# Agilent Approaches for Amino Acid Analysis

Mark Powell
Columns and Supplies Technical Support
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Agilent InfinityLab

Agilent

# Outline

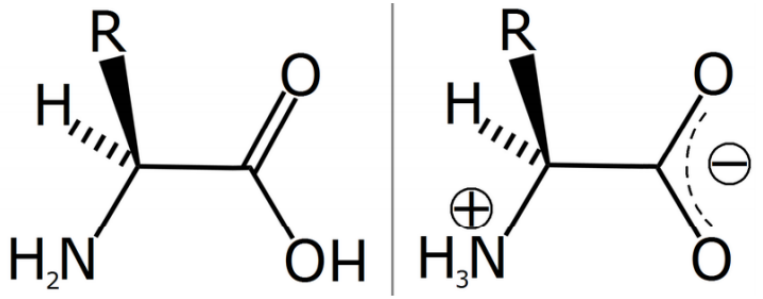
- History of Amino Acid Analysis at Agilent
- Reverse phase LC/UV analysis of derivatized amino acids
- HILIC LC/MS of underivatized amino acids and other metabolites
- Ion pairing analysis of underivatized amino acids
- Chiral analysis of amino acids

# Why is Amino acid analysis important?

- Important for protein and peptide identification and quantitation
- Part of reverse-phase characterization in biopharma
- Important for monitoring cell culture media
- Used for the analysis of metabolic intermediates
- Flavor analysis

# Some Basics

- Detection by UV or FL of amino acids is improved by derivatization
  - OPA/FMOC, Ninhydrin, Dansyl chloride, and PITC are common reagents used
- Derivatization can be done precolumn or post column
  - OPA/FMOC, Dansyl chloride, and PITC are common reagents used for precolumn
  - Ninhydrin is common for post column methods
- The are multiple methods to perform amino acid analysis:
  - GC, CE, HPAE-PAD
  - LC/UV/FL, LC/MS



# Precolumn and Postcolumn Derivatization

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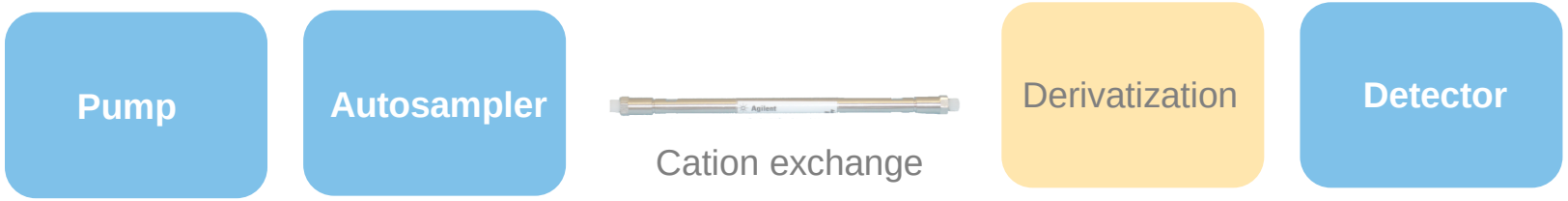
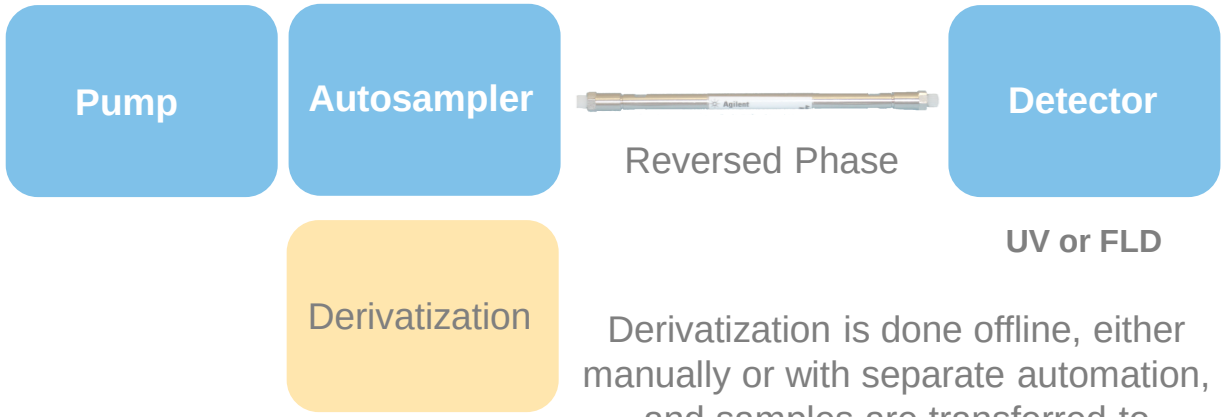
## Postcolumn derivatization – the historic Gold Standard of dedicated Amino Acid Analyzers

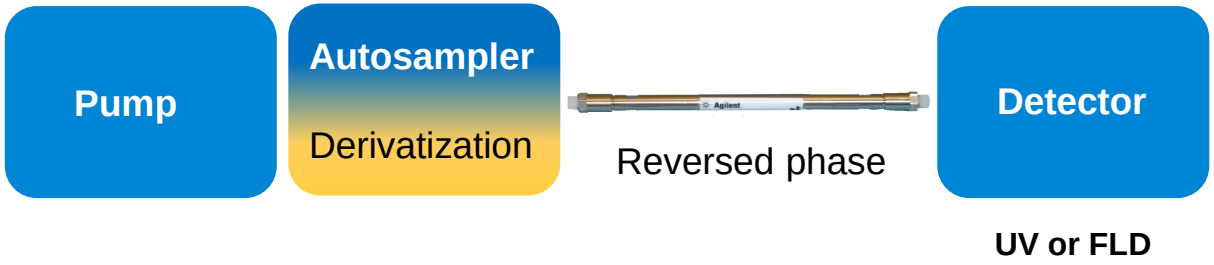



## Precolumn derivatization – offline:




Derivatization is done offline, either manually or with separate automation, and samples are transferred to autosampler

## Precolumn derivatization – online:




Derivatization is done online, in the autosampler, which eliminates errors associated with manual sample handling and produces highly consistent, reproducible results



Agilent

# History of AAA at Agilent

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## AminoQuant (c. 1990)

- Extensive guide for HP 1090
- Hypersil AA ODS column

## ZORBAX Eclipse AAA (c. 2000)

- User guide pub no. 5980-3088EN

## ZORBAX Eclipse Plus C18 (c. 2010)

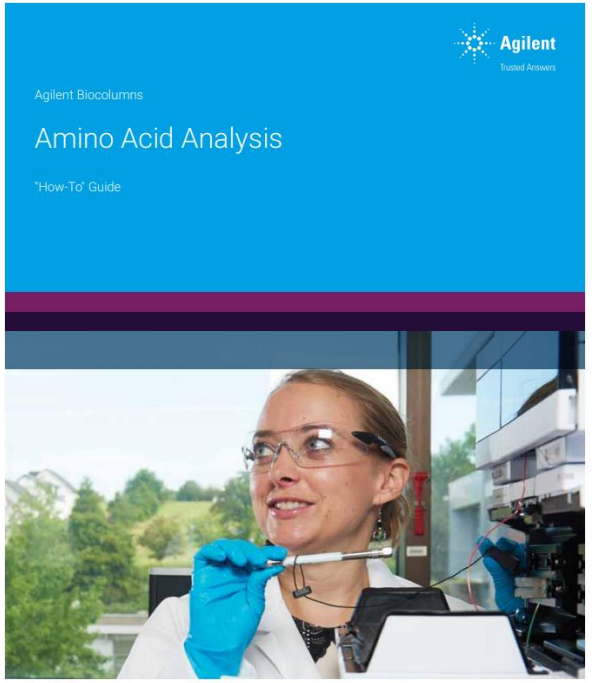
- *Improved Amino Acid Methods using Agilent ZORBAX Eclipse Plus C18 Columns for a Variety of Agilent LC Instrumentation and Separation Goals*
- Pub no. 5990-4547EN

## AdvanceBio AAA (c. 2017)

- Amino Acid Analysis “How-To” Guide
- Poroshell particle
- Pub no. 5991-7694EN

## AdvanceBio MS Spent Media (c. 2018)

- HILIC, MS compatible
- Pub no. 5991-8816EN

# Agilent AdvanceBio AAA

## Previous Agilent AAA method

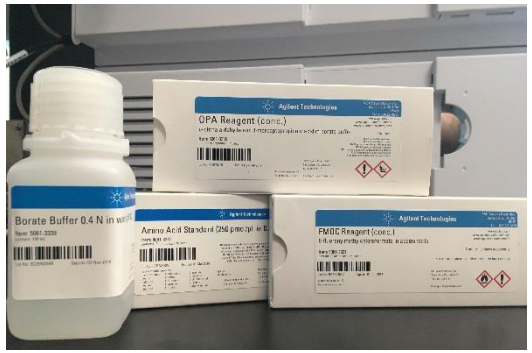
Agilent has a well-established solution for Amino Acid Analysis

- Based on automated precolumn derivatization capabilities of Agilent autosamplers
- Uses ZORBAX Eclipse AAA column
- Well established method using reagents and standards from Agilent

## What's updated?

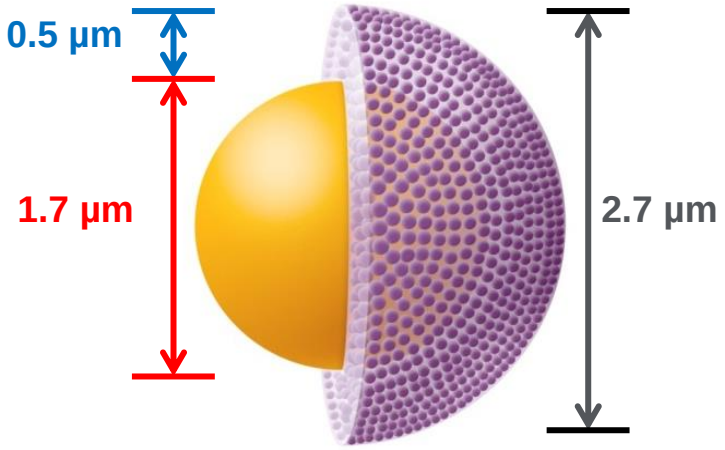
- Reagents conveniently kitted together under a single part number
- Introduced an HPH chemistry on a Poroshell particle for improved column lifetime
  - Traditional silica columns dissolve above neutral pH, but HPH chemistry stabilizes column
  - AA derivatization and separation are most efficient at higher pH
  - Poroshell particle: high efficiency, fast separations, rugged

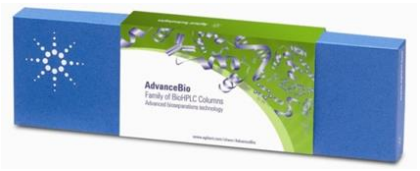
# The Agilent Amino Acid Analysis Solution




Ready to use AdvanceBio AAA kit (standards and reagents)

All Agilent LC systems, including Infinity II systems




AdvanceBio AAA columns
Poroshell particles
Fast and rugged

# Automated Derivatization in the Autosampler

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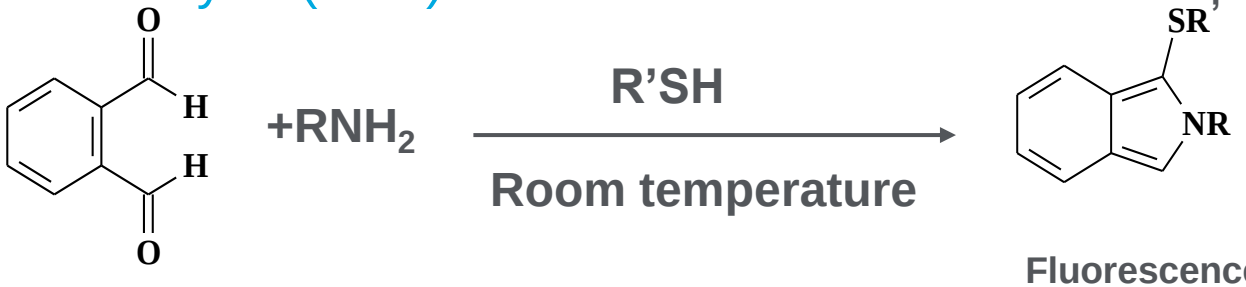
1. Allows visualization by UV or FL
2. Helps retain very polar compounds

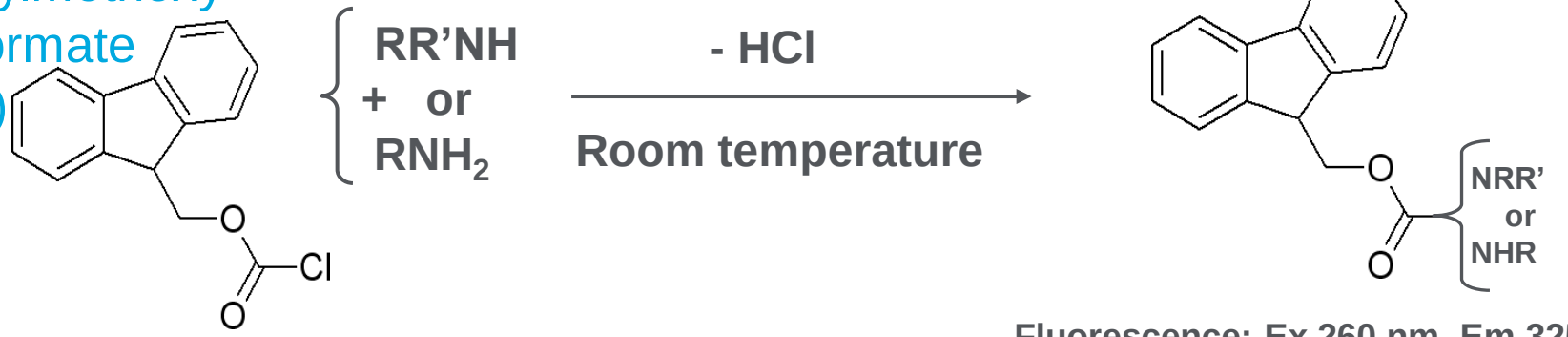
## ortho-Phthalaldehyde (OPA)




**Nonfluorescent**
**Does not absorb at 338 nm**

**Fluorescence: Ex 340 nm, Em 450 nm**
**DAD: 338, 10 nm; Ref. 390, 20 nm**

## Fluorenylmethoxy chloroformate (FMOC)



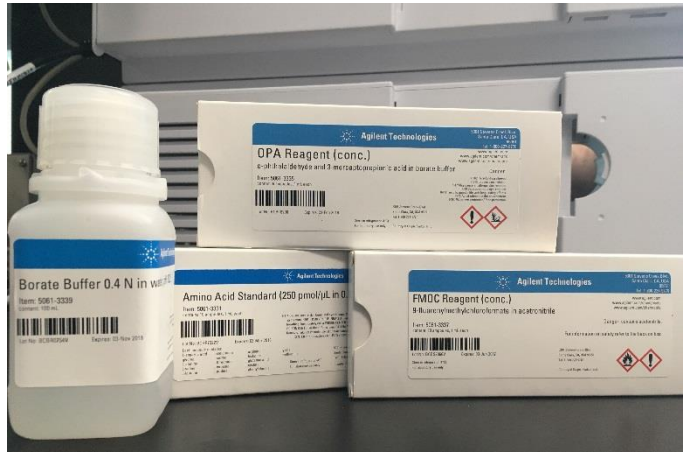

**Fluorescent**
**Absorbs at 262 nm and fluoresces at 324 nm**

**Fluorescence: Ex 260 nm, Em 325 nm**
**DAD: 262, 16 nm; Ref. 324,8 nm**

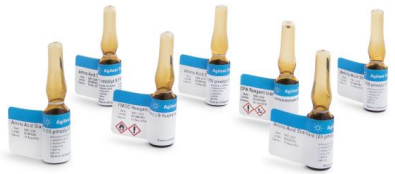
Optimal pH for reaction with AA: ~10.0

# AdvanceBio AAA Reagent Kit

| Material | Part Number |
|---|---|
| Borate buffer, 100 mL | 5061-3339 |
| FMOC reagent, 10 ampoules, 1 mL each | 5061-3337 |
| OPA reagent/3-mercaptopropionic acid, 6 ampoules, 10 mg/mL | 5061-3335 |
| Dithiodipropionic acid (DTDPA) | 5062-2479 |
| AA standards, 1 nmol, 10/pk | 5061-3330 |
| AA standards, 250 pmol, 10/pk | 5061-3331 |
| AA standards, 100 pmol, 10/pk | 5061-3332 |
| AA standards, 25 pmol, 10/pk | 5061-3333 |
| AA standards, 10 pmol, 10/pk | 5061-3334 |
| Amino acids supplement kit, 1 g each of norvaline, sarcosine, asparagine, glutamine, tryptophan, and 4-hydroxyproline | 5062-2478 |




Order components individually, or together as part of a kit with a single part number:
**5190-9426**



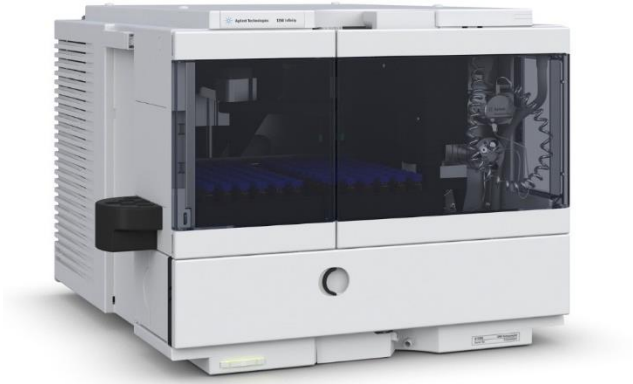
# Online Derivatization/Injection Program

- Draw 2.5 µL from borate vial (Agilent p/n 5061-3339)
- Draw 1.0 µL from sample vial
- Mix 3.5 µL in wash port five times
- Wait 0.2 min
- Draw 0.5 µL from OPA vial (Agilent p/n 5061-3335)
- Mix 4.0 µL in wash port 10 times default speed
- Draw 0.4 µL from FMOC vial (Agilent p/n 5061-3337)
- Mix 4.4 µL in wash port 10 times default speed
- Draw 32 µL from injection diluent vial
- Mix 20 µL in wash port eight times
- Inject
- Wait 0.1 min
- Valve bypass

1290 Infinity II Multisampler

Method can be programmed into any Agilent autosampler:

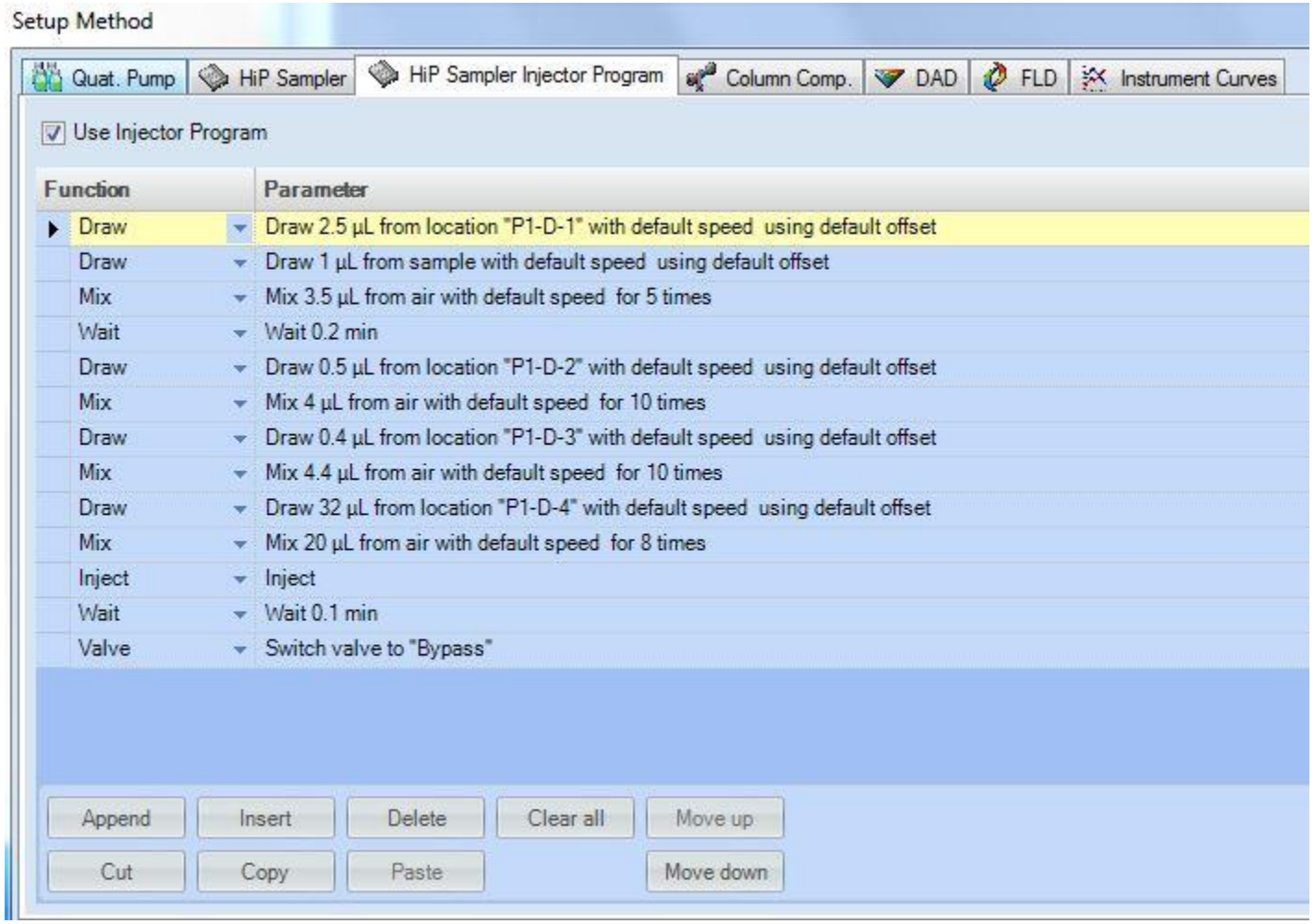
- Eliminates manual labor and variability
- Enables highly precise data



1260 Infinity II Vialsampler

# Online Derivatization/Injection Program

OpenLab ChemStation C.01




1290 Infinity II Multisampler

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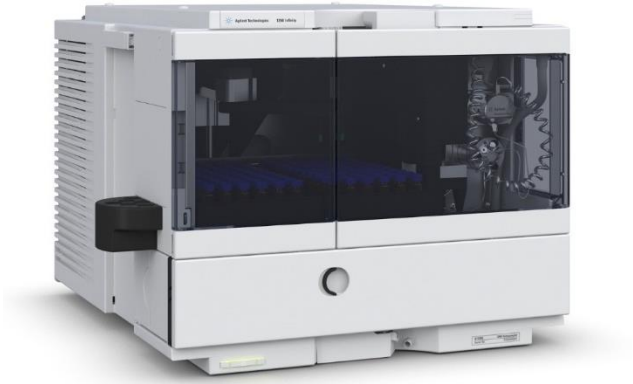


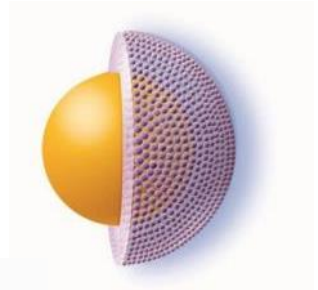
1260 Infinity II Vialsampler

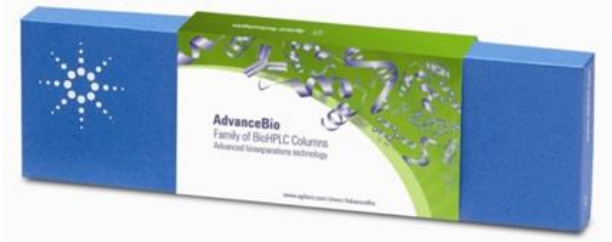
# Robust Columns for AAA

*A robust, high efficiency Fast LC column with resistance to elevated pH and temperature offering users performance comparable to that of sub-2 µm alternatives but with up to 50% less backpressure.*

Utilizes a proprietary technology for particle synthesis


- 2.7 µm particles, 100 Å pore size
- Two dimensions available: 3.0 x 100 mm, 4.6 x 100 mm
  - Guard columns also available in each id
- Each individual column is tested for efficiency
- Each batch is tested with amino acid standards to ensure performance





# AdvanceBio AAA columns

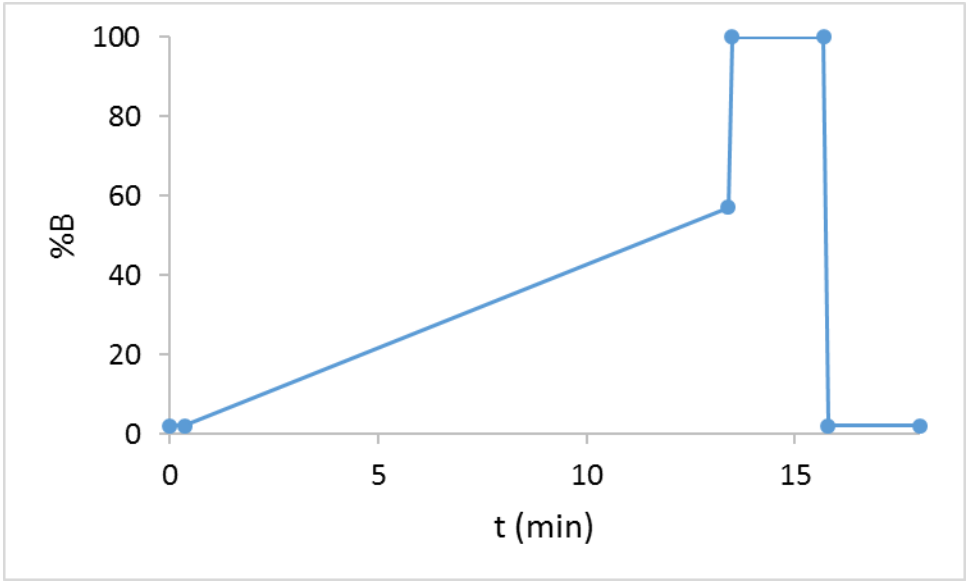
| Bonded Phase | id (mm) | Particle size (µm) | Length (mm) | Pore Size (Å) | Temp Limit | pH Range | Endcapping | Part Number |
|---|---|---|---|---|---|---|---|---|
| C18 | 3.0 | 2.7 | 100 | 100 | 65 °C | 3.0 – 11.0 | Double | 695975-322 |
| C18 | 4.6 | 2.7 | 100 | 100 | 65 °C | 3.0 – 11.0 | Double | 655950-802 |
| C18 | 3.0 | 2.7 | 5 | 100 | 65 °C | 3.0 – 11.0 | Double | 823750-946 (3/pk guards) |
| C18 | 4.6 | 2.7 | 5 | 100 | 65 °C | 3.0 – 11.0 | Double | 820750-931 (3/pk guards) |

# Derivatization Kit – How Many Samples?

- Once opened, the OPA and FMOC ampoules need to be used in about a week
- Typically, the OPA and FMOC reagents are opened on Monday and used for five days
- I would estimate 50 samples a day is easily accomplished, so 250 samples in a week
- The OPA comes as a pack of six ampoules (so let's estimate it's good for roughly 1500 samples)
- The FMOC comes as a pack of 10 (so roughly 2500 samples)


# Chromatographic Method

| AdvanceBio Amino Acid Analysis | |
|---|---|
| **Column** | Agilent AdvanceBio Amino Acid Analysis |
| **Column Temp** | 40 °C |
| **Mobile Phase** | A = 10 mM $Na_2HPO_4$ and 10 mM $Na_2B_4O_7$, pH 8.2<br>B = Acetonitrile:methanol:water (45:45:10, v:v:v) |
| **Flow Rate** | 1.5 mL/min for 4.6 mm i.d.<br>0.62 mL/min for 3.0 mm i.d. |
| **Gradient Program** | **Time** **% B**<br>0 2<br>0.35 2<br>13.4 57<br>13.5 100<br>15.7 100<br>15.8 2<br>18 stop |
| **Injection volume** | 1 µL, with 7s needle wash at wash port |
| **Detection** | UV – 338 and 262 nm<br>FLD – Ex λ 340 nm, Em λ 450 nm; Ex λ 260 nm, Em λ 325 nm |



# Order of Elution for OPA and FMOC derivatives

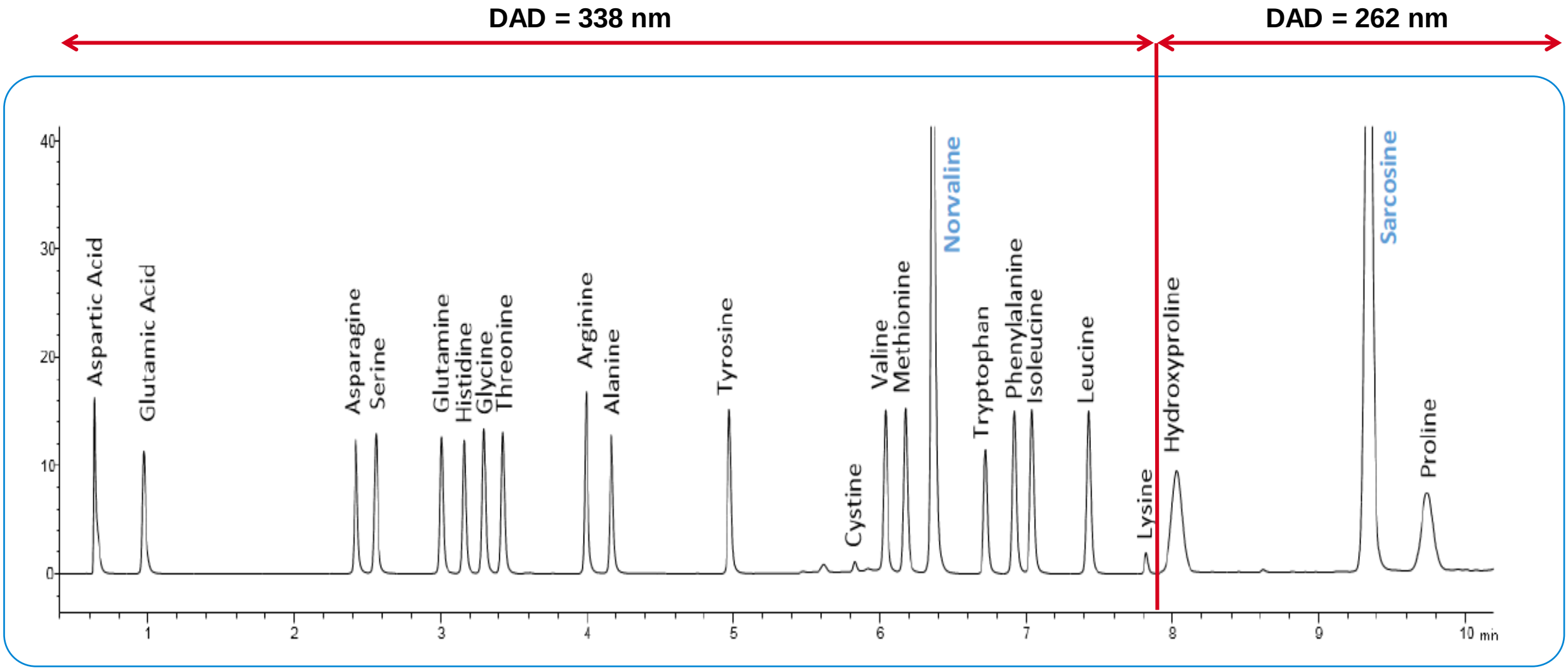
| Peak # | AA Name | AA Abbreviation | Derivative Type |
|---|---|---|---|
| 1 | Aspartic Acid | Asp | OPA |
| 2 | Glutamic Acid | Glu | OPA |
| 3 | Asparagine | Asn | OPA |
| 4 | Serine | Ser | OPA |
| 5 | Glutamine | Gln | OPA |
| 6 | Histidine | His | OPA |
| 7 | Glycine | Gly | OPA |
| 8 | Threonine | Thr | OPA |
| 9 | Arginine | Arg | OPA |
| 10 | Alanine | Ala | OPA |
| 11 | Tyrosine | Tyr | OPA |
| 12 | Cysteine | Cys-Cys | OPA |
| 13 | Valine | Val | OPA |
| 14 | Methionine | Met | OPA |
| 15 | Norvaline | Nva | OPA |
| 16 | Tryptophan | Trp | OPA |
| 17 | Phenylalanine | Phe | OPA |
| 18 | Isoleucine | Ile | OPA |
| 19 | Leucine | Leu | OPA |
| 20 | Lysine | Lys | OPA |
| 21 | Hydroxyproline | Hyp | FMOC |
| 22 | Sarcosine (IS) | Sar | FMOC |
| 23 | Proline | Pro | FMOC |

**Primary AA**

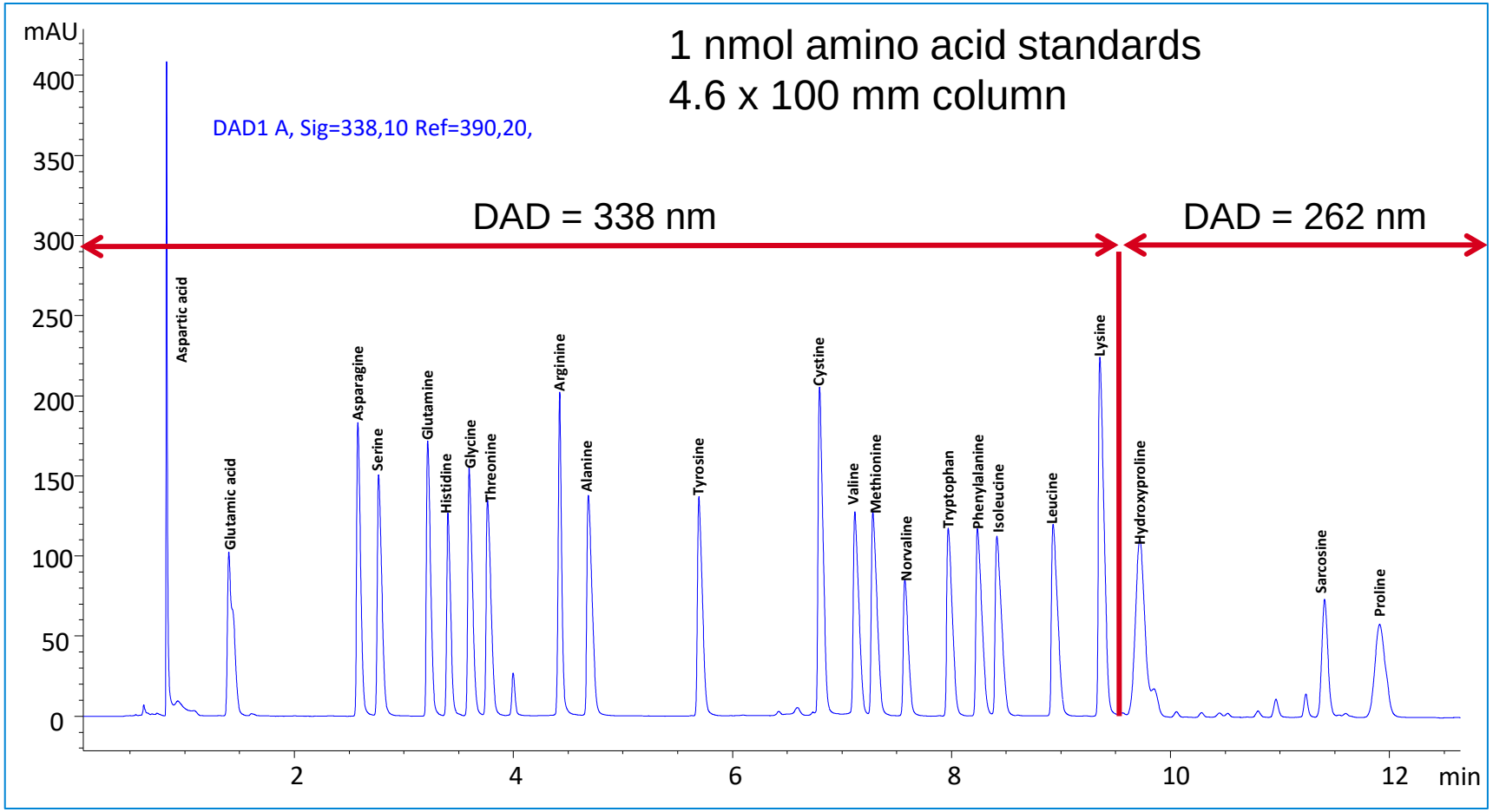
**Secondary AA**

# Fast and Rugged Amino Acids Separation

Agilent InfinityLab



# Reproducible Separations

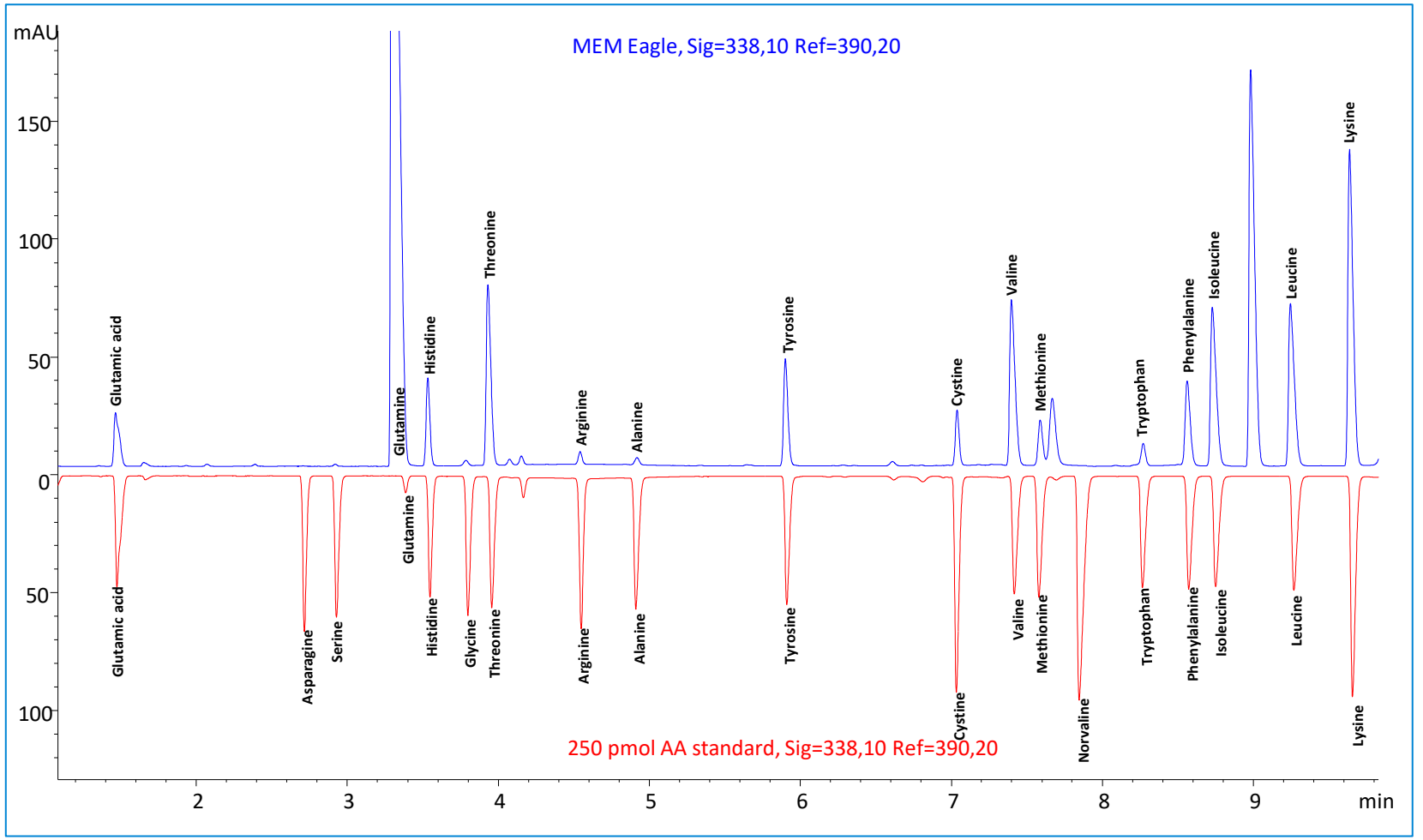



| Amino Acids | RT RSD (%) | Area RSD (%) |
|---|---|---|
| 1. Aspartic acid | 1.270 | 1.066 |
| 2. Glutamic acid | 0.973 | 1.85 |
| 3. Asparagine | 0.605 | 1.79 |
| 4. Serine | 0.629 | 1.82 |
| 5. Glutamine | 0.470 | 1.56 |
| 6. Histidine | 0.430 | 1.22 |
| 7. Glycine | 0.477 | 1.92 |
| 8. Threonine | 0.440 | 1.95 |
| 9. Arginine | 0.251 | 2.15 |
| 10. Alanine | 0.280 | 3.06 |
| 11. Tyrosine | 0.128 | 1.65 |
| 12. Cystine | 0.067 | 1.9 |
| 13. Valine | 0.084 | 2.47 |
| 14. Methionine | 0.073 | 1.82 |
| 15. Norvaline | 0.073 | 1.72 |
| 16. Tryptophan | 0.054 | 1.57 |
| 17. Phenylalanine | 0.051 | 1.66 |
| 18. Isoleucine | 0.047 | 1.72 |
| 19. Leucine | 0.03 | 1.7 |
| 20. Lysine | 0.028 | 1.66 |
| 21. Hydroxyproline | 0.021 | 4.13 |
| 22. Sarcosine | 0.026 | 1.15 |
| 23. Proline | 0.021 | 4.36 |

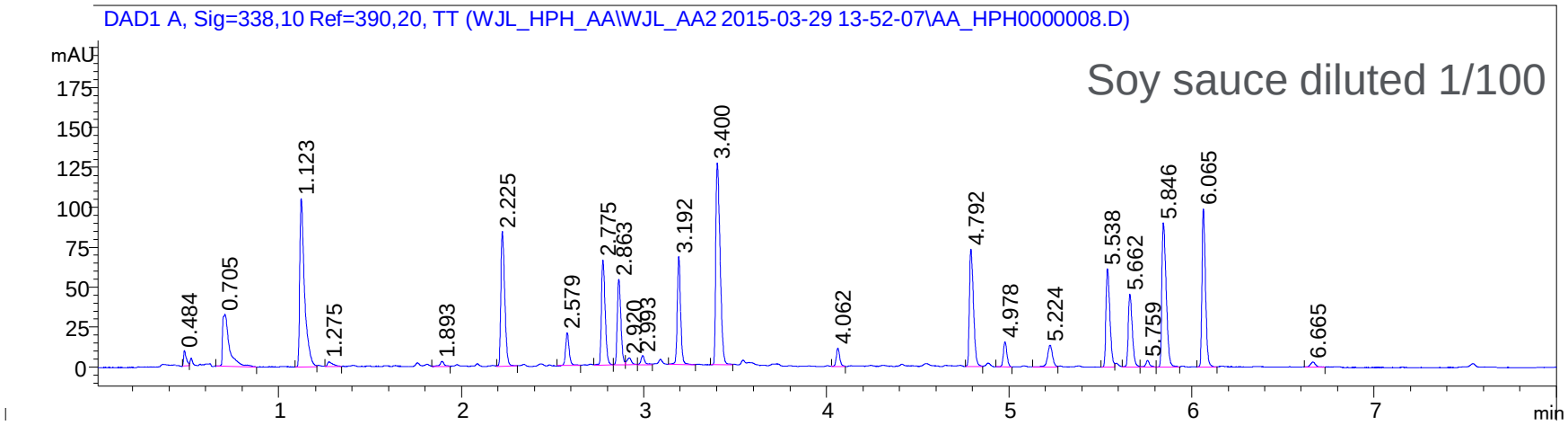
- Retention time %RSD mostly under 1%
- Peak area %RSD mostly under 3%

# AAA of Cell Culture Media – MEM

L-Arginine, L-Cystine, L-Glutamine, L-Histidine, L-Isoleucine, L- Leucine, L-Lysine, L-Methionine, L- Phenylalanine, L-Threonine, L-Tryptophan, L- Tyrosine, L-Valine, L-Glutamic acid

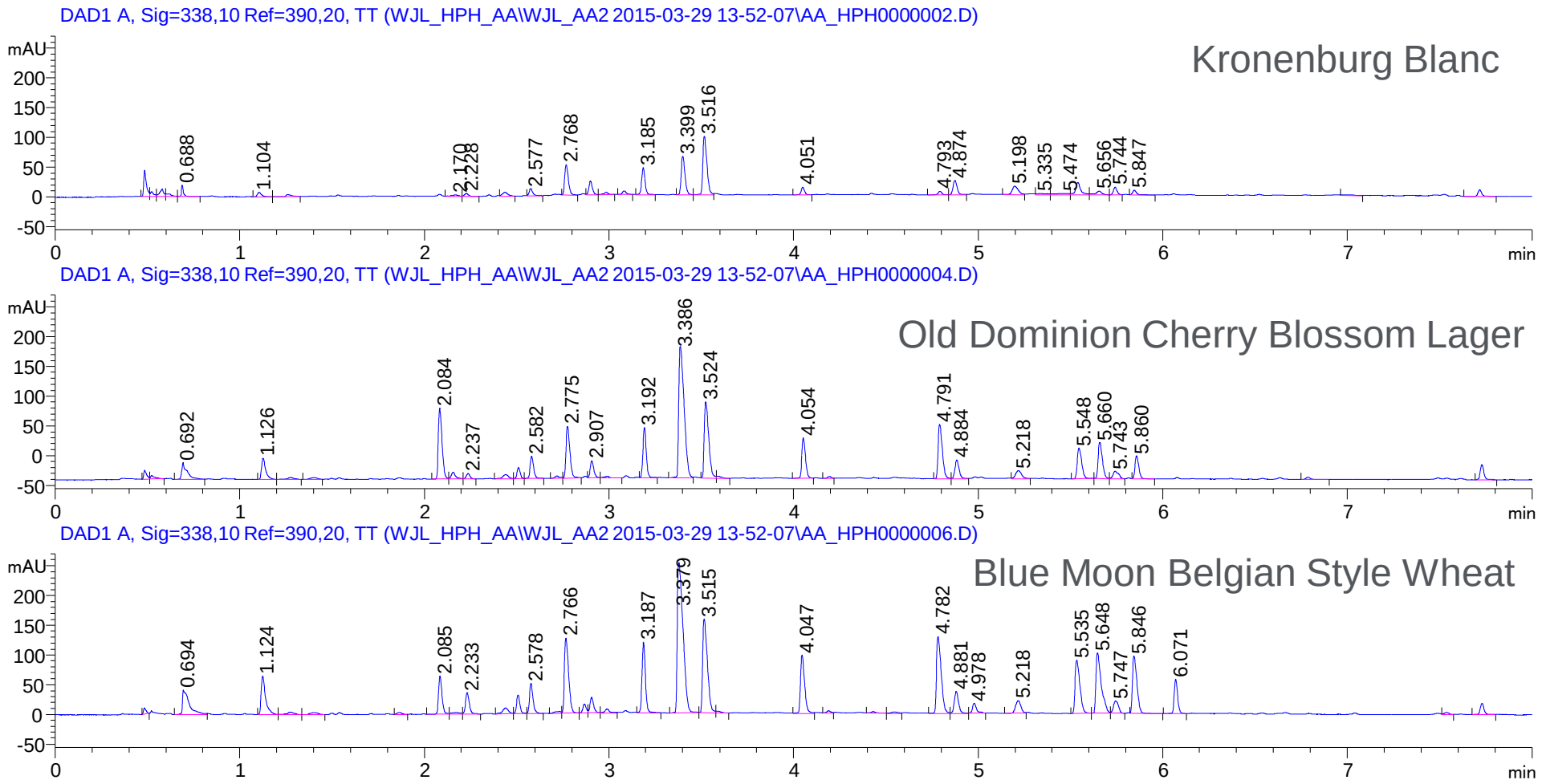


# Amino Acid Analysis in Fermentation Applications




Many other foods (such as soy sauce) and pharmaceuticals that are produced using fermentation processes are monitored by AAA

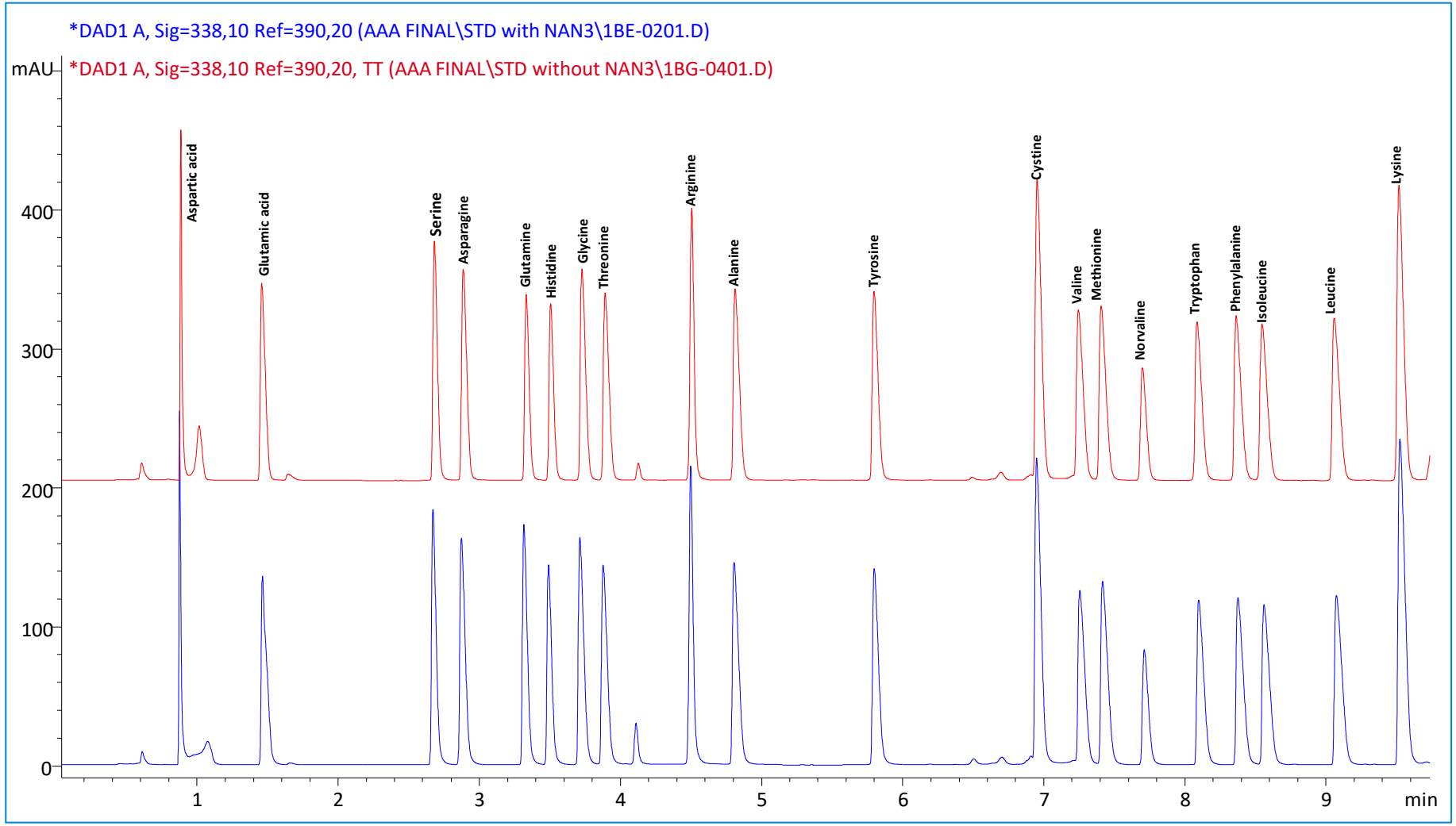
# Amino Acids Analysis for Batch Comparison




Quantity and diversity of amino acids is evident. Can be used to monitor and compare batches.

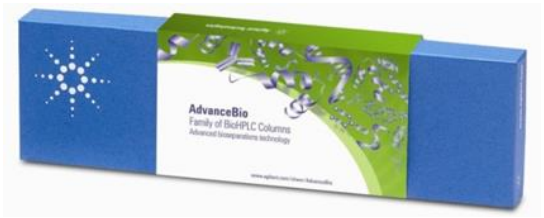
# Elution Profile With and Without Sodium Azide

- Historically $NaN_3$ has been added to aqueous mobile phase to reduce bacterial growth
- $NaN_3$ is highly toxic
- No effect on the separation
- Highly recommend filtering mobile phases (0.45 or 0.2 µm) to reduce bacterial growth



# Tips and Tricks – Maintenance

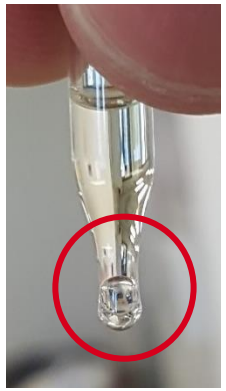
- Replace derivatization reagent, borate buffer, amino acid standard daily
- Recalibrate for retention times and response factors weekly
- Check column and guard column performance by following specs (Rs for two pairs of AA)
- Replace mobile phase A and B with fresh ones every other day
- Exchange guard column if high backpressure develops
- Avoid using max mixing speed during sample derivatization
  - The max speed on newer LCs is much faster than older LCs (1100/1200)
  - Can cause excessive wear on the autosampler

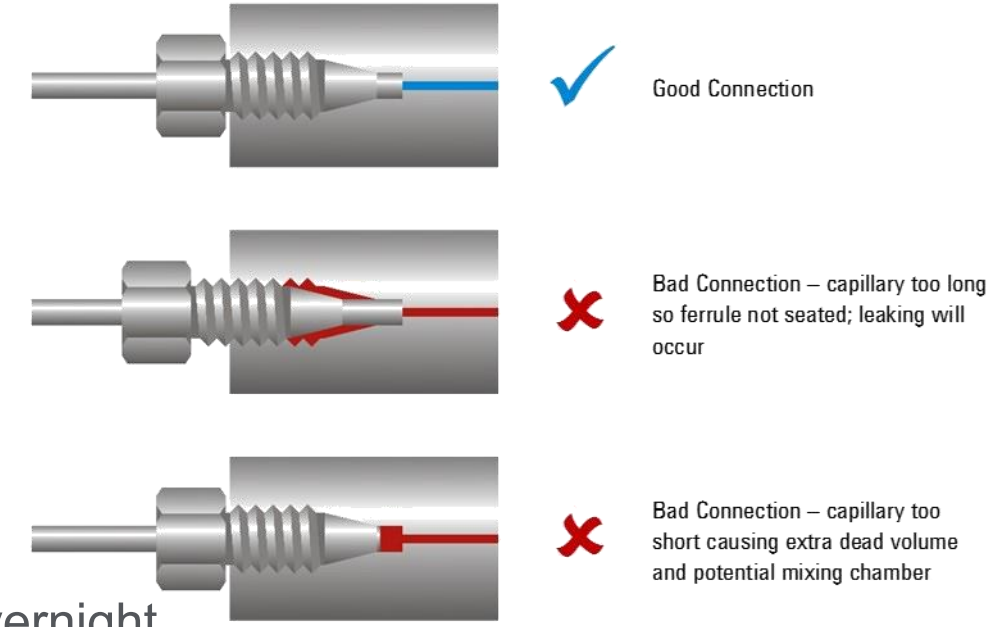



# Tips and Tricks – Troubleshooting

## Poor chromatographic resolution?

- Cell culture media does not require any sample preparation, however appropriate dilutions must be made to suit detector response
- In all cases, use the low-volume heat exchanger with short red tubing to minimize extra column volume
- Ensure proper connections
- Damaged guard or analytical column





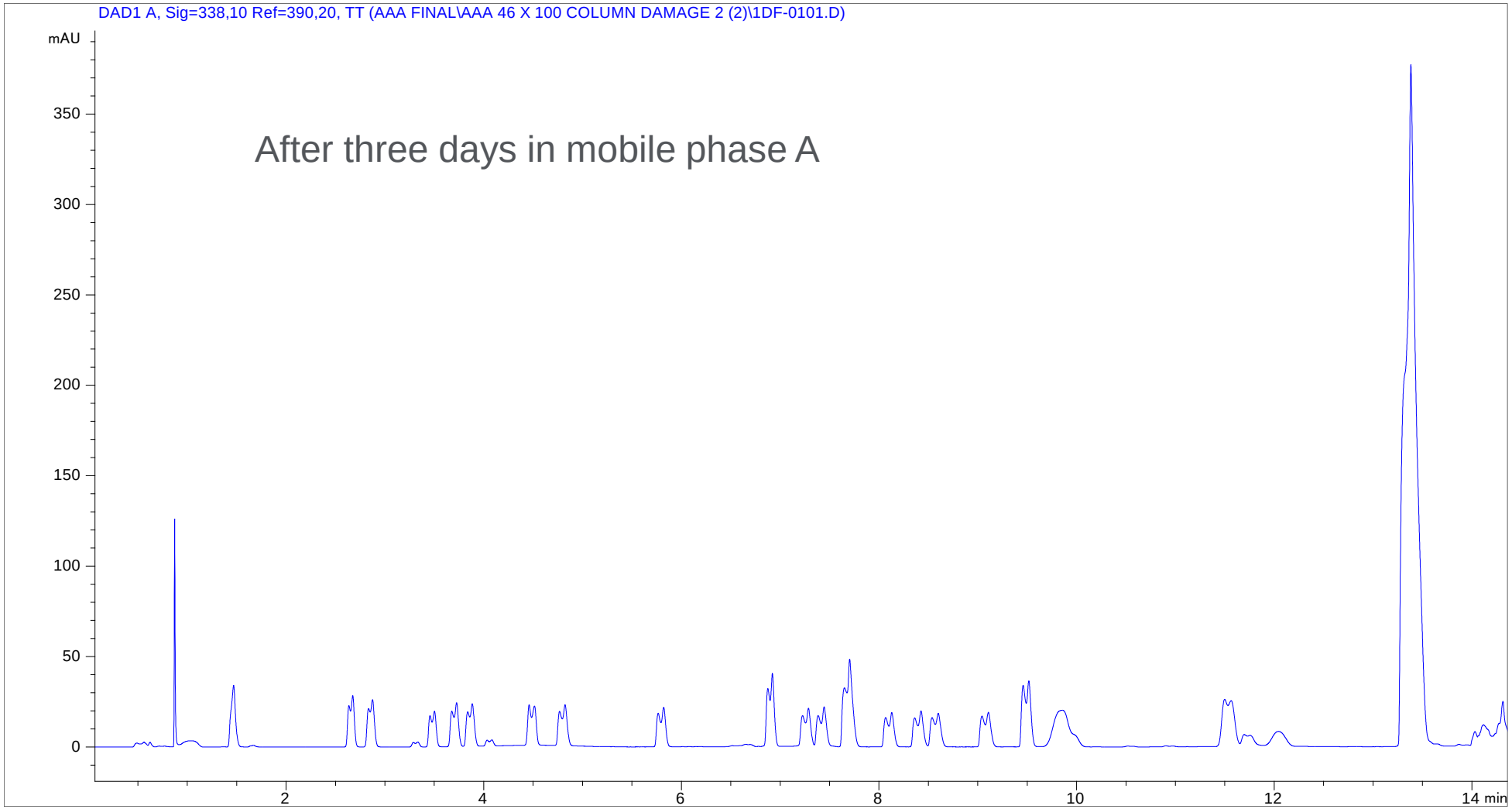

## Low intensity chromatogram?

- OPA/FMOC reagent deteriorated
- Air bubble in vial insert

## Column storage?

- Never leave the column in mobile phase A even if it's just overnight
- For short term always store the column in mobile phase B
- For long term, store column in 50/50 acetonitrile/$H_2O$

# Damaged Column



# Outline

- History of Amino Acid Analysis at Agilent
- Reverse phase LC/UV analysis of derivatized amino acids
- HILIC LC/MS of underivatized amino acids and other metabolites
  - **H**ydrophobic **I**nteraction **LI**quid **C**hromatography
  - Stationary phase is more polar than reversed phase
  - Acetonitrile is weak solvent, water is strong solvent (and typically a volatile buffer)
  - AdvanceBio MS Spent Media column
- Ion pairing analysis of underivatized amino acids
- Chiral analysis of amino acids

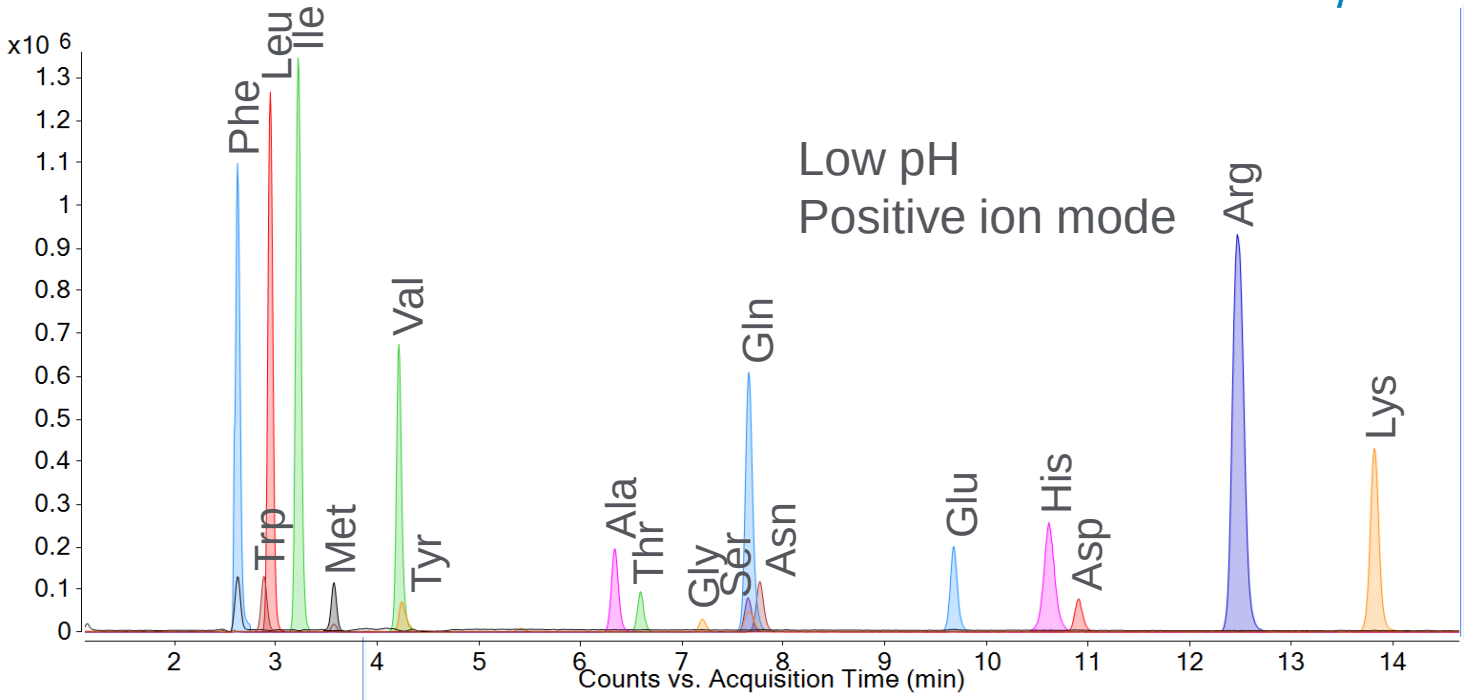
# Method – Amino Acids by HILIC

| Suggested Starting Conditions – LC/MS | |
|---|---|
| **Column** | Agilent AdvanceBio MS Spent Media, 2.1 x 100 mm, p/n 675775-901 |
| **Column Temp** | 30 °C |
| **Mobile Phase** | **Low pH, Positive Ion Mode MS Detection:**<br>A = 10% 200 mM ammonium formate in water pH 3, 90% water<br>B = 10% 200 mM ammonium formate in water pH 3, 90% acetonitrile<br>*Final salt concentration is 20 mM.*<br><br>**High pH, Negative Ion Mode MS Detection:**<br>A = 10% 100 mM ammonium acetate in water pH 9, 90% water<br>B = 10% 100 mM ammonium acetate in water pH 9, 90% acetonitrile<br>*Final salt concentration is 10 mM.*<br><br>*We recommend preparing mobile phases from a concentrated buffer stock to ensure robust and consistent mobile phases.* |
| **Flow Rate** | 0.5 mL/min |
| **Gradient Program** | **Time** **% B**<br>0 100<br>15 80<br>15.5 100<br>20 100 |
| **Sample** | Cell culture media, diluted 5-fold with Mobile Phase B |
| **Detection** | Agilent 6230 TOF LC/MS |

# Standard Amino Acids

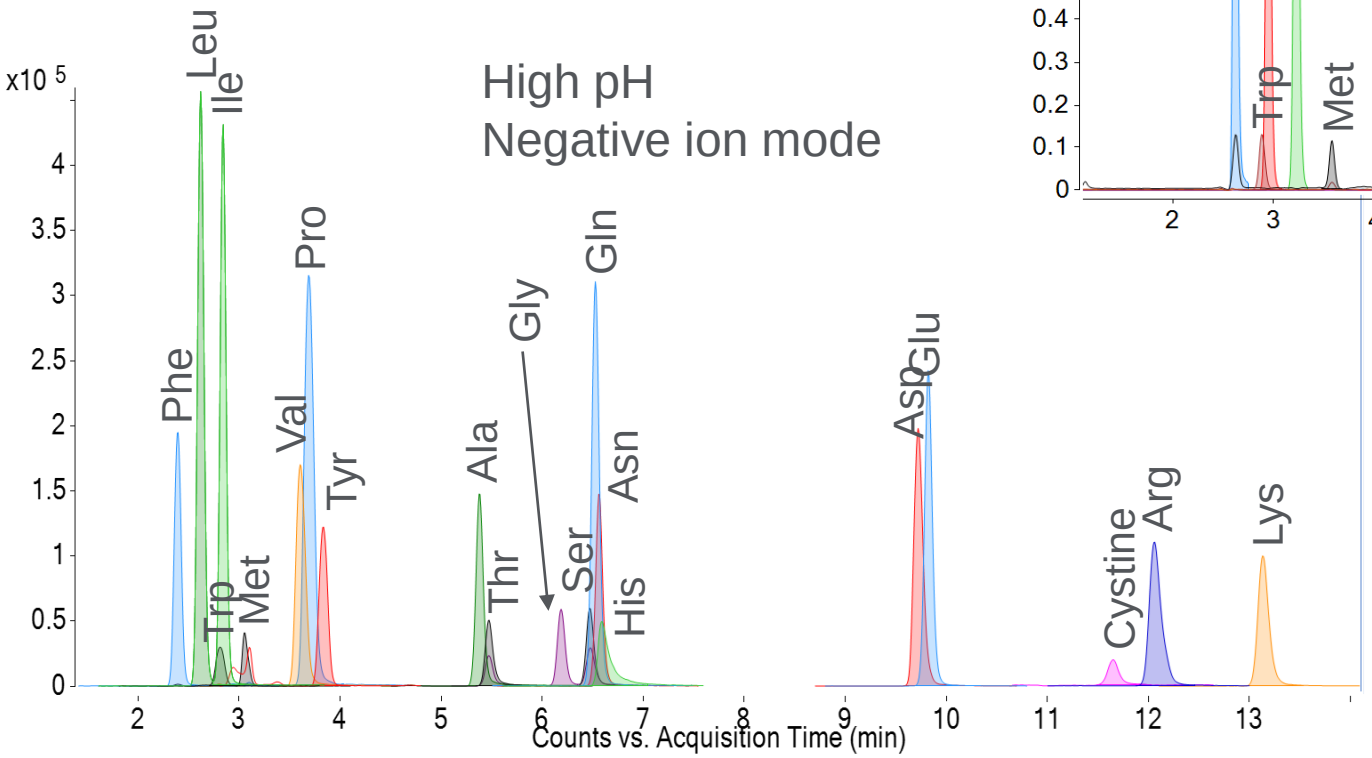
Agilent InfinityLab

Amino acids can be analyzed in either positive or negative ion mode since they are zwitterionic.



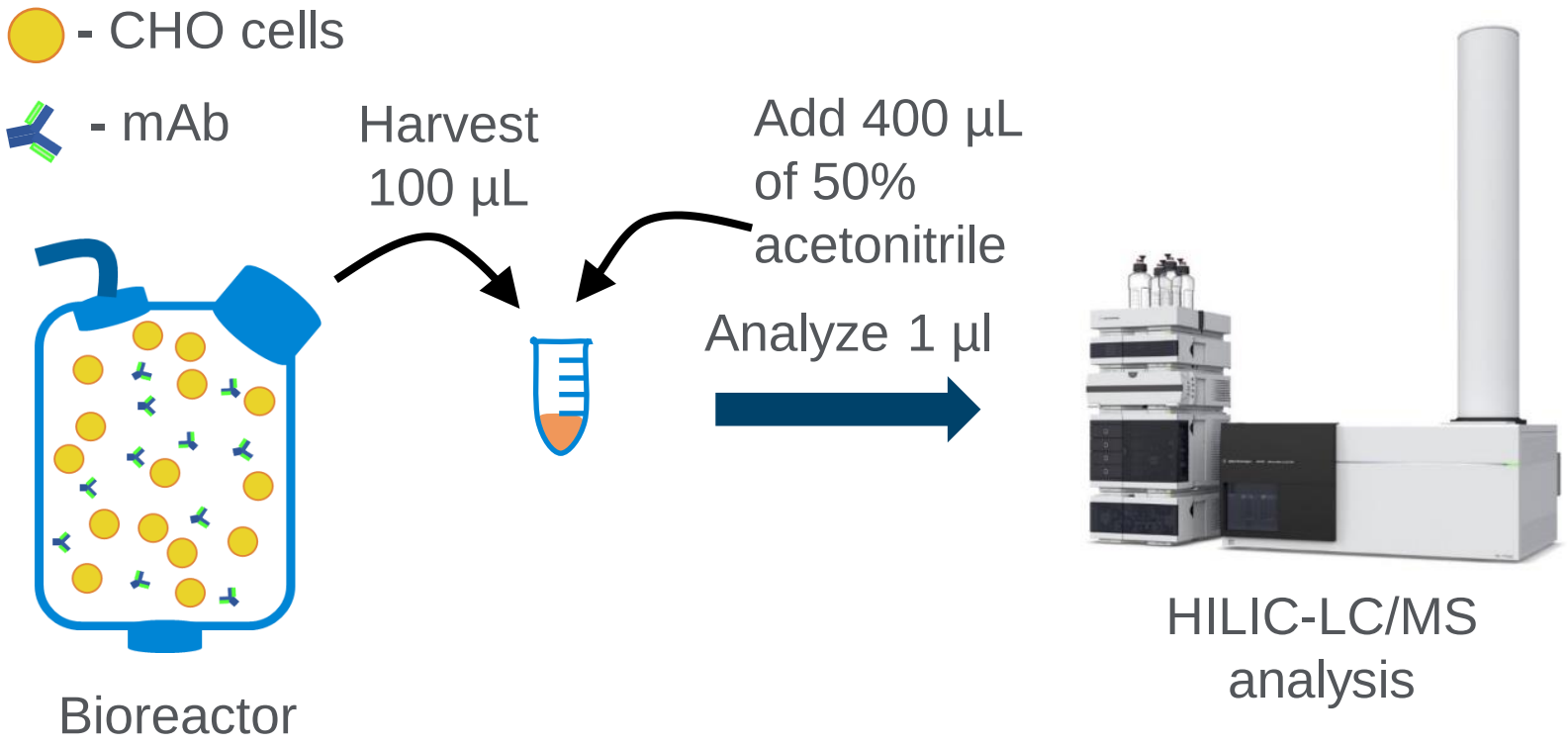

Low pH
Positive ion mode

High pH
Negative ion mode

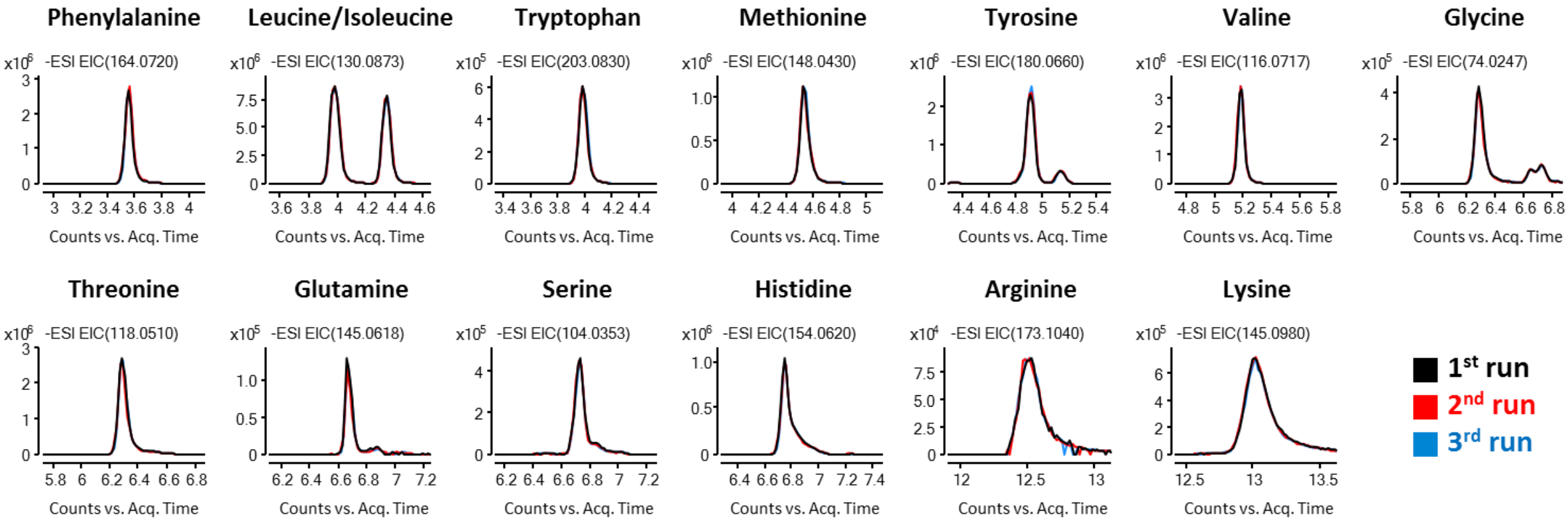



Generally positive ion mode will be more sensitive, though which mode is used may also depend on what other analytes need to be monitored simultaneously.

# A Fast and Simple Approach to Profiling Cell Culture Media

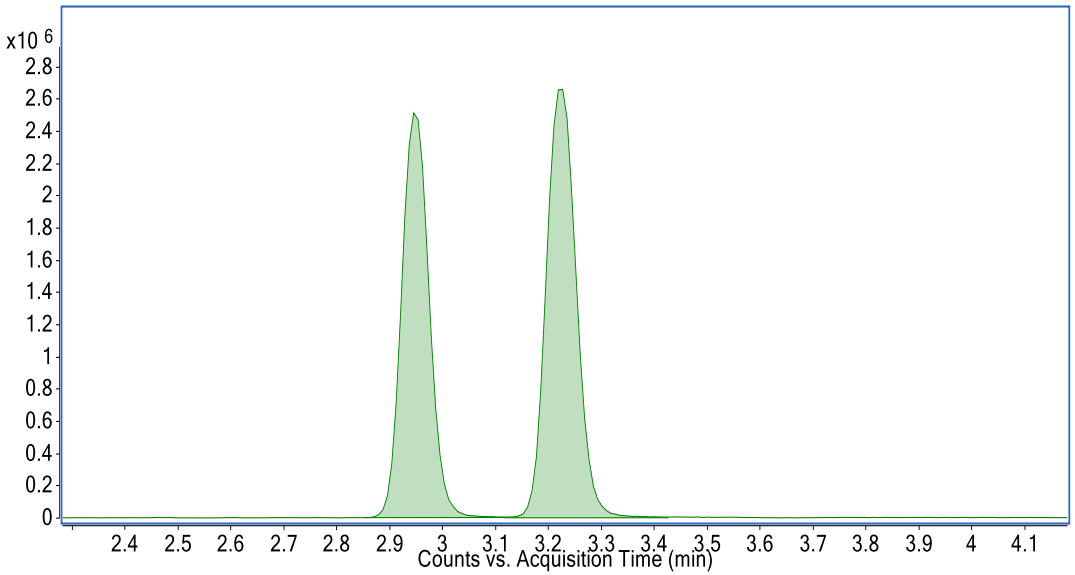


# Reproducibility Test



# Leu/Ile Resolution

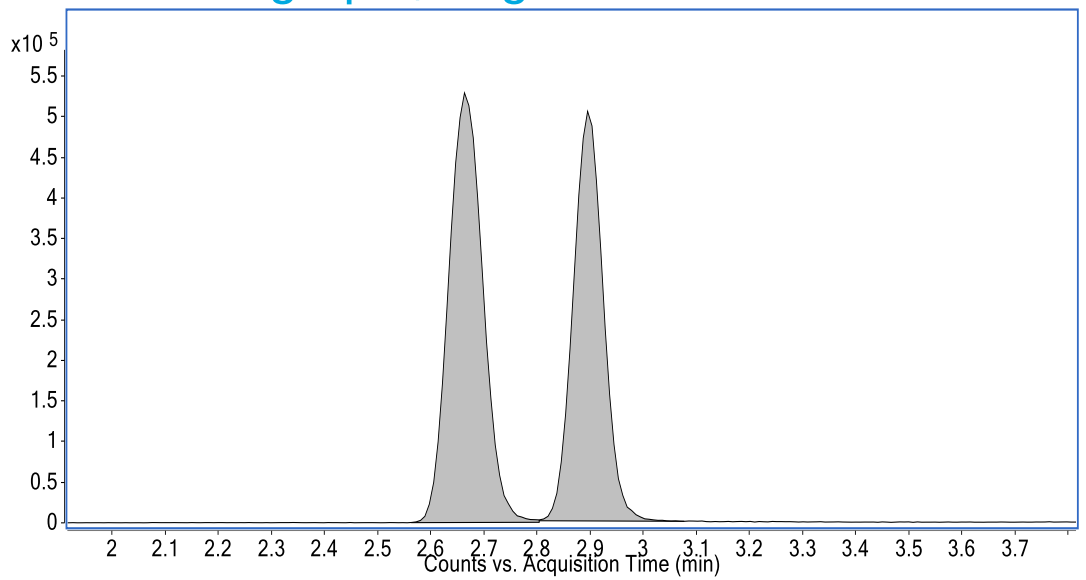
## Low pH, positive ion mode




**System Suitability**

| RT | Area | Height | Symmetry | Width | Plates | Resolution |
|---|---|---|---|---|---|---|
| 2.946 | 9080018.68 | 2511695.61 | 0.71 | 0.265 | 17910 | 66.9 |
| 3.224 | 10510201.33 | 2657458.46 | 1 | 0.306 | 16575 | 3 |

## High pH, negative ion mode

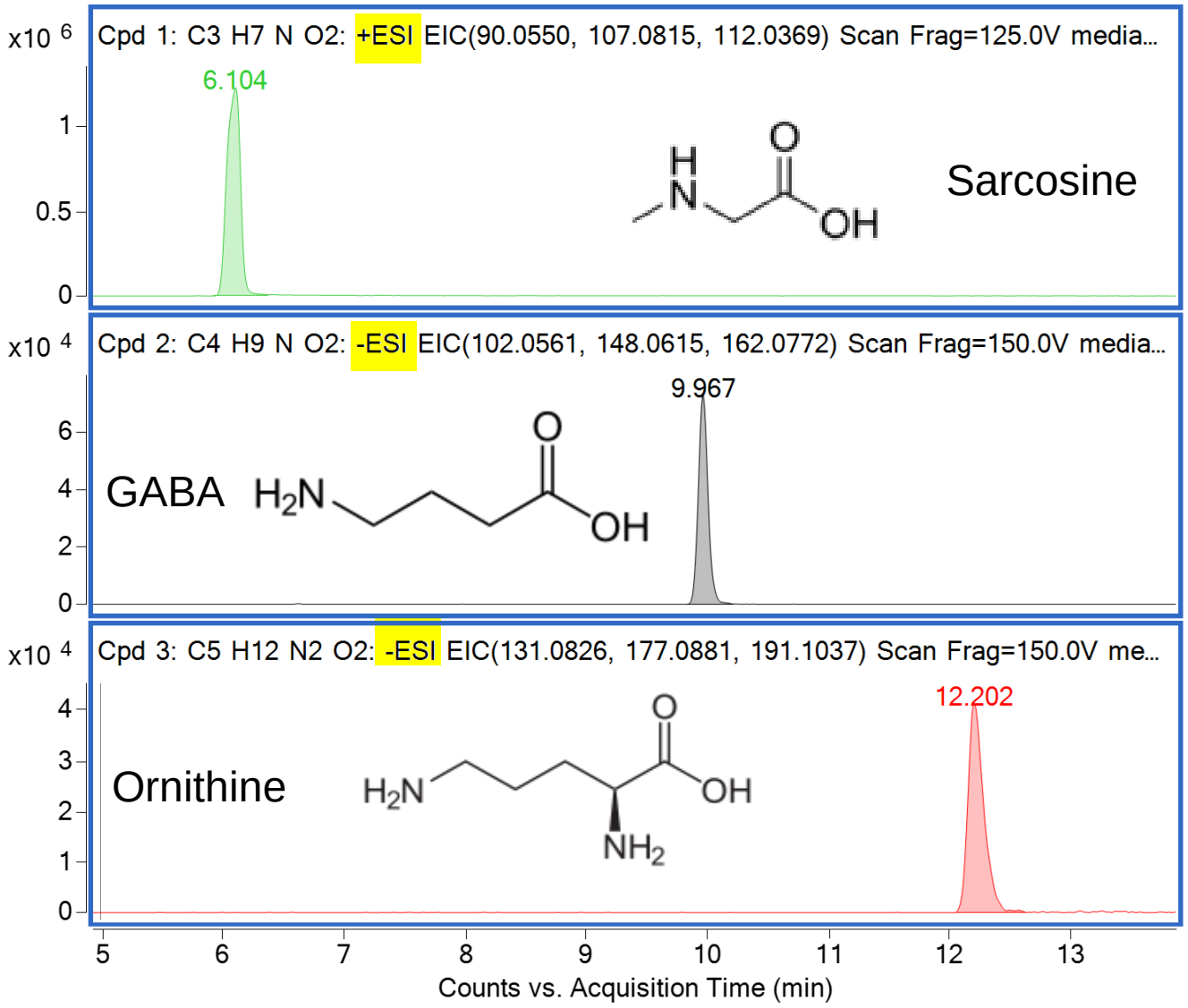



**System Suitability**

| RT | Area | Height | Symmetry | Width | Plates | Resolution |
|---|---|---|---|---|---|---|
| 2.664 | 2371609.84 | 529034.04 | 0.88 | 0.24 | 8347 | 45.7 |
| 2.896 | 1997455.1 | 504458.71 | 1 | 0.273 | 14420 | 2.2 |

Resolution > 1.5
(European Pharmacopeia requirement)

# Nonstandard Amino Acids in Cell Culture Media



# Application Note – Experimental Details and Results

Agilent InfinityLab


## Analysis of Underivatized Amino Acids by LC/MS for Bioreactor Cell Culture Monitoring

Pub no. 5991-8816EN

**Authors**

Jordy Hsiao, Te-Wei Chu, Andrew Kennedy, Adam Bivens, and Anne Blackwell

**Abstract**

This Application Note presents a solution for LC/MS analysis of amino acids in fermentation media. The polar nature of amino acids makes analysis by reversed-phase liquid chromatography challenging, so derivatization is often used to improve retention. However, hydrophilic interaction chromatography (HILIC) is capable of retaining and separating complex amino acid mixtures without derivatization, while still offering a similar workflow to traditional reversed-phase. The combination of HILIC and mass spectrometry offers a particularly simple and powerful solution for underivatized amino acid analysis.

# Amino Acid Workflows

Derivatization?

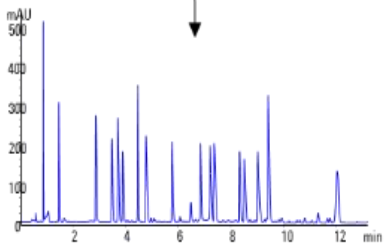
AdvanceBio Amino Acid Analysis
Ready to use reagents

AdvanceBio Amino Acid Analysis
LC/UV

Yes

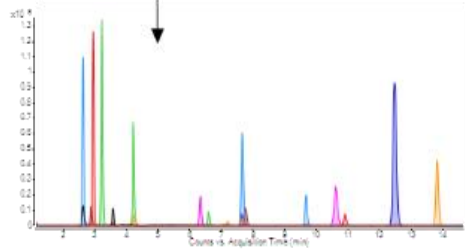
No

AdvanceBio MS Spent Media
LC/MS




Amino Acids

Fast, reliable results



Amino Acids & Cell Culture Metabolites

# Cell Culture Media Analysis – Choosing an Approach

## Derivatized amino acid analysis: LC/UV

- Industry standard, widely used
- Any Agilent LC
- Minimal instrumentation and expertise investment
- Reverse phase separation – very robust

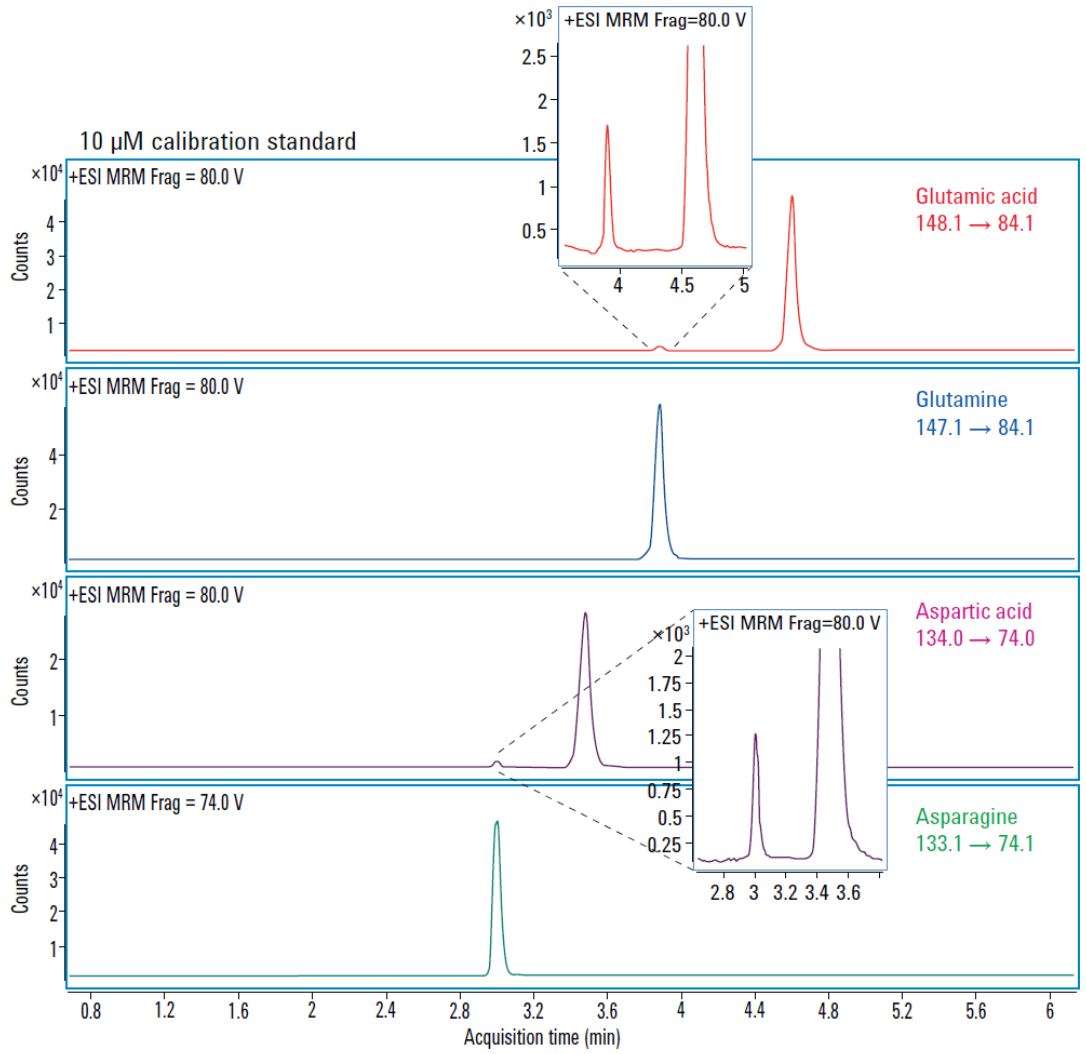

## Underivatized amino acid analysis: LC/MS

- Any LC/MS
- More expertise required, at least initially
- HILIC separations less familiar
- Higher sensitivity
- Savings:
  - No time spent derivatizing
  - No reagents to purchase
  - Combine assays for amino acids and other metabolites into a single method
  - *Possibly* - faster method since baseline chromatographic resolution isn't necessary with MS
    - Must still resolve isomers (Leu/Ile)

# Outline

- History of Amino Acid Analysis at Agilent
- Reverse phase LC/UV analysis of derivatized amino acids
- HILIC LC/MS of underivatized amino acids and other metabolites
- Ion pairing analysis of underivatized amino acids
- Chiral analysis of amino acids

# Amino Acids by Ion Pairing




| LC Conditions | | | |
|---|---|---|---|
| Column | Agilent ZORBAX SB-C18 RRHT column, 3.0 x 150 mm, 1.8 µm, part number 829975-302 | | |
| Column temperature | 25 °C | | |
| Injection volume | 1 µL | | |
| Autosampler temperature | 4 °C | | |
| Needle wash | 10 seconds in wash port | | |
| Mobile phase | A = 0.5 % formic acid and 0.3 % HFBA in water<br>B = 0.5 % formic acid and 0.3 % HFBA in acetonitrile | | |
| Flow rate | 0.4 mL/min | | |
| Gradient program | Time (min) | A (%) | B (%) |
| | Initial | 100 | 0 |
| | 5.00 | 95 | 5 |
| | 5.01 | 10 | 90 |
| | 6.00 | 10 | 90 |
| | 6.01 | 100 | 0 |
| Post time | 1 min | | |
| **Triple quadrupole MS source conditions** | | | |
| Ion mode | Positive | | |
| Drying gas temperature | 275 °C | | |
| Drying gas flow | 9 L/min | | |
| Sheath gas temperature | 325 °C | | |
| Sheath gas flow | 12 L/min | | |
| Nebulizer pressure | 40 psi | | |
| Capillary voltage | 3750 V | | |
| Nozzle voltage | 0 V | | |
| Delta EMV | 0 V | | |

# Chiral Analysis of Amino Acids (Chiral Column)

| Parameter | Value |
|---|---|
| Column | Agilent InfinityLab Poroshell 120 Chiral-T, 2.1 × 100 mm, 2.7 µm (p/n 685775-603) |
| Mobile phase | Premix 70/30 methanol/ammonium formate, pH 3.0, 25 mM |
| Flow rate | 0.21 m/min |
| Temperature (column) | 30 °C |
| Injection volume | 1 µL |
| Sample concentration | 2 mg/mL in water |

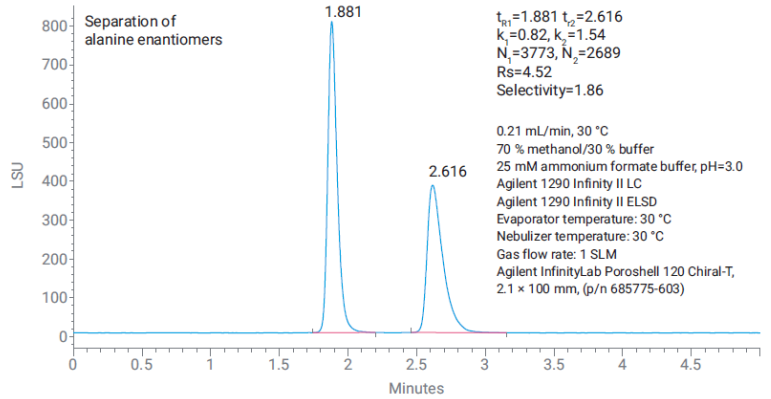

Chiral Analysis of Hydrophobic Amino Acids with Agilent InfinityLab Poroshell 120 Chiral-T Column

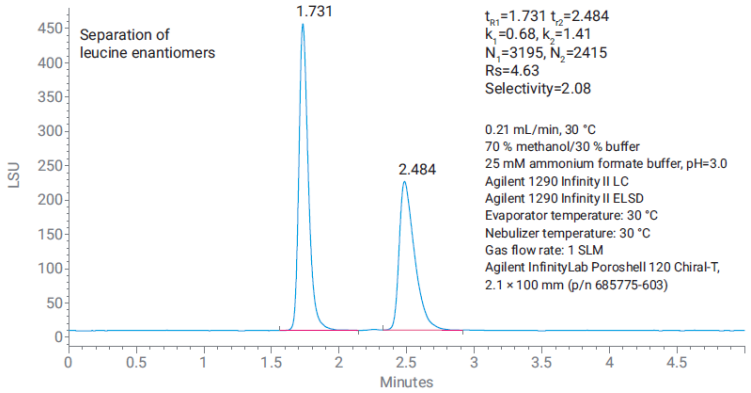
**Author**
William J. Long
Agilent Technologies, Inc.

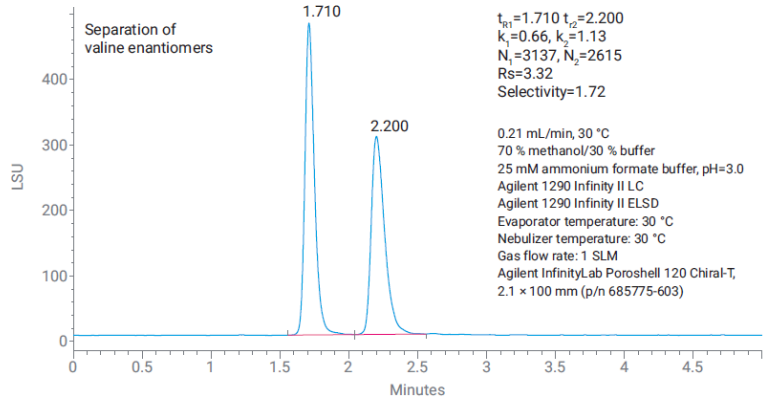
**Abstract**

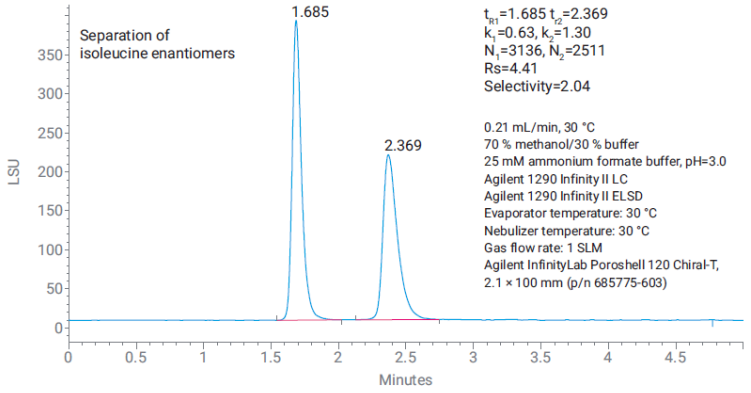
The chiral separation of a series of underivatized aliphatic amino acids was performed using an Agilent InfinityLab Poroshell 120 Chiral-T column using a methanol/ammonium formate buffer mobile phase. The separation of these D- and L-enantiomers is monitored using an ELSD detector. The L-enantiomer elutes first in all four cases.

Pub no. 5994-1366EN

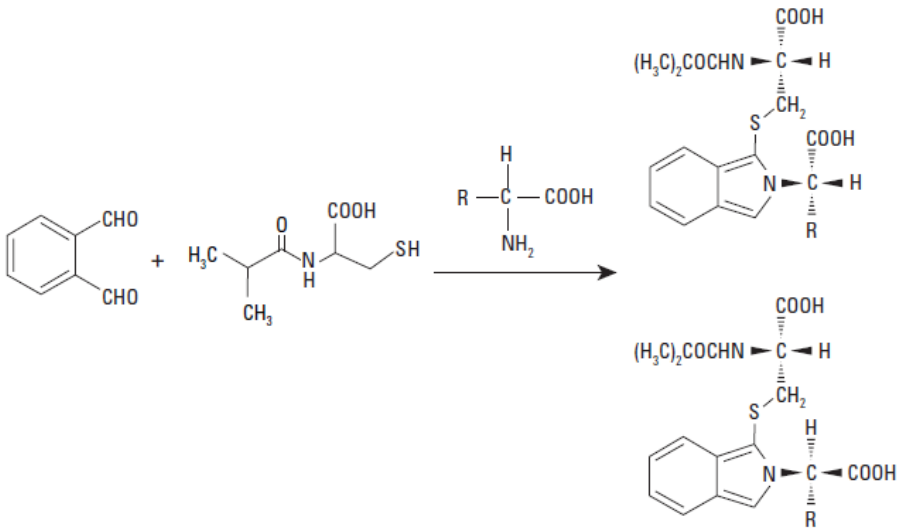


# Chiral Analysis of Amino Acids (By Derivatization)

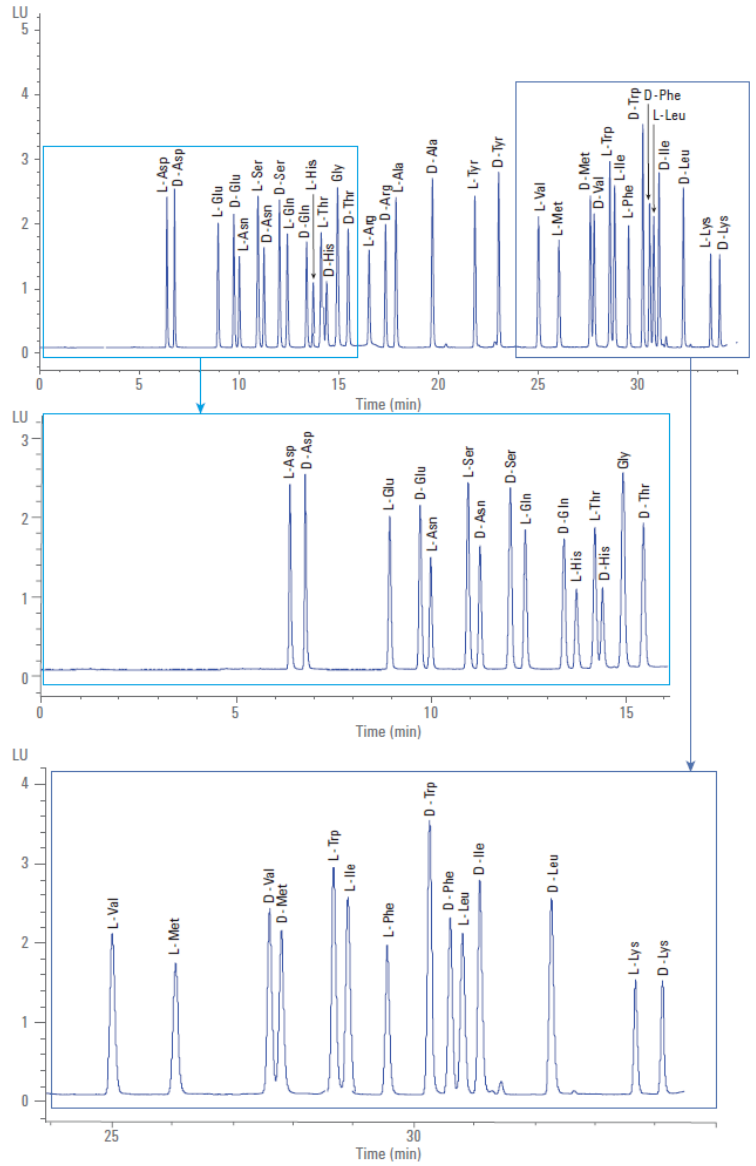


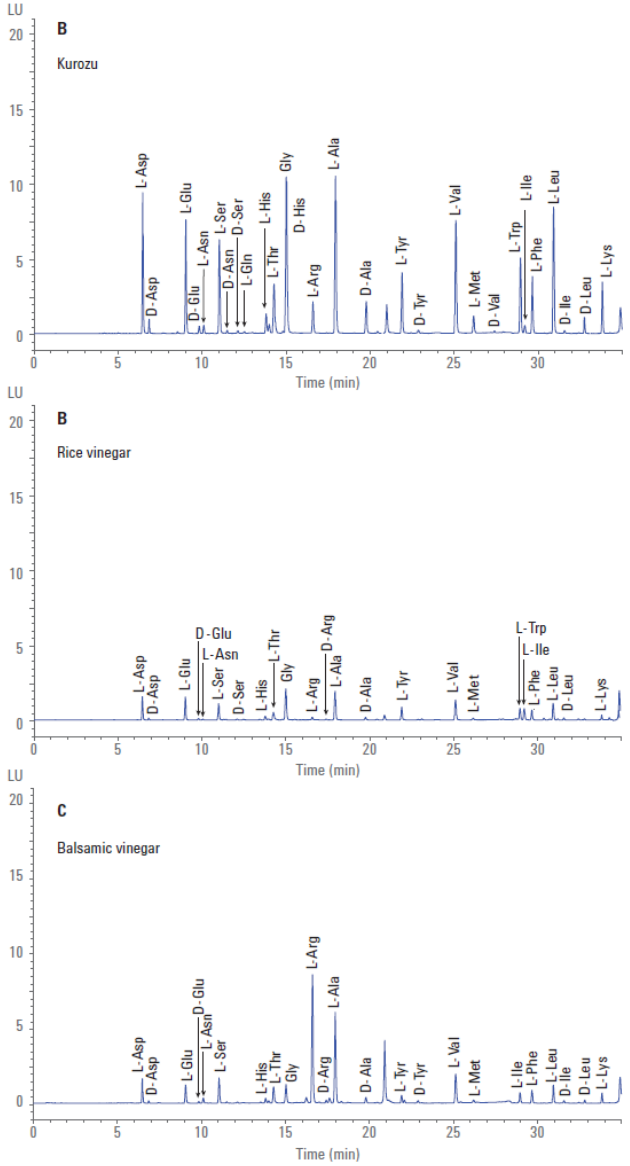
- Diastereomers can often be analyzed by reversed phase on a C18 column
- Derivatized with OPA and N-isobutyryl-L-cysteine in borate buffer
- Similar program for the online derivatization

| Parameter | Value |
|---|---|
| Column | Agilent Poroshell HPH-C18, 3.0 × 150 mm, 2.7 µm (p/n 693975-502) |
| Mobile phase | A) 50 mM sodium acetate (pH 6.0)<br>B) Acetonitrile/methanol/water 45/45/10 |
| Flow rate | 0.7 mL/min |
| Gradient Pump | 0 to 2.0 minutes, 4 %B<br>2.0 to 4.0 minutes, 10 %B<br>4.0 to 15 minutes, 20 %B<br>15 to 27 minutes, 35 %B<br>27 to 35 minutes, 50 %B<br>35 to 37 minutes, 100 %B<br>37 to 42 minutes, 100 %B |
| Post time | 10 minutes at 4 %B |
| Column temperature | 30 °C |
| Injection | See injector program |
| Needle wash | 40 °C |
| Detection | Ex. 230 nm, Ex. 450 nm |

| Step | Mode | Action |
|---|---|---|
| 1 | Draw | Draw 2.5 µL from location 1 with default speed |
| 2 | Draw | Draw 0.5 µL from sample |
| 3 | Wash | Wash needle in flush port with S1 for 3 seconds with 100 µL /min |
| 4 | Mix | Mix 3.0 µL from air at maximum speed 10 times |
| 5 | Wait | Wait 0.5 minutes |
| 6 | Draw | Draw 0.25 from location 2 with 100 µL/min speed |
| 7 | Mix | Mix 3.25 µL from air at maximum speed 20 times |
| 8 | Wait | Wait 0.5 minutes |
| 9 | Draw | Draw 15 µL from location 3 with default speed |
| 10 | Mix | Mix 20 µL from air at maximum speed 10 times |
| 11 | Wait | Wait 0.1 minutes |
| 12 | Inject | Injection |
| 13 | Wait | Wait 0.5 minutes |
| 14 | Valve | Switch valve to **Bypass** |

# Chiral Analysis of Amino Acids (By Derivatization)



# Agilent Resources for Support

- Resource page http://www.agilent.com/chem/agilentresources
  - Quick reference guides, product catalogs
  - Online selection tools, “How-to” videos
  - Column user guides - https://www.agilent.com/en-us/support/liquid-chromatography/kb005965
  - Biocolumn user guides - https://www.agilent.com/en/support/liquid-chromatography/kb005960
- Tech support: http://www.agilent.com/chem/techsupport
- InfinityLab LC Supplies catalog (5991-8031EN)
- Agilent University http://www.agilent.com/crosslab/university
- YouTube – Agilent Channel
- Your local product specialists
- Subscribe to Agilent Peak Tales podcasts at peaktales.libsyn.com


# Contact Agilent Chemistries and Supplies Technical Support

Available in the USA & Canada 8-5 all time zones

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Option 2 for LC and LC/MS columns and supplies

Option 3 for sample preparation, filtration and QuEChERS

Option 4 for spectroscopy supplies

Option 5 for chemical standards

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chem-standards-support@agilent.com