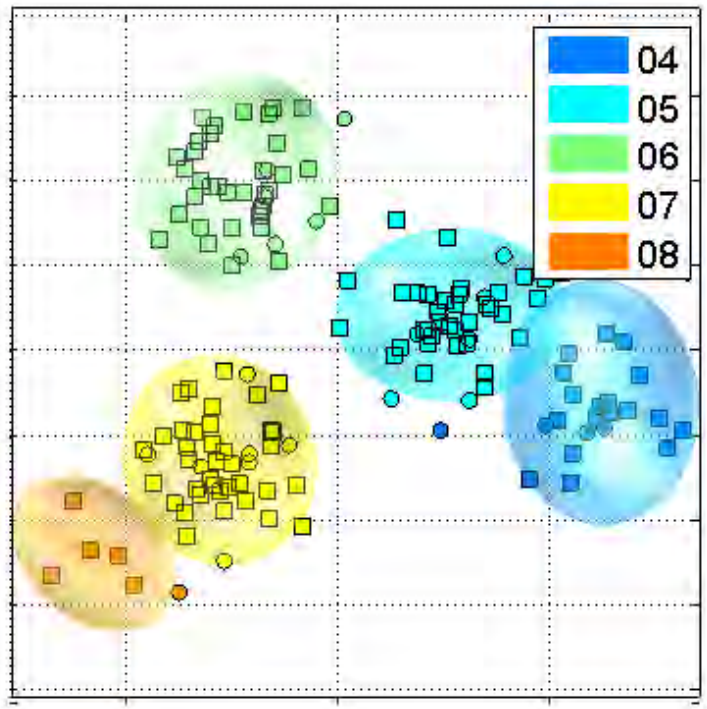
Confusion matrix
混淆矩阵

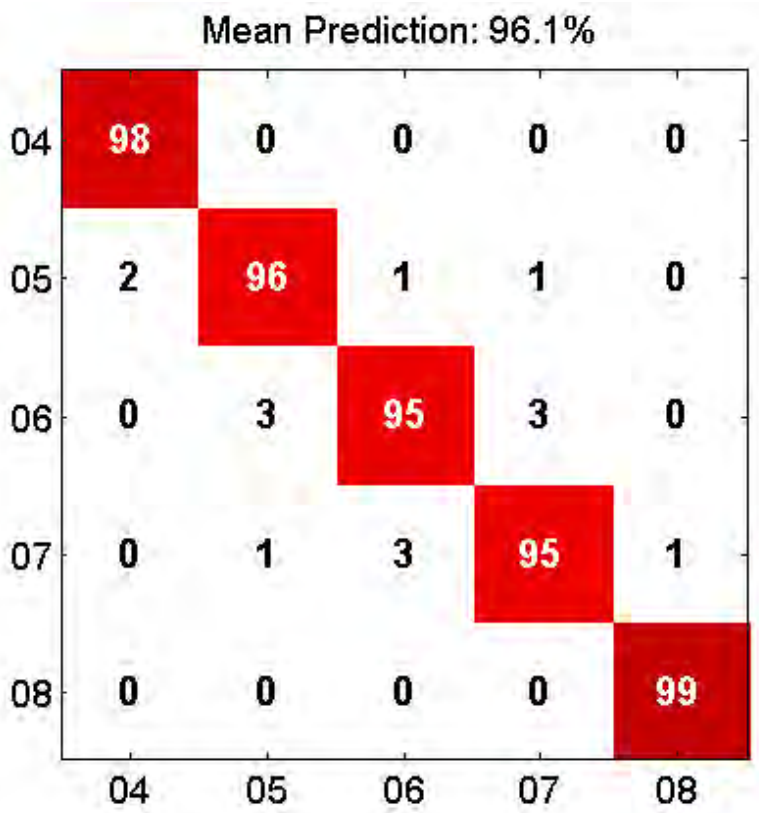
判断分类结果的精度

Avg.=92.6%



Vintages of German Wine (DLR)
德国葡萄酒年份分类的验证





Cooperation with **D**ienstleistungszentrum **L**ändlicher **R**aum Rheinland-Pfalz

All the samples kindly from DLR Rheinland-Pfalz, Riesling excluded

Results by NMR indicating problems confirmed by conventional AnthocyanAnalysis

Analysis Report Wine-Profiling™

Sample ID: 6136723

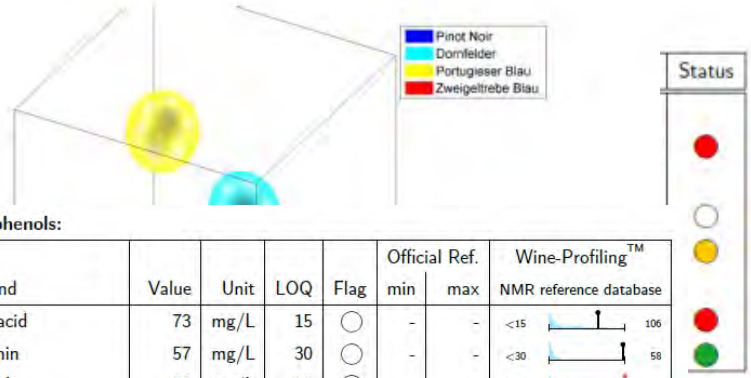
Additional Sample Information

Variety:	Portugieser Blau
Country:	Austria
Region:	undefined
Vintage:	2011
Type of Wine:	red

Classification Analysis

Model: Red Wine Variety (Germany/Austria)

Result: Declared variety *Portugieser Blau* is not consistent with classification result.



(Poly-)phenols:

Compound	Value	Unit	LOQ	Flag	Official Ref. min	Official Ref. max	Wine-Profiling™ NMR reference database
caftaric acid	73	mg/L	15	○	-	-	<15 … 106
epicatechin	57	mg/L	30	○	-	-	<30 … 58
gallic acid	56	mg/L	25	○	-	-	<25 … 32
shikimic acid	59	mg/L	20	○	-	-	<20 … 67
trigonelline	14	mg/L	10	○	-	-	<10 … 15

Stabilising Agents:

Compound	Value	Unit	LOD	Flag	Official Ref. min	Official Ref. max	Wine-Profiling™ NMR reference database
benzoic acid	<10	mg/L	10	○	-	LOD	not available
sorbic acid	<10	mg/L	10	○	-	200	not available
salicylic acid	<20	mg/L	20	○	-	LOD	not available

Untersuchungsbericht 6136723 - 2

Freigabe von: Herbert Witowski, Laborleitung am 07.05.13

Bezeichnung der Probe

Probeneingangsdatum 18.03.13 Behälterart: 0,75L Glasflasche
Untersuchungszeitraum vom 18.03.13 bis 07.05.13

WeinNr:F13 3966 LN 672/12 13,5 %vol [redacted]
Weinviertel Blauer Portugieser Blauer Portugieser N 100;00% Qualitätswein rot
Stillwein Stahltank trocken 3966

Anthocyanspektrum HPLC (QMP 11)

Parameter	Ergebnis	Einheit
Dp-3-Glucosid	7.73	%
Cy-3-Glucosid	n.n.	%
Pt-3-Glucosid	9.93	%
Po-3-Glucosid	5.25	%
Mv-3-Glucosid	73.14	%
Po-3-acyl	n.n.	%
Mv-3-acyl	n.n.	%
Po-3-cugl	0.39	%
Mv-3-cugl	3.57	%
Mv-3.5-Diglucosid	< 1.0	mg/l
Summe nicht acylierter Anthocyane	96.05	%
Summe acylierter Anthocyane	3.95	%
Summe acetylierter Anthocyane	0.00	%
Summe cumarylierter Anthocyane	3.95	%
Verhältnis acetylierter : cumarylierter Anthocyane	0.00	

Portugieser = 9-26%

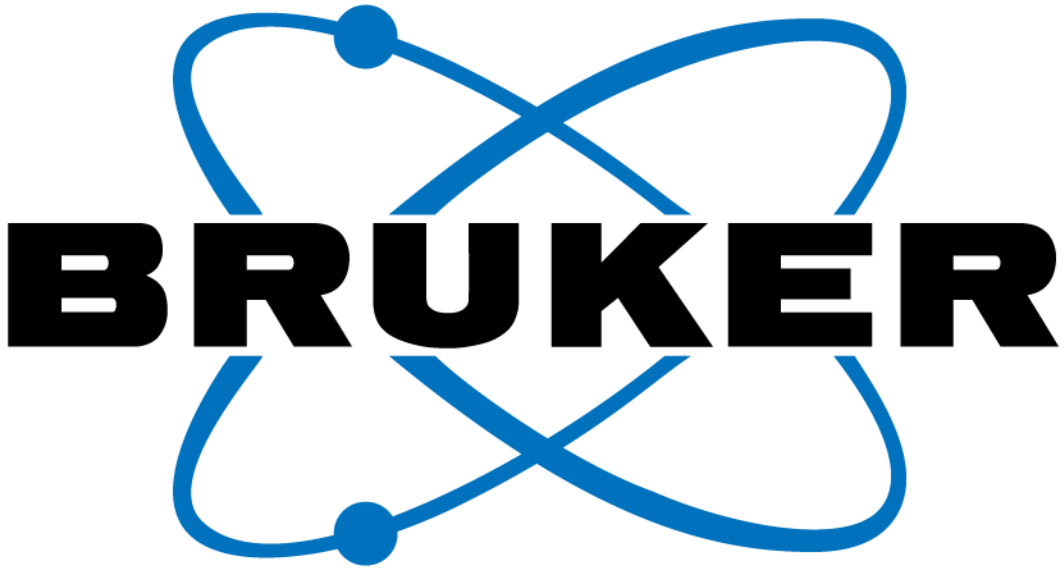
Portugieser 0,7 – 1,8%

Untersuchungsergebnis

Parameter	Ergebnis	Einheit	Methode	
Gesamtalkohol	110.0	g/l	1.1	
Gesamtalkohol	13.93	%vol	1.1	
Vorhandener Alkohol	107.3	g/l	2.9 NIR**/*	
Vorhandener Alkohol	13.59	%vol	ber.	
Gesamtextrakt	32.9	g/l	3.3	
Zuckerfreier Extrakt	27.2	g/l	3.3	
Glucose	0.7	g/l	4.5**	BG: 0,1g/l
Fructose	4.8	g/l	4.5**	BG: 0,2g/l
Zucker vor Inversion	5.5	g/l	4.5** ber.	
Zucker nach Inversion	5.7	g/l	4.5** ber.	
Gesamtsäure ber. als Weinsäure	4.9	g/l	5.3**	
Freie SO2	< 5	mg/l	6.3phot. */**	
Gesamte SO2	37	mg/l	7.7 phot */**	
Relative Dichte	0.9951		8.4**	

Conclusions

- NMR is an efficient high throughput tool for food screening under full automation.
  基于NMR的食品筛选是一种高通量、全自动化的有效分析手段
- One measurement, but many parameters combines targeted and non-targeted screening .
  通过一次检测（约15分钟）可以获得大量的参数，包含靶向和非靶向的分析结果
- NMR is now well established in fruit juice, wine and honey screening; the developed procedure can be taken as a model for many other materials.
  目前筛选平台可应用于果汁、葡萄酒和蜂蜜。这个成熟的平台还可以扩展到其他类型的食品
- Before a screening is started, one must define and check SOPs. Decision made here often cannot be corrected later.
  在对其他类型的食品建立新的筛选方案之初应该确定好SOP
- Extensive validation guarantees correct results.
  对结果进行广泛的验证，确保结果的正确性
- We offer SOPs for juice, wine, honey, edible oils and coffee. Biofluid and tissue SOPs are also available.
  我们提供果汁、葡萄酒、蜂蜜、食用油和咖啡的SOP，也提供临床上的体液和组织的SOP



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Q & A

是否有问题？

请您在 Q&A 面板中提交您的问题

我们做的如何？

当您退出webinar的时候，请填写您对此次webinar的评价，我们非常感谢您的反馈。

Thank you!

Wenxin.xu@bruker.com
Lu.shan@bruker.com