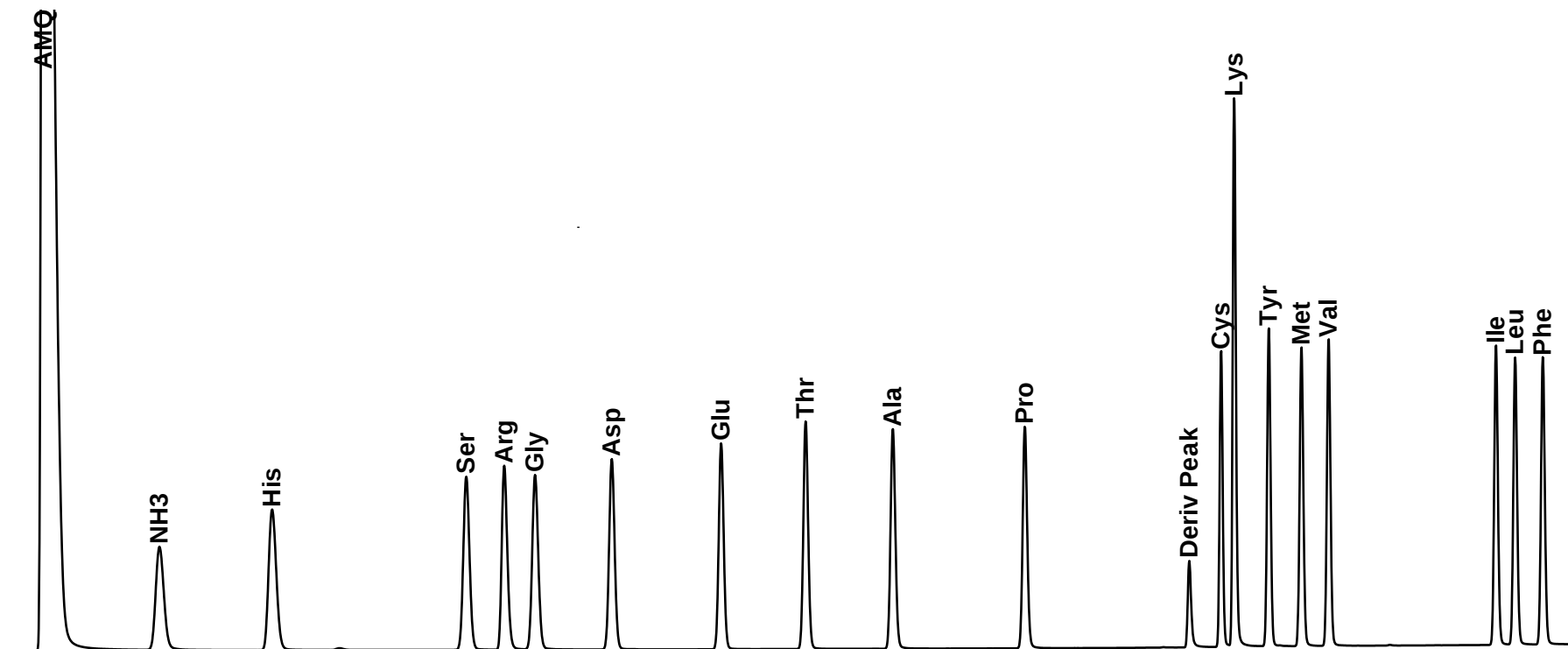


UPLC Amino Acid Analysis Solution

AAAFCO Meeting
Washington, DC
August 2, 2009

UPLC® Amino Acid Analysis Solution

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AccQ•Tag™ Ultra
UPLC™ Amino Acid Analysis

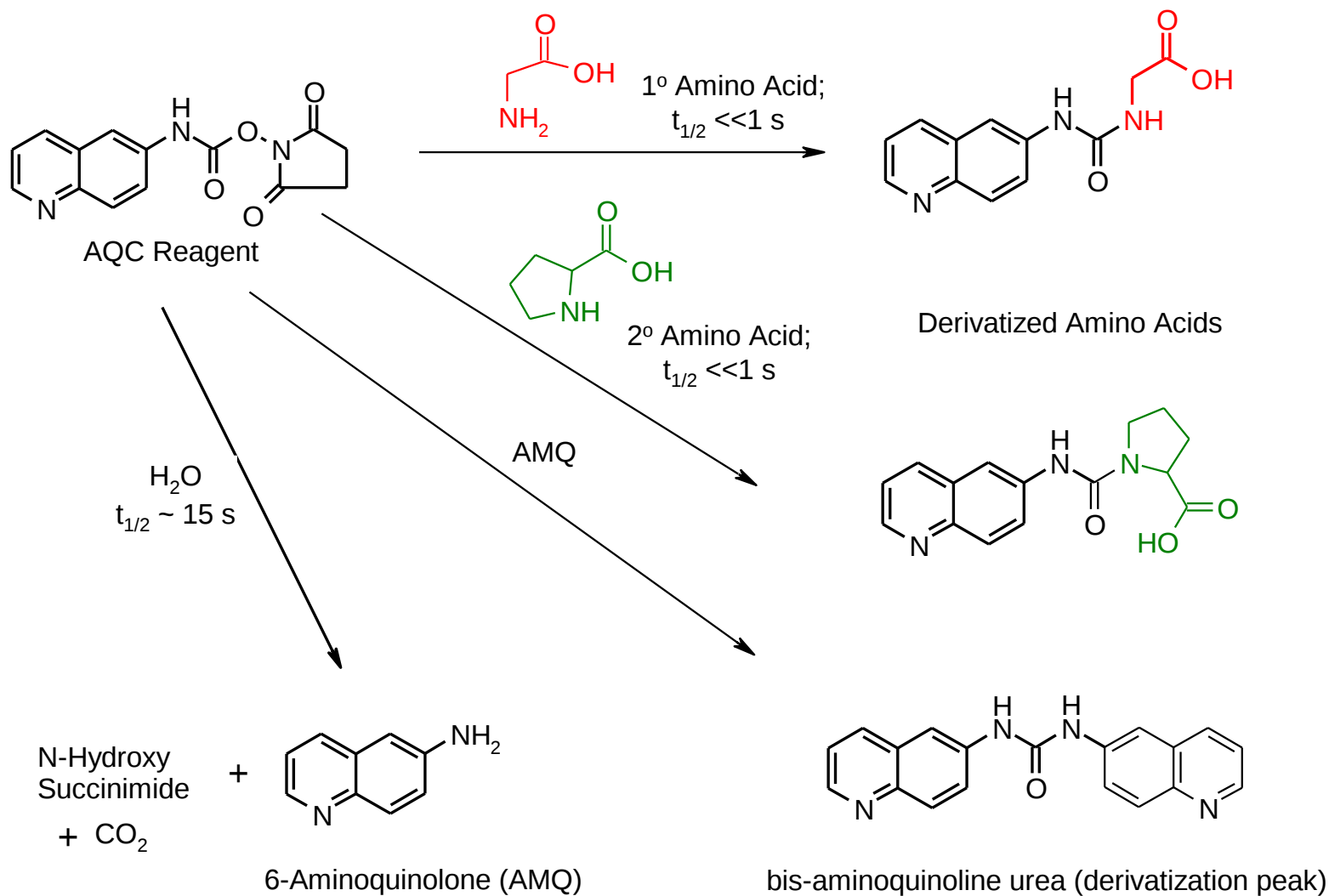
UPLC® Application Solution for Amino Acid Analysis

- Turn-key UPLC® Amino Acid Analysis application
 - Optimized for the ACQUITY UPLC System
 - Dedicated QC-tested column and reagents
 - Application-specific Performance Qualification
 - Same result day-to-day, instrument-to-instrument, lab-to-lab, around the world
- Target Applications
 - Protein and peptide ID and quantitation
 - Monitoring cell culture media
 - Nutritional content of food and feed

Total System Approach Assured Performance

- ACQUITY UPLC® System with TUV Detector
- Empower software control
- Application Start Up Kit Containing:
 - Manual
 - CD with method templates and example data/calculations
 - Necessary instrument accessories
- AccQ●Tag™ Ultra Chemistry Kit
 - Reagent
 - Buffers/diluents/eluents
 - QC tested ACQUITY UPLC BEH 2.1 x 100mm, 1.7 µm C18 column
- Add-on Kit for existing ACQUITY UPLC Users

Chemistry of AQC Derivatization

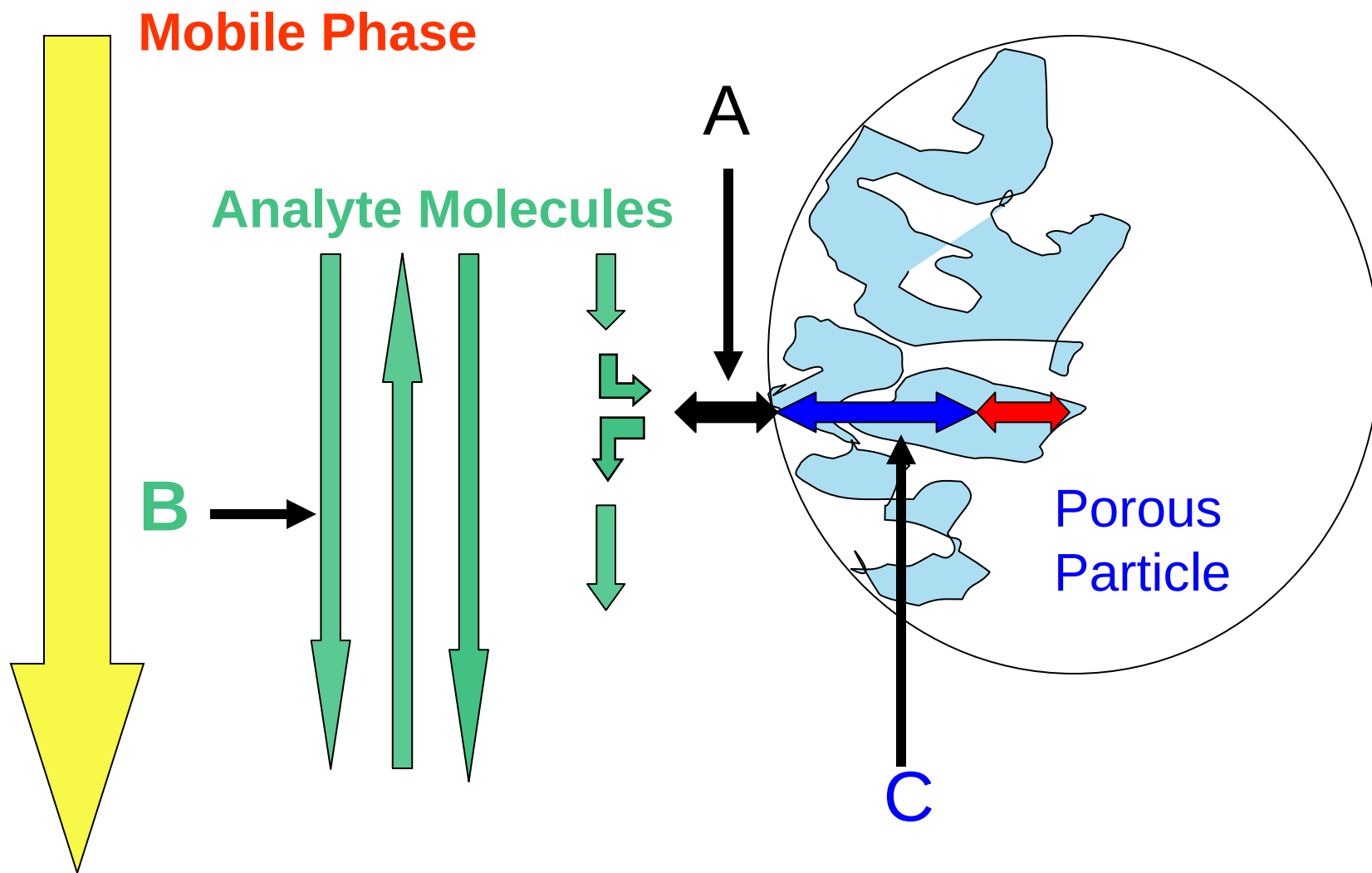


- A new class of separation science
 - Based on chromatography columns with very small particles
 - Based on instruments designed to take advantage of the small particles
- Provides improved Resolution, Speed, and Sensitivity with no compromises
- Suitable for chromatographic applications in general
 - Appropriate for developing new methods
 - Appropriate for improving existing methods

Chromatography Principles

Mass Transfer / Diffusion

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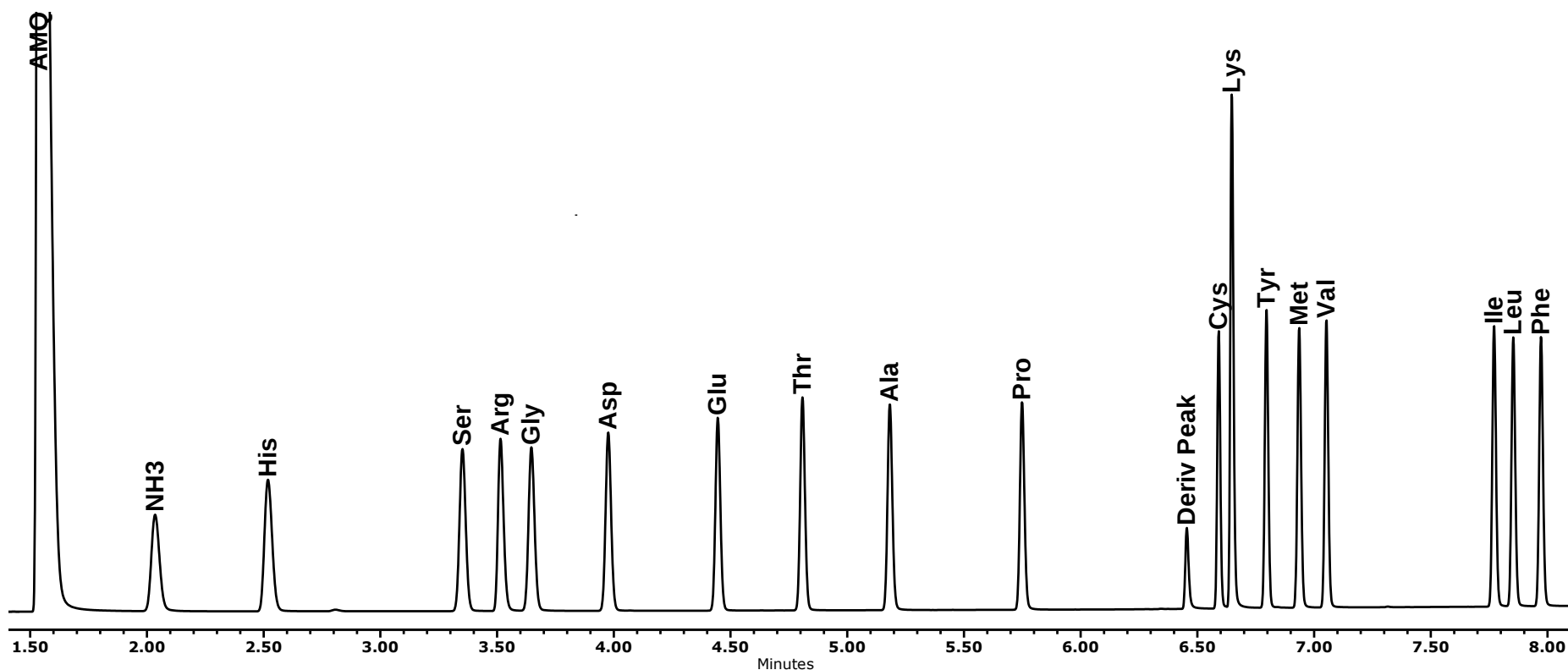
UPLC® Amino Acid Analysis Application Solution

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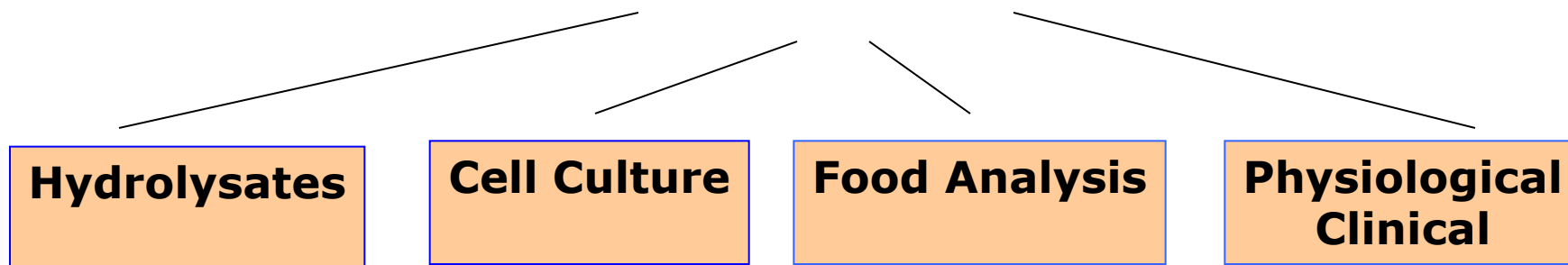
AccQ•Tag™ Ultra
UPLC™ Amino Acid Analysis

Amino Acid Analysis

Application Types

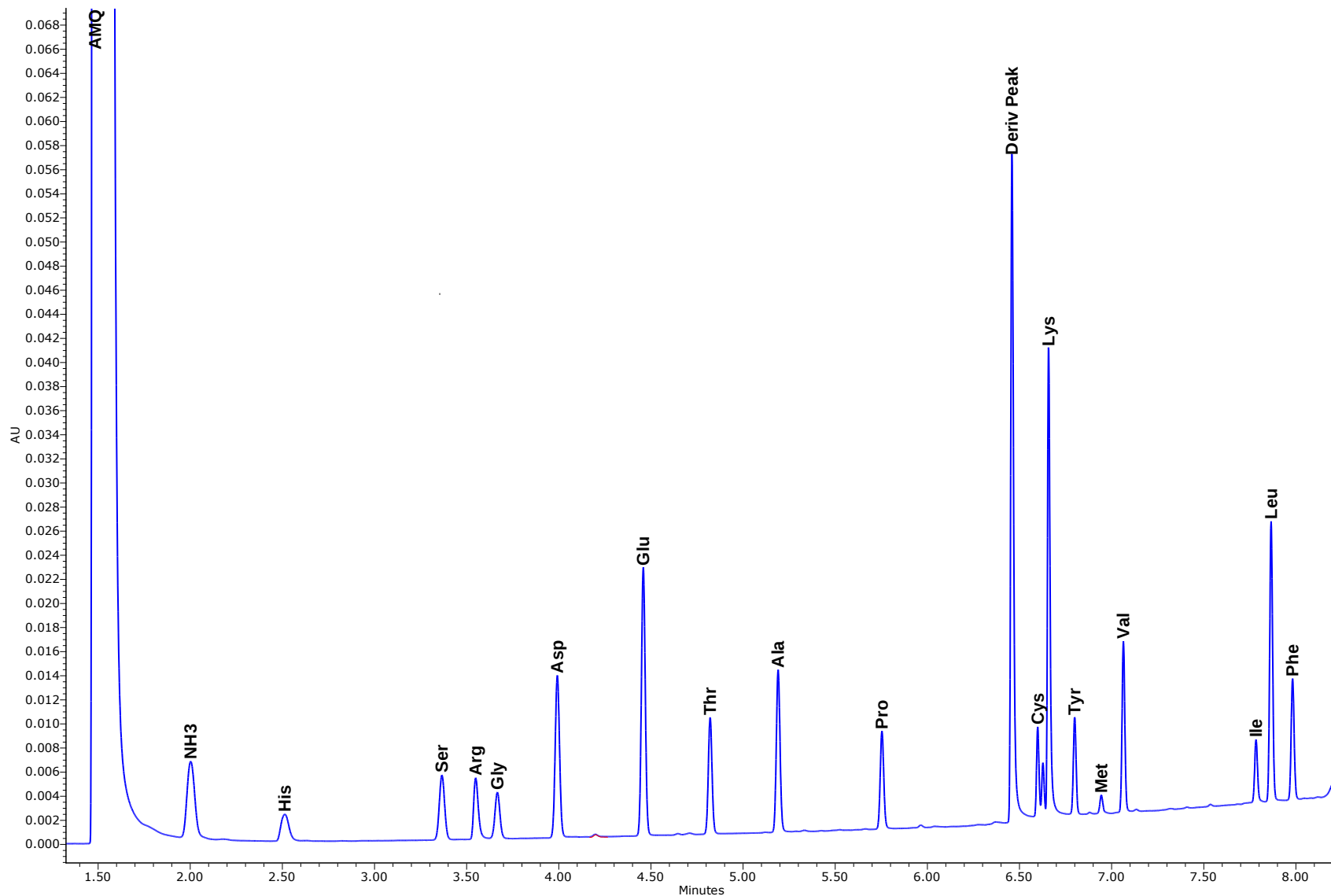
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Amino Acid Analysis



Analysis of Acid Hydrolyzed BSA 9ng on column

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Accuracy of Compositional Analysis Bovine Serum Albumin

Amino Acid	Expected # Residues	*Observed # Residues
His	17	15.36 ± 0.19
Ser	28	26.00 ± 0.08
Arg	23	22.37 ± 0.08
Gly	16	17.68 ± 0.20
Asp	54	55.47 ± 0.21
Glu	79	80.68 ± 0.20
Thr	33	31.92 ± 0.06
Ala	47	47.51 ± 0.15
Pro	28	28.35 ± 0.14
Lys	59	57.78 ± 0.38
Tyr	20	20.19 ± 0.08
Met	4	4.16 ± 0.15
Val	36	35.67 ± 0.16
Ile	14	13.15 ± 0.15
Leu	61	63.13 ± 0.19
Phe	27	26.57 ± 0.13

* Mean ± Standard
Deviation for 75
independent
analyses

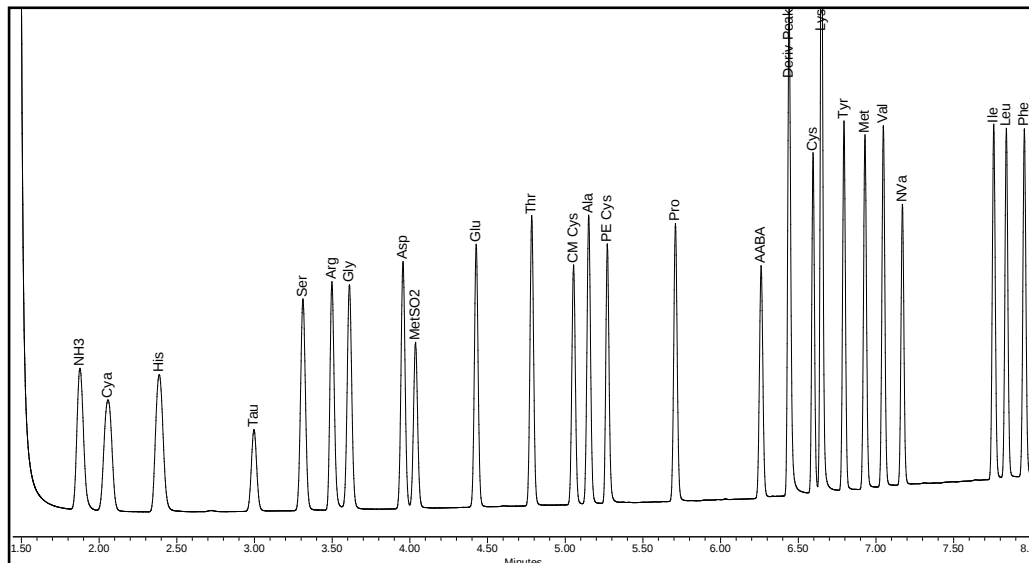
Measuring Protein Concentration Amino Acid Analysis

Sample ID	Total µg AA / mL Hydrolysate
BSA 1-1	699.64
BSA 1-2	694.09
BSA 1-3	697.88
BSA 1-4	698.21
BSA 1-5	695.43
BSA 2-1	716.66
BSA 2-2	717.87
BSA 2-3	715.65
BSA 2-4	709.75
BSA 2-5	707.93
BSA 3-1	714.93
BSA 3-2	711.50
BSA 3-3	708.46
BSA 3-4	708.14
BSA 3-5	708.10
BSA 4-1	674.16
BSA 4-2	678.43
BSA 4-3	683.07
BSA 4-4	678.62
BSA 4-5	678.94
BSA 5-1	591.4
BSA 5-2	604.3
BSA 5-3	571.78
BSA 5-4	571.57
BSA 5-5	599.18
Mean	677.43
Standard Deviation	47.98
% RSD	7.08

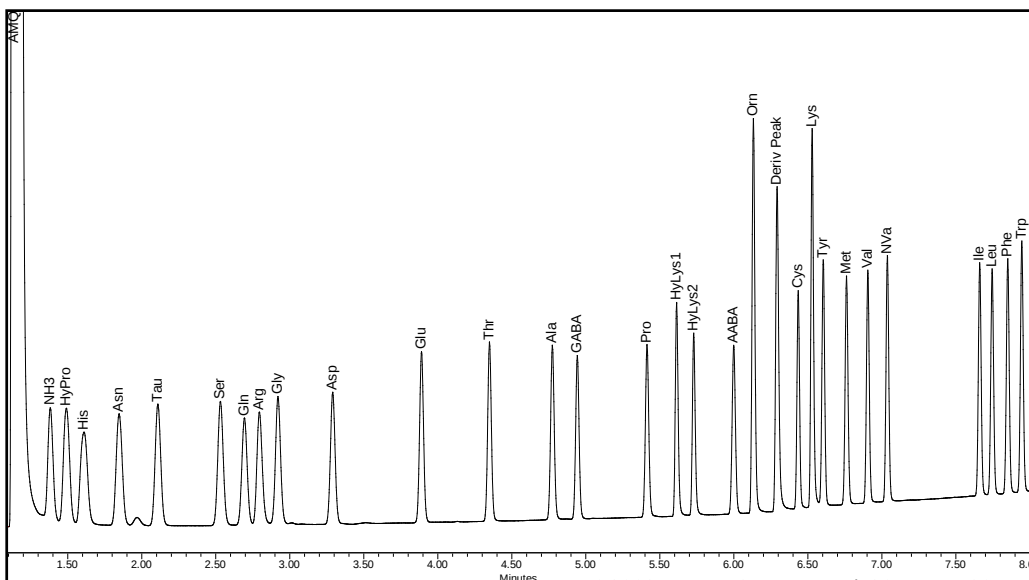
Method Options

Bound vs. Free Amino Acids

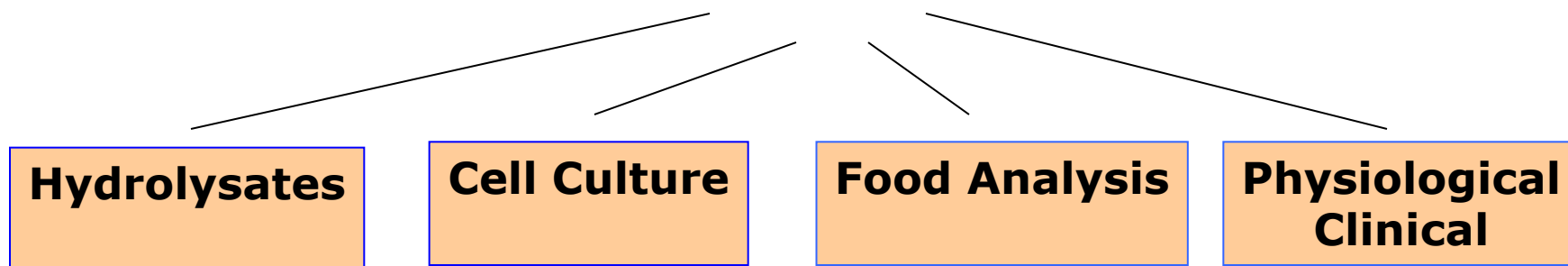
Protein Hydrolysate
20X Dilution of
Concentrate A
55



Cell Culture Media
10X Dilution of
Concentrate A
60



Amino Acid Analysis



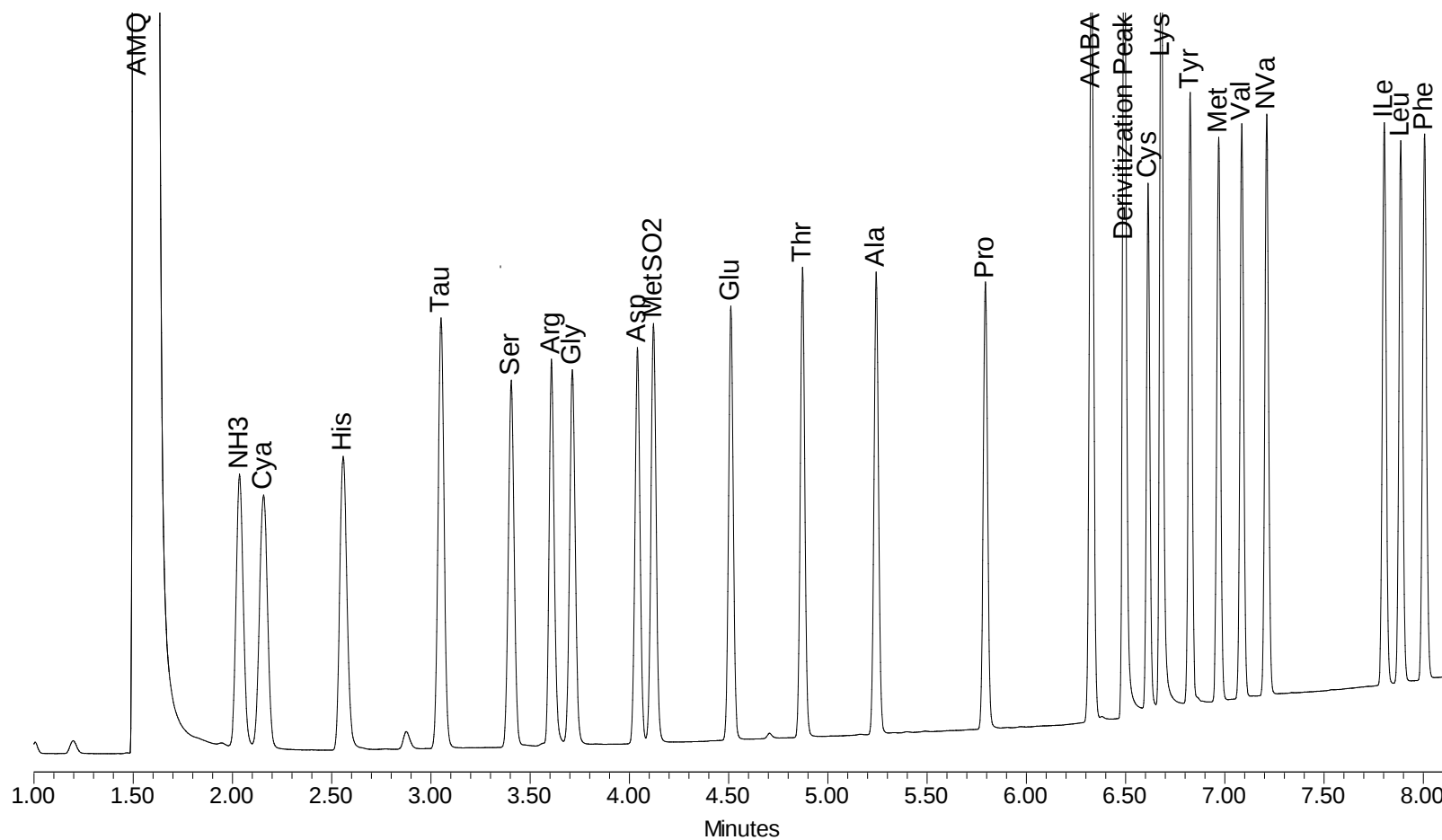
- Nutritional Content
 - Absolute amount of amino acids
 - Complex sample handling
- Identification/Adulteration
 - Unusual amino acids
 - Marker amino acids
 - Proprietary
- Criteria
 - Unequivocal identification
 - Accuracy, precision, linearity
 - Sensitivity and speed
 - Rugged and robust

- Nutritional monitoring
 - Total protein required to support health
 - Proportions of specific amino acids required
 - Essential amino acids that are not synthesized by the animal
 - Growth-limiting nutrients
- Identity testing
 - Free amino acids related to metabolic pathways
 - Free amino acids derived from the environment of the product

Foods and Feeds Analysis

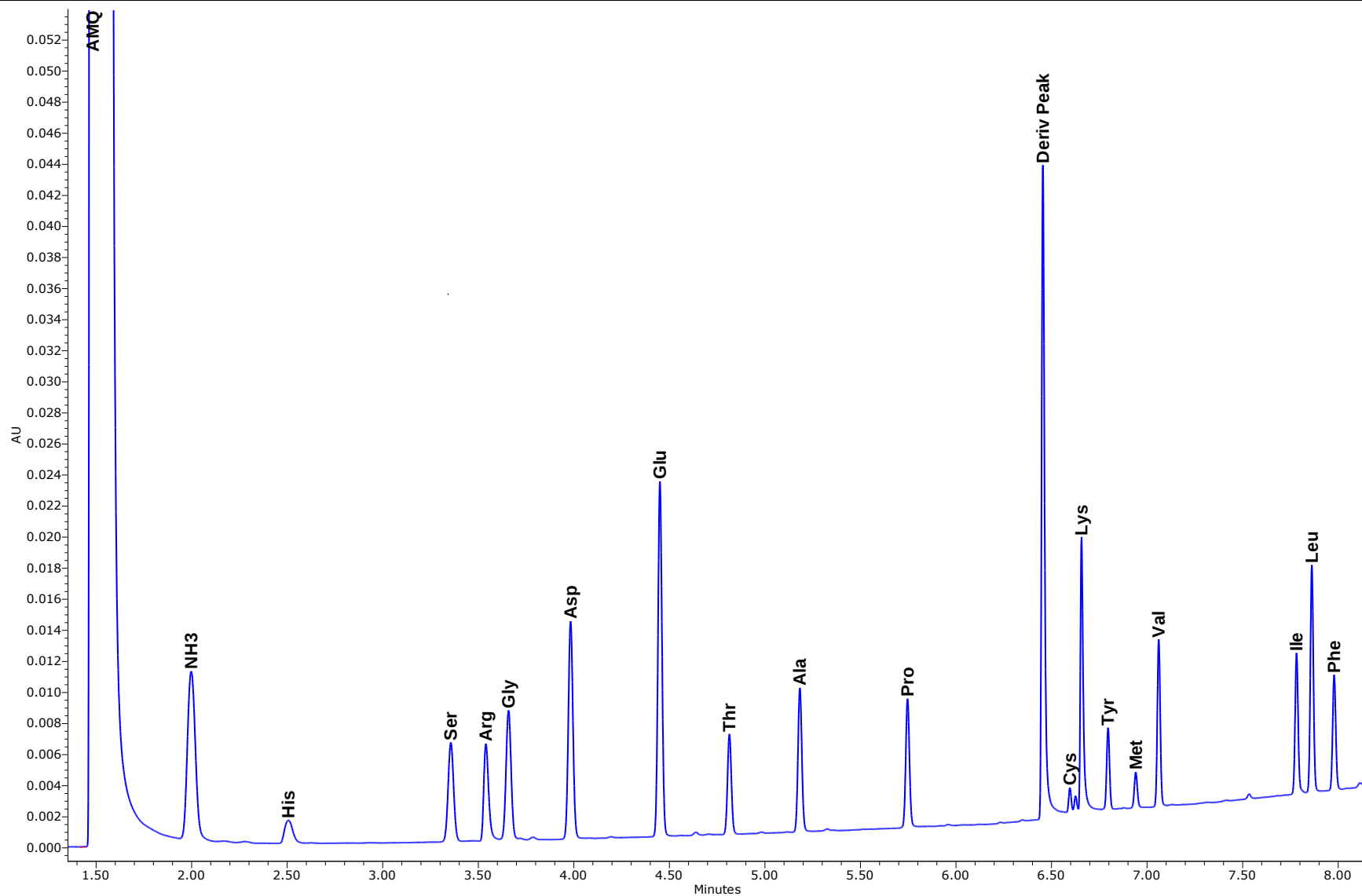
10 pmol Standard on Column

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Analysis of Soybean Meal Hydrolysate

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Analysis of Soybean Meal Hydrolysate

Amino Acid	Weight %	Amino Acid	Weight %
His	1.87 ± 0.03	Pro	3.86 ± 0.03
Ser	3.65 ± 0.02	Lys	4.67 ± 0.03
Arg	5.82 ± 0.03	Tyr	2.90 ± 0.02
Gly	2.98 ± 0.02	Met	1.08 ± 0.02
Asp	9.06 ± 0.05	Val	3.66 ± 0.02
Glu	14.49 ± 0.08	Ile	3.45 ± 0.02
Thr	2.92 ± 0.02	Leu	6.12 ± 0.03
Ala	3.17 ± 0.02	Phe	3.92 ± 0.02

Mean ± Standard Deviation for 75 independent replicate analyses

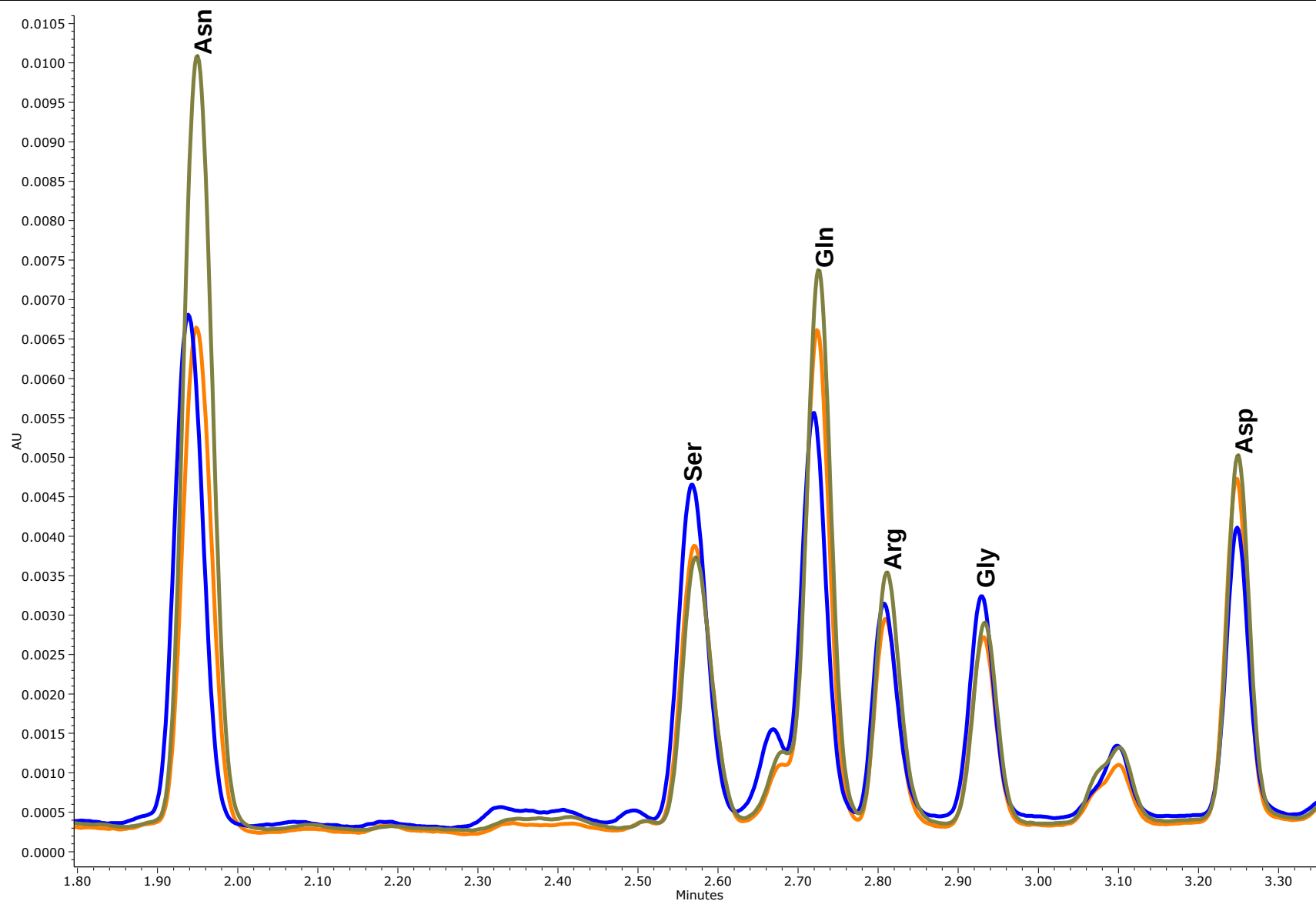
Analysis of Hydrolyzed Grains and Complex Feeds

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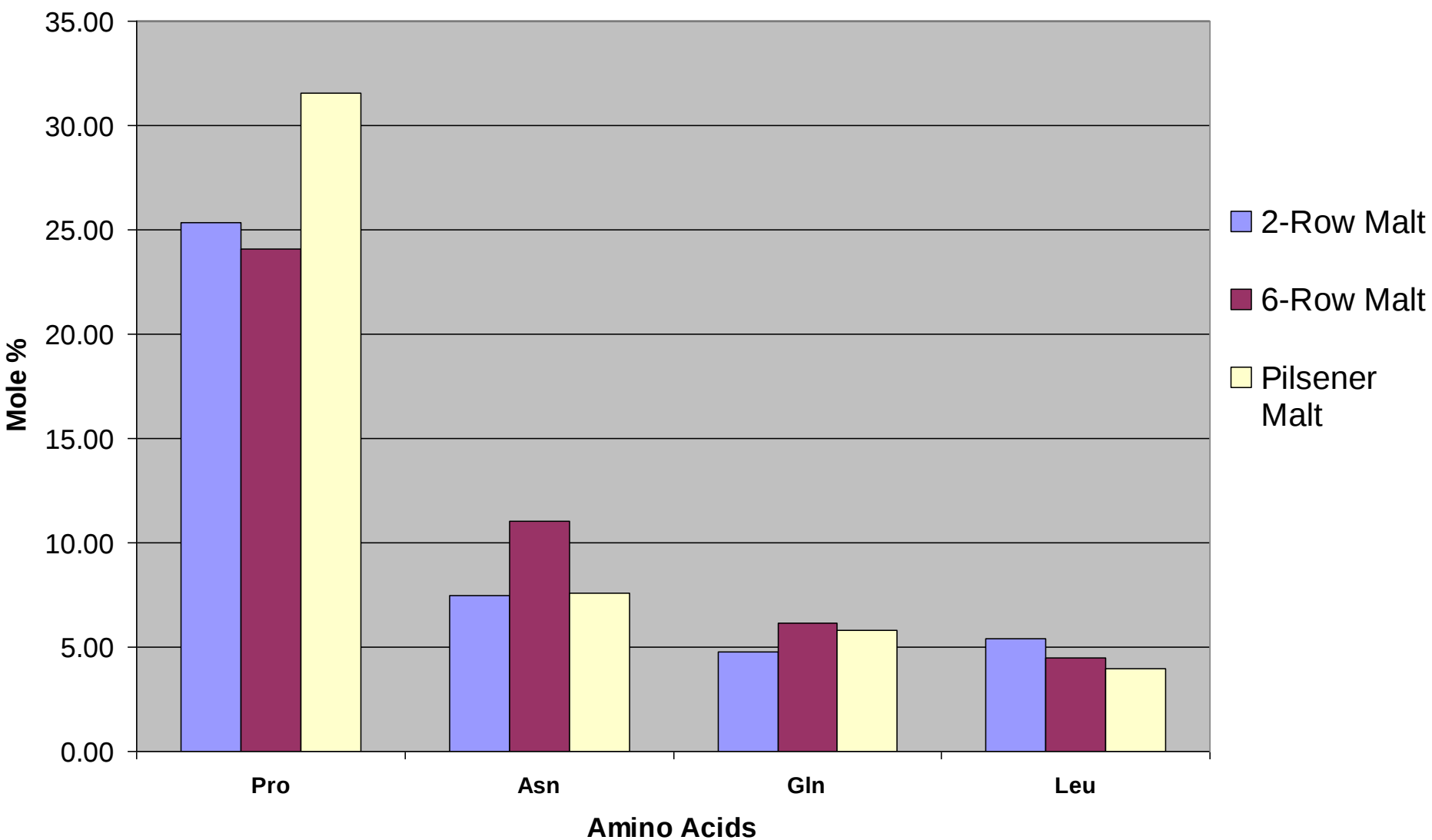
Free Amino Acids in Starting Malts for Brewing Process

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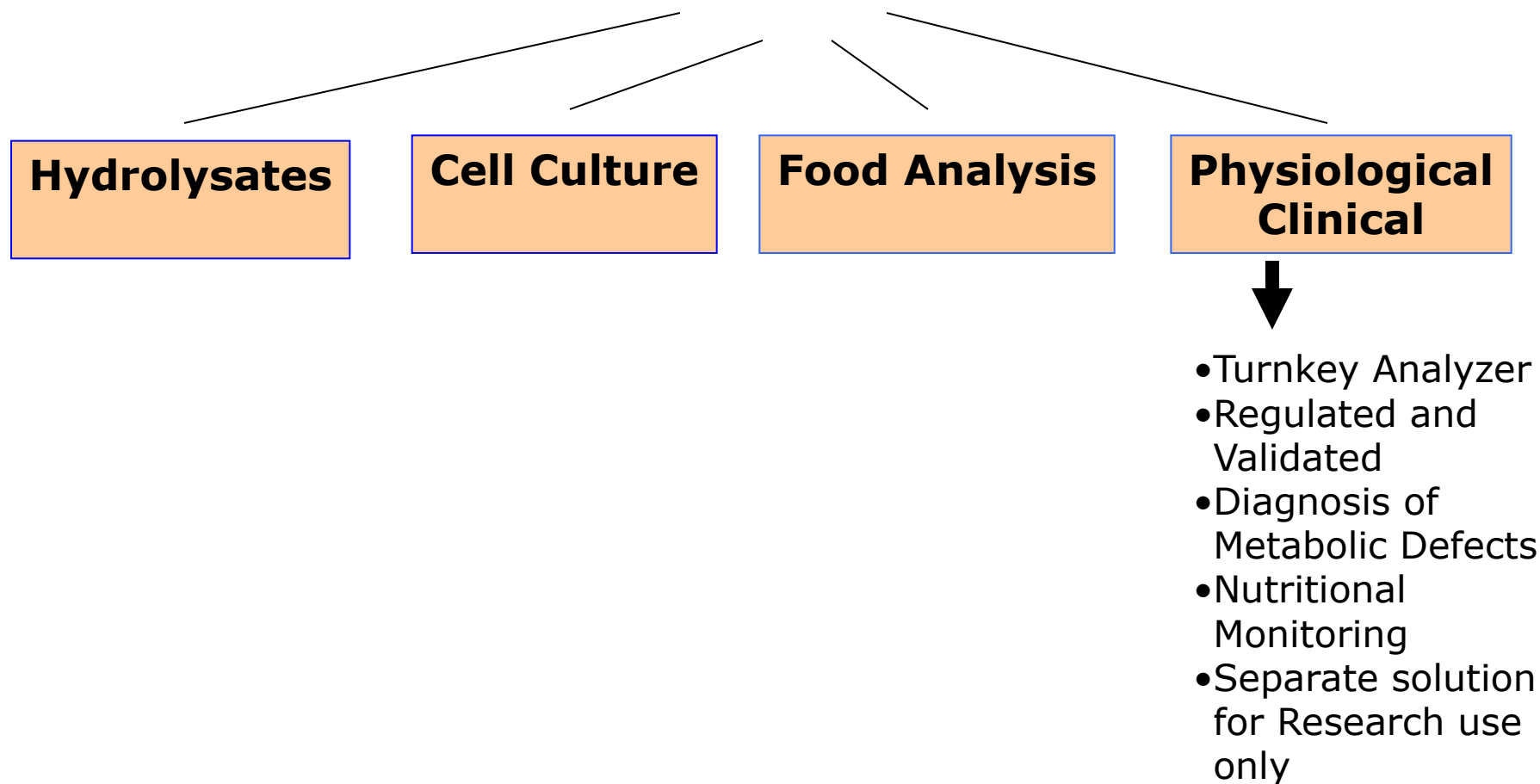


Free Amino Acids Levels Starting Malt

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Amino Acid Analysis



MassTrak™ Amino Acid Analysis Solution

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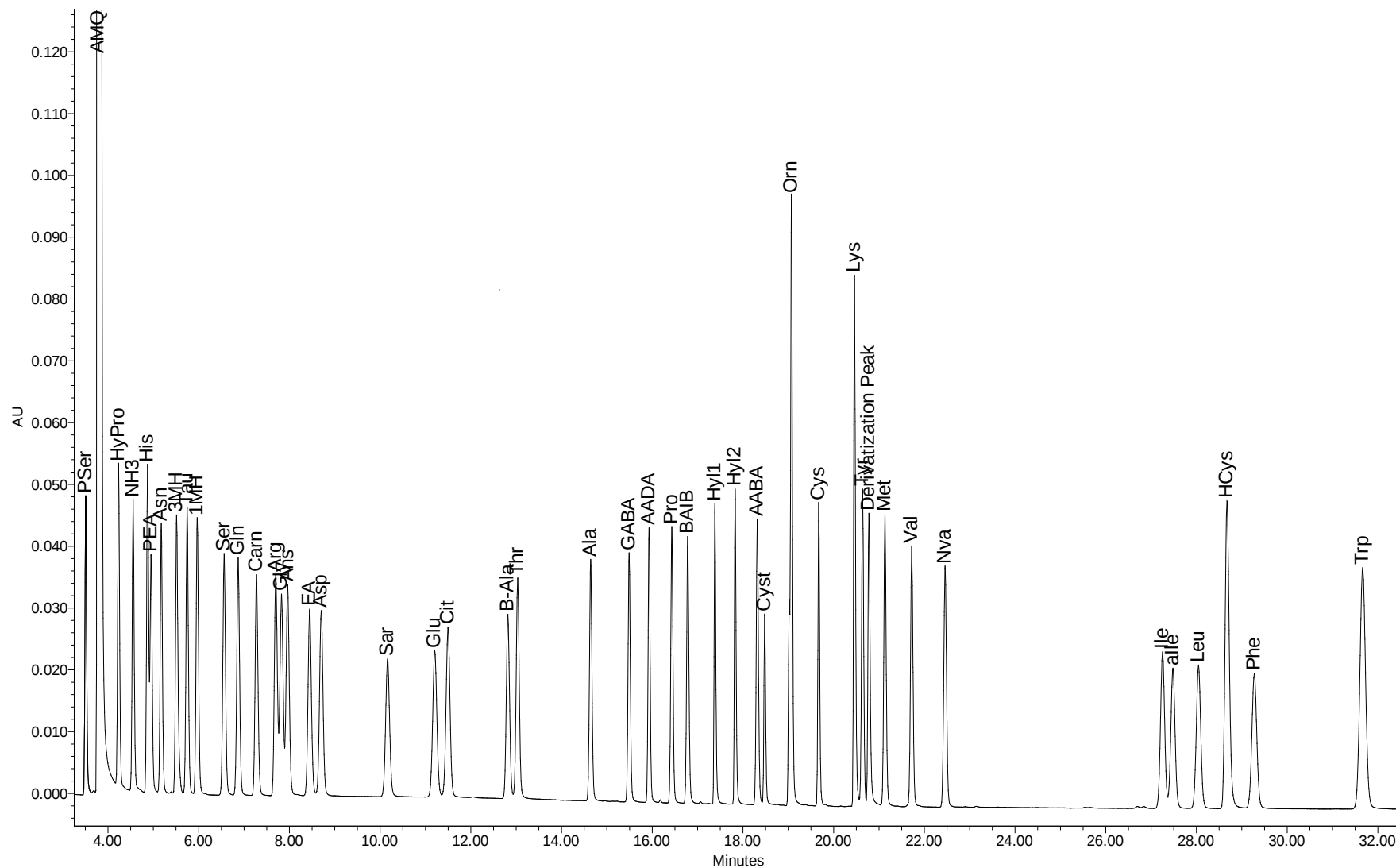
“A pre-configured set of – instrumentation, informatics, chemistries – designed to work together, made and supported by the same company for a specific application.”



**FOR RESEARCH USE ONLY.
NOT FOR USE IN DIAGNOSTIC PROCEDURES**

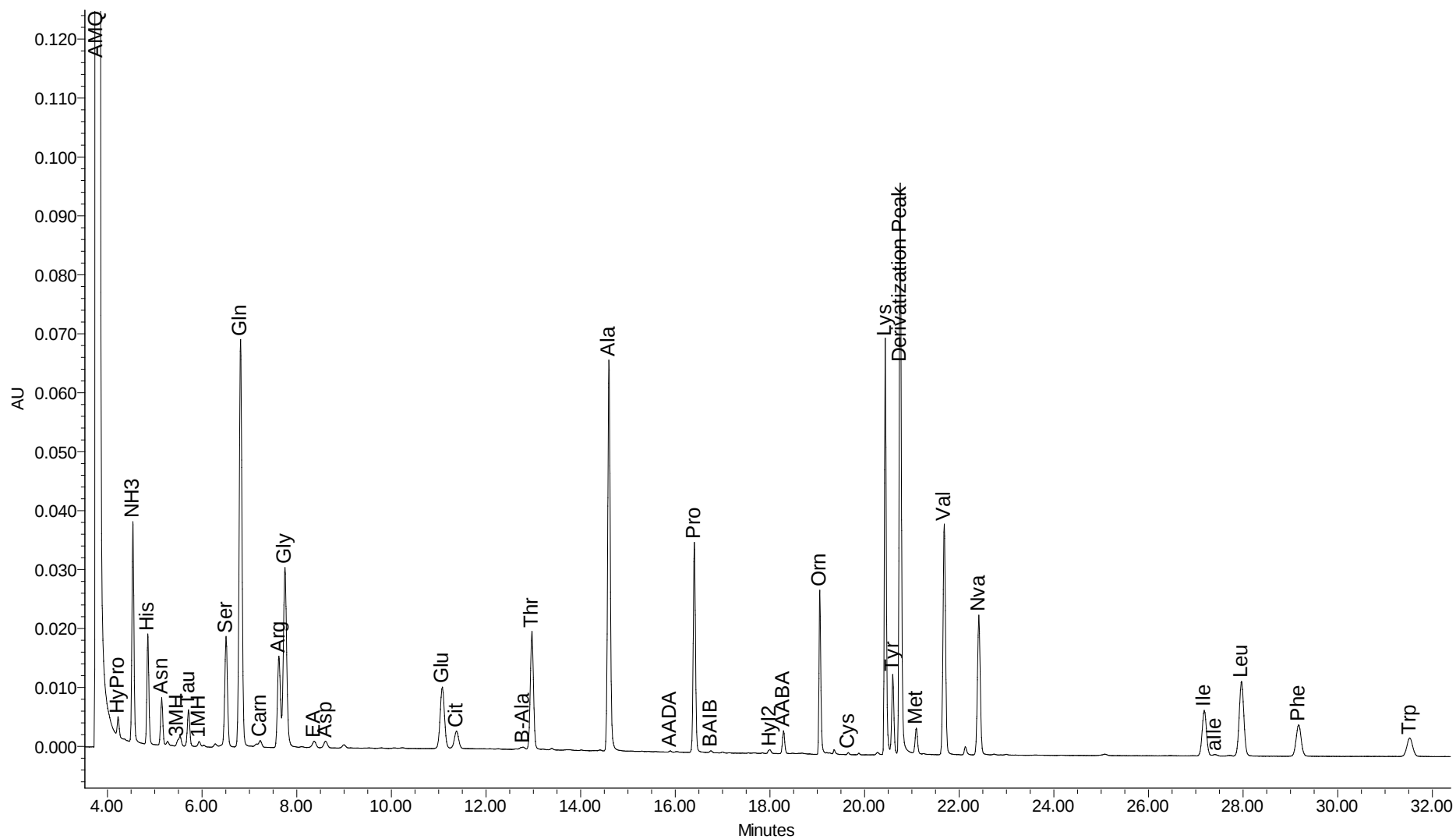
Physiological Amino Acid Standard

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MassTrak™ Amino Acid Analysis Plasma Sample

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UPLC® Amino Acid Analysis Application Solution

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- Turnkey System
 - Newly developed analytical method
 - Based on established chemistry and instruments
 - Components tested for ruggedness
- Demonstrated for most common applications
 - Protein structure
 - Cell culture
 - Nutritional analysis of foods and feeds
- Right Answer Assured