SHIMADZU
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Application News

No. L568

High Performance Liquid Chromatography

Analysis of Foods Using Nexera™ Post-Column Amino Acid Analysis System

Amino acids are essential nutrients that make up the proteins which form the human body. Analysis of amino acids has a long history of applying firstly to research purposes such as structural analysis and diagnoses of diseases and then to quality control of food products and drugs and other fields. Responding to increasing requests for higher speed analysis in recent years, Shimadzu Corporation developed an online pre-column method* that enables more rapid analysis. On the other hand, the post-column method is still frequently used today because of its wide range of applicability, even though it is a time-consuming technique.

Shimadzu post-column method employs an automatic analysis system that utilizes fluorescence derivatization by *o*-phthalaldehyde (OPA) in detection after separation of the amino acids by gradient elution using a cation exchange column. OPA derivatized amino acids have excellent sensitivity and linearity, and highly sensitive detection of proline and other secondary amino acids is also possible. Because the derivatizatoin is carried out after separation, reaction efficiency is less prone to sample matrix effects. Therefore, this post-column method can detect amino acids with high sensitivity and high selectivity. As separation techniques, two modes are available, the Na type for analysis of proteinogenic amino acids and the Li type for analysis of free amino acids. Although simultaneous analysis of amino acids is conducted using the optimum time programs for these respective separation modes, highly accurate amino acid analysis is possible without labor-intensive preparation using a dedicated amino acid mobile phase kit and reaction reagent kit.

This article introduces amino acid analyses of various food products using the Nexera post-column amino acid analysis system, which was recently introduced in the market.

* : Please refer to Application News No. L529B.

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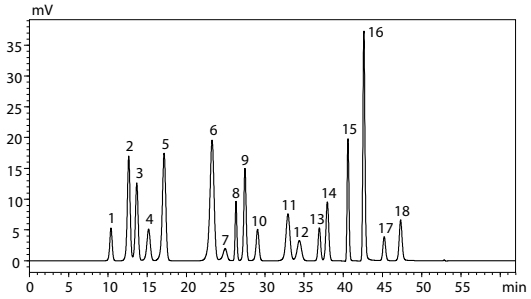
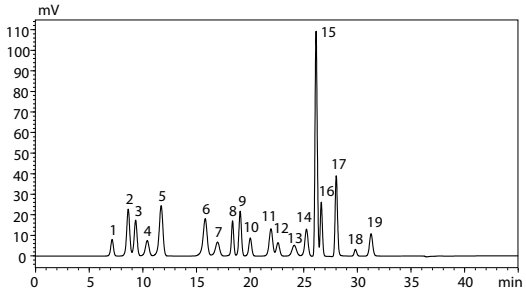
Analysis of Proteinogenic Amino Acids (Na Type) [Standard Sample]

Analysis of proteinogenic amino acids (Na type) enables separation and quantitation of approximately 20 types of amino acids obtained by hydrolyzing protein. The Na type method provides three separation conditions, the high resolution condition for normal analysis, a high speed condition, and a tryptophan included condition. Figs. 1 to 3 show analyses of standard amino acids under these three conditions, and Table 1 shows the analytical conditions of the Na type method.



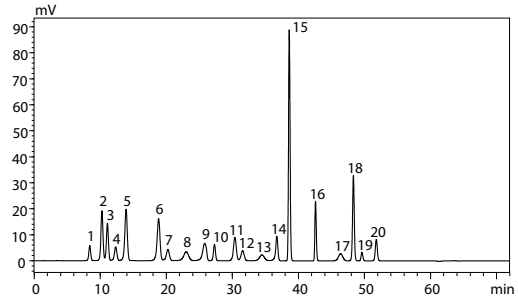
1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. Histidine
16. Lysine
17. Ammonia
18. Arginine

Fig. 1 Analysis of 18 Standard Amino Acids (0.1 µmol/L, Na Type High Resolution Condition)



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. γ –Aminobutyric acid (GABA)
16. Histidine
17. Lysine
18. Ammonia
19. Arginine

Fig. 2 Analysis of 19 Standard Amino Acids (0.1 µmol/L, Na Type High Speed Condition)



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. γ –Aminobutyric acid (GABA)
16. Histidine
17. Tryptophan
18. Lysine
19. Ammonia
20. Arginine

Fig. 3 Analysis of 20 Standard Amino Acids (0.1 µmol/L, Na Type Tryptophan Included Condition)

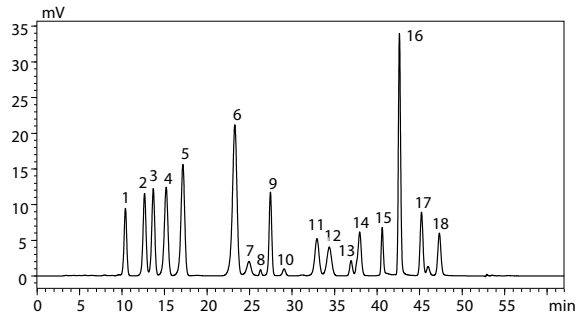
Table 1 Analytical Conditions of Na Type Method (Common)

Column	:	Shim-pack™ Amino-Na (100 mm × 6.0 mm I.D., 5 µm) *1
Ammonia trap column	:	Shim-pack ISC-30/S0504Na (50 mm×4.0 mm I.D.) *2
Mobile phase	:	Amino Acid Mobile Phase Kits (Na type) *3 Gradient Elution
Flow rate	:	0.4-0.6 mL/min
Column temp.	:	60 °C
Injection vol.	:	10 µL
Vial	:	Shimadzu Vials, LC, 1.5 mL, Glass *4
Reaction reagent	:	Amino Acid Reagent Kits *5
Flow rate of reagent	:	0.2 mL/min for each
Reaction temp.	:	60 °C
Detection	:	Ex. 350 nm Em. 450 nm

*1 : P/N S228-18837-91, *2 : P/N S228-14206-91, *3 : P/N S228-21195-94, *4 : P/N S228-15652-92, *5 : P/N S228-21195-93

Analysis under Na Type High Resolution Condition [Hydrochloric Acid Hydrolysis: Soybeans, Liquid Formula, Protein Powder]

The following introduces analyses using the Na type high resolution condition. Figs. 4 to 9 show analyses of 18 proteinogenic amino acids obtained by hydrochloric acid hydrolysis. Among these, 9 essential amino acids (threonine, valine, methionine, isoleucine, leucine, phenylalanine, histidine, tryptophan, lysine) must be obtained from foods, as they cannot be synthesized in the human body.



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. Histidine
16. Lysine
17. Ammonia
18. Arginine

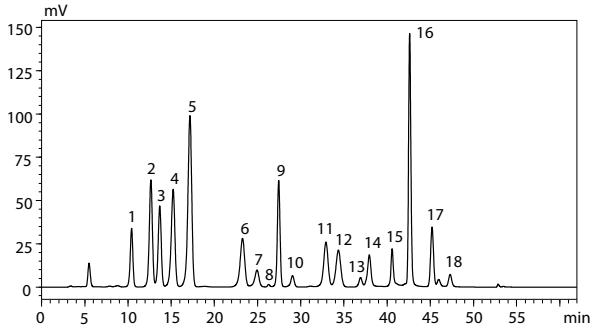
Fig. 4 Analysis of Ground Soybeans (Hydrochloric Acid Hydrolysis) (Na Type High Resolution Condition)

Ground soybeans 5.03 mg
- 6 mol/L HCl (500 µL)
  Seal vessel under reduced pressure
  Stand at 110 °C, 22 hours
  Evaporate to dryness under nitrogen atmosphere
- 0.2 mol/L sodium citrate buffer (pH 2.2) (500 µL)
  Dissolve with vortex mixer
  Filtration (0.45 µm, 10000 rpm × 10 minutes)

Filtrate 50 µL
- 0.2 mol/L sodium citrate buffer (pH 2.2) (950 µL)

HPLC

Fig. 5 Pretreatment Protocol for Ground Soybeans (Hydrochloric Acid Hydrolysis)



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. Histidine
16. Lysine
17. Ammonia
18. Arginine

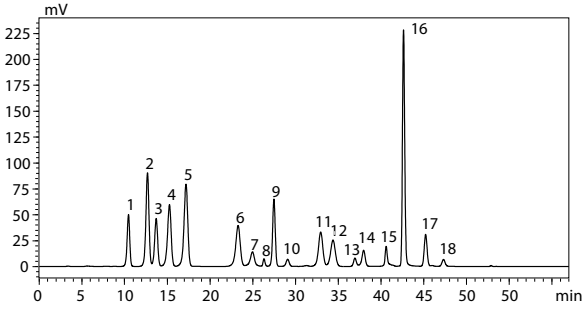
Fig. 6 Analysis of Baby Liquid Formula (Hydrochloric Acid Hydrolysis) (Na Type High Resolution Condition)

Baby liquid formula 250 µL
- 12 mol/L HCl (250 µL)
  Seal vessel under reduced pressure
  Stand at 110 °C, 22 hours
  Evaporate to dryness under nitrogen atmosphere
- 0.2 mol/L sodium citrate buffer (pH 2.2) (500 µL)
  Dissolve with vortex mixer
  Filtration (0.45 µm, 10000 rpm × 10 minutes)

Filtrate 100 µL
- 0.2 mol/L sodium citrate buffer (pH 2.2) (900 µL)

HPLC

Fig. 7 Pretreatment Protocol for Baby Liquid Formula (Hydrochloric Acid Hydrolysis)



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. Histidine
16. Lysine
17. Ammonia
18. Arginine

Fig. 8 Analysis of Protein Powder (Hydrochloric Acid Hydrolysis) (Na Type High Resolution Condition)

Protein powder 5.05 mg
- 6 mol/L HCl (500 µL)
  Seal vessel under reduced pressure
  Stand at 110 °C, 22 hours
  Evaporate to dryness under nitrogen atmosphere
- 0.2 mol/L sodium citrate buffer (pH 2.2) (500 µL)
  Dissolve with vortex mixer
  Filtration (0.45 µm, 10000 rpm × 10 minutes)

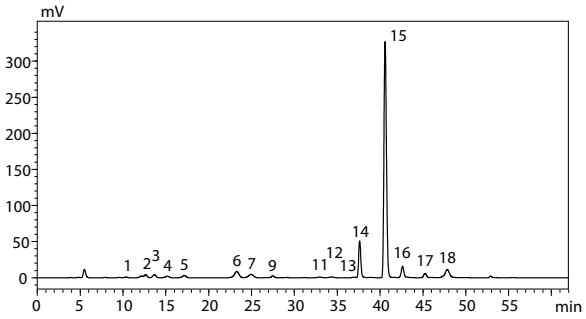
Filtrate

HPLC

Fig. 9 Pretreatment Protocol for Protein Powder (Hydrochloric Acid Hydrolysis)

Analysis under Na Type High Resolution Condition [Chicken Breast Meat, Soy Meat, Nampler (Fish Sauce), Amino Acid Supplement]

Figs. 10 to 17 show analyses of 18 free amino acids obtained by extraction from samples. Here, an ultrafiltration cartridge was used in deproteinization.



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
9. Valine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. Histidine
16. Lysine
17. Ammonia
18. Arginine

Fig. 10 Analysis of Chopped Chicken Breast Meat (Na Type High Resolution Condition)

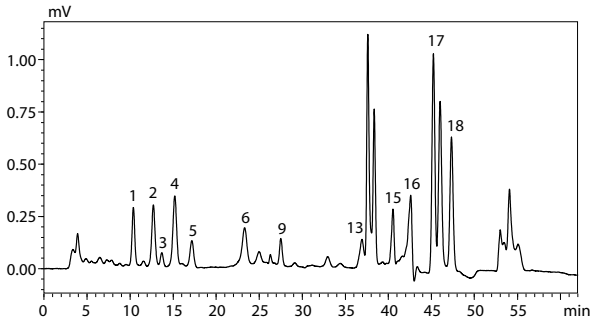
Chopped chicken breast meat 1.668 g
- 0.2 mol/L sodium citrate buffer (pH 2.2) (10 mL)
- Homogenize (1 minute)
- Centrifuge (5000 rpm × 5 minutes)

Supernatant
- Ultrafiltration filter (10K)
- Centrifuge (10000 rpm × 10 minutes)

Filtrate 100 µL
- 0.2 mol/L sodium citrate buffer (pH 2.2) (900 µL)

HPLC

Fig. 11 Pretreatment Protocol for Chopped Chicken Breast Meat



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
9. Valine
13. Tyrosine
15. Histidine
16. Lysine
17. Ammonia
18. Arginine

Fig. 12 Analysis of Dry Soy Meat (Na Type High Resolution Condition)

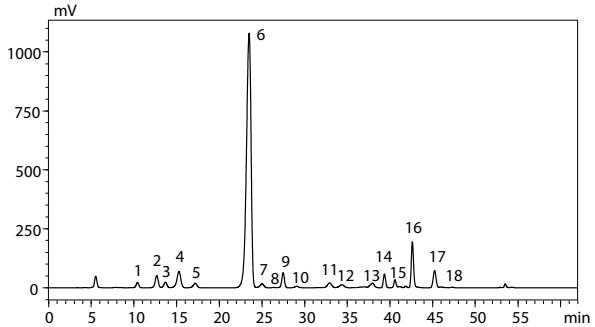
Dry soy meat 109 mg
- 0.2 mol/L sodium citrate buffer (pH 2.2) (20 mL)
- Stand at room temperature, 2 hours
- Homogenize (1 minute)

Supernatant
- Ultrafiltration filter (10K)
- Centrifuge (10000 rpm × 10 minutes)

Filtrate 100 µL
- 0.2 mol/L sodium citrate buffer (pH 2.2) (400 µL)

HPLC

Fig. 13 Pretreatment Protocol for Dry Soy Meat



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. Histidine
16. Lysine
17. Ammonia
18. Arginine

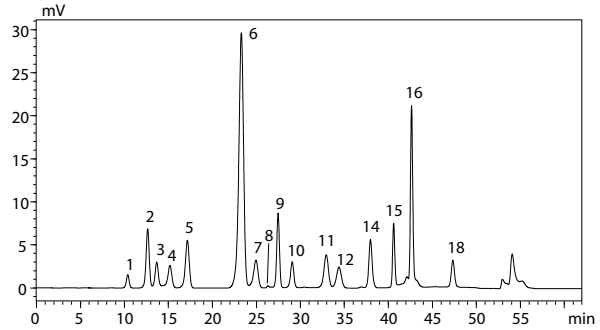
Fig. 14 Analysis of Nampler (Fish Sauce) (Na Type High Resolution Condition)

Nampler (fish sauce) 50 µL
- 0.2 mol/L sodium citrate buffer (pH 2.2) (4.95 mL)
- Ultrafiltration filter (10K)
- Centrifuge (10000 rpm × 10 minutes)

Filtrate

HPLC

Fig. 15 Pretreatment Protocol for Nampler (Fish Sauce)



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
14. Phenylalanine
15. Histidine
16. Lysine
18. Arginine

Fig. 16 Analysis of Amino Acid Supplement (Na Type High Resolution Condition)

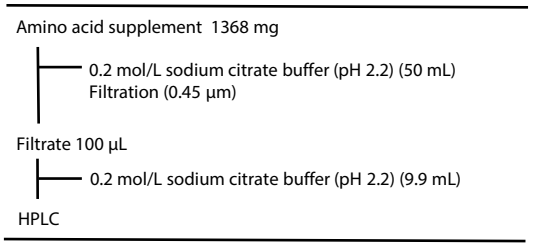
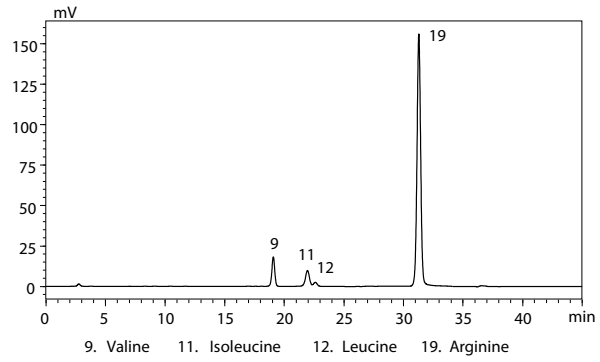


Fig. 17 Pretreatment Protocol for Amino Acid Supplement

Analysis under Na Type High Speed Condition [Sports Drink, Beer, Yogurt]

Figs. 18 to 23 show analyses under the Na type high speed condition. This separation condition has the shortest analysis time (45 min) and can be used with samples that contain comparatively few contaminants.



9. Valine 11. Isoleucine 12. Leucine 19. Arginine

Fig. 18 Analysis of Sports Drink (Na Type High Speed Condition)

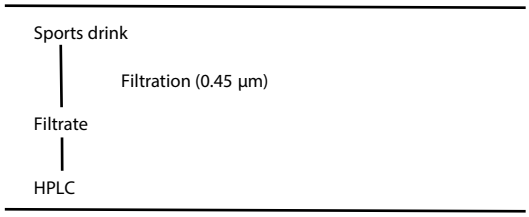
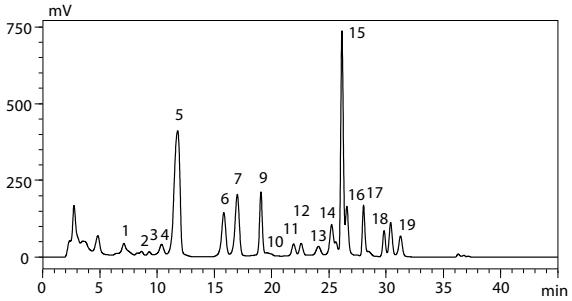


Fig. 19 Pretreatment Protocol for Sports Drink



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. γ –Aminobutyric acid (GABA)
16. Histidine
17. Lysine
18. Ammonia
19. Arginine

Fig. 20 Analysis of Beer (Na Type High Speed Condition)

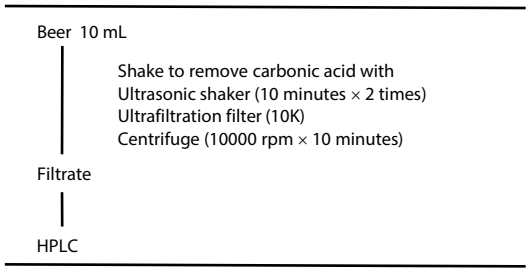
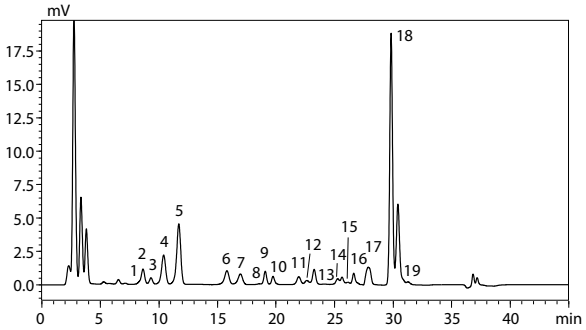


Fig. 21 Pretreatment Protocol for Beer



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. γ –Aminobutyric acid (GABA)
16. Histidine
17. Lysine
18. Ammonia
19. Arginine

Fig. 22 Analysis of Yogurt (Na Type High Speed Condition)

Yogurt 2.074 g
- 0.2 mol/L sodium citrate buffer (pH 2.2) (20 mL)
  Mix with vortex mixer
  Ultrafiltration filter (10K)
  Centrifuge (10000 rpm × 10 minutes)

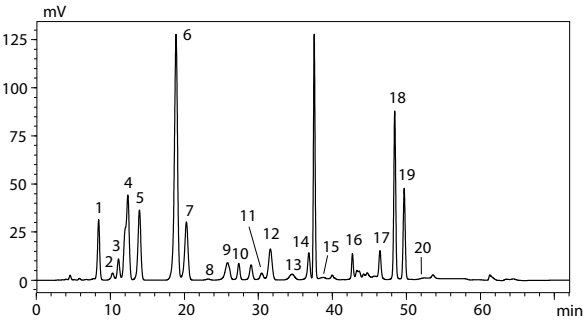
Filtrate

HPLC

Fig. 23 Pretreatment Protocol for Yogurt

Analysis under Na Type Tryptophan Included Condition [Chicken Breast Meat (Alkali Hydrolysis), Vegetable Juice, Miso (Soybean Paste)]

Figs. 24 to 29 show analyses under the Na type tryptophan included condition. Alkali hydrolysis is used when analyzing proteinogenic amino acids containing tryptophan due to the decomposition of tryptophan by acid hydrolysis.



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
8. Cystine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. γ –Aminobutyric acid (GABA)
16. Histidine
17. Tryptophan
18. Lysine
19. Ammonia
20. Arginine

Fig. 24 Analysis of Chopped Chicken Breast Meat (Alkali Hydrolysis) (Na Type Tryptophan Included Condition)

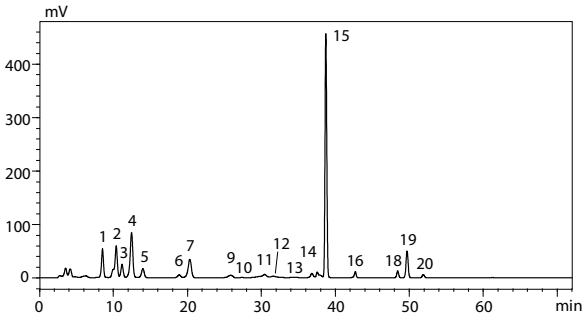
Chopped chicken breast meat 15.16 mg
- 5.2 mol/L NaOH (500 µL)
- 2,2`-Thiodiethanol (100 µL)
  Seal vessel under reduced pressure
  Stand at 110 °C, 22 hours
- Neutrialization with 6 mol/L HCl
- Water

5 mL
- Filtration (0.45 µm, 10000 rpm × 10 minutes)

Filtrate

HPLC

Fig. 25 Pretreatment Protocol for Chopped Chicken Breast Meat (Alkali Hydrolysis)



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. γ –Aminobutyric acid (GABA)
16. Histidine
18. Lysine
19. Ammonia
20. Arginine

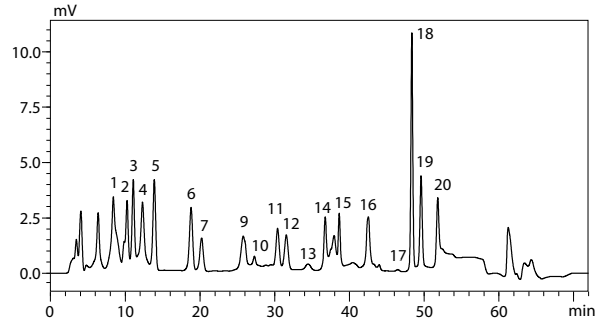
Fig. 26 Analysis of Vegetable Juice (Na Type Tryptophan Included Condition)

Vegetable juice 500 µL
- 0.2 mol/L sodium citrate buffer (pH 2.2) (4.5 mL)
  Ultrafiltration filter (10K)
  Centrifuge (10000 rpm × 15 minutes)

Filtrate

HPLC

Fig. 27 Pretreatment Protocol for Vegetable Juice



1. Aspartic Acid
2. Threonine
3. Serine
4. Glutamic Acid
5. Proline
6. Glycine
7. Alanine
9. Valine
10. Methionine
11. Isoleucine
12. Leucine
13. Tyrosine
14. Phenylalanine
15. γ –Aminobutyric acid (GABA)
16. Histidine
17. Tryptophan
18. Lysine
19. Ammonia
20. Arginine

Fig. 28 Analysis of Miso (Japanese Soybean Paste) (Na Type Tryptophan Included Condition)

Miso (Japanese soybean paste) 2.1971 g
- 0.2 mol/L sodium citrate buffer (pH 2.2) (20 mL)
  Mix with vortex mixer
  Centrifuge (3500 rpm × 3 minutes)

Supernatant
- Ultrafiltration filter (10K)
  Centrifuge (10000 rpm × 15 minutes)

Filtrate 20 µL
- 0.2 mol/L sodium citrate buffer (pH 2.2) (980 µL)

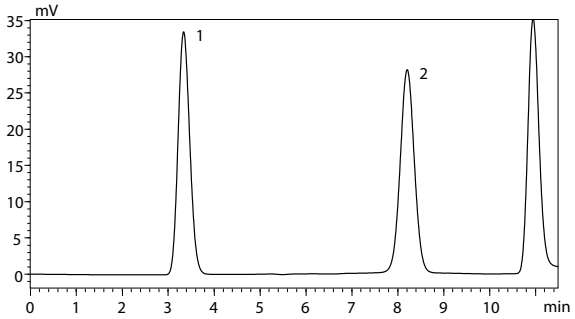
HPLC

Fig. 29 Pretreatment Protocol for Miso (Japanese Soybean Paste)

Analysis of Cysteic Acid and Methionine Sulfone under Na Type Condition [Standard Sample, Chicken Breast Meat (Performic Acid Oxidation-Hydrolysis)]

Cysteine, cystine, and methionine are decomposed by hydrochloric acid hydrolysis. Therefore, in order to measure these compounds, performic acid oxidation-hydrolysis is conducted to convert cysteine and cystine to cysteic acid and methionine to methionine sulfone to be determined in the analysis.

Fig. 30 shows a standard analysis of cysteic acid and methionine sulfone, Fig. 31 shows an analysis of chopped chicken breast meat by performic acid oxidation-hydrolysis, and Table 2 shows the analytical conditions.



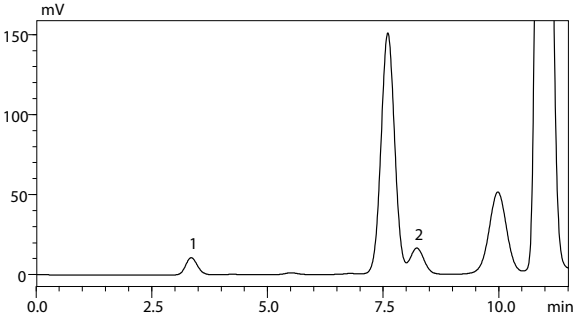
1. Cysteic Acid 2. Methionine sulfone

Fig. 30 Analysis of Standard Sample (Cysteic Acid, Methionine Sulfone, 0.1 mmol/L Each)

Table 2 Analytical Conditions for Cysteic Acid and Methionine Sulfone

Column	: Shim-pack Amino-Na (100 mm × 6.0 mm I.D., 5 µm)
Mobile phase	: A*6) 67 mmol/L sodium citrate containing 7% ethanol and 0.15 mol/L perchloric acid B) 0.2 mol/L NaOH Gradient Elution
Flow rate	: 0.4 mL/min
Column temp.	: 60 °C
Injection vol.	: 10 µL
Reaction reagent	: Amino acid reagent kits not add sodium hypochlorite to reagent A
Flow rate of reagent	: 0.2 mL/min each
Reaction temp.	: 60 °C
Detection	: Ex. 350 nm Em. 450 nm

*6 : Sodium citrate 2-hydrate 19.6 g, ethanol 70 mL, perchloric acid (60%) 14 mL into 1 L of water



1. Cysteic Acid 2. Methionine sulfone

Fig. 31 Analysis of Chopped Chicken Breast Meat (Performic Acid Oxidation-Hydrolysis)

Chopped chicken breast meat 14.87 mg
- Performic acid (800 µL)
  Stand at room temperature, 1 hour
  Store in ice through night
- Hydrobromic acid (100 µL)
  Evaporate to dryness under nitrogen atmosphere
- 6 mol/L HCl (500 µL)
  Seal Vessel under reduced pressure
  Stand at 110 °C, 22 hours
  Evaporate to dryness under nitrogen atmosphere
- 0.2 mol/L Sodium citrate buffer (pH 2.2) (500 µL)
  Dissolve with vortex mixer
  Filtration (0.45 µm, 10000 rpm × 10 minutes)

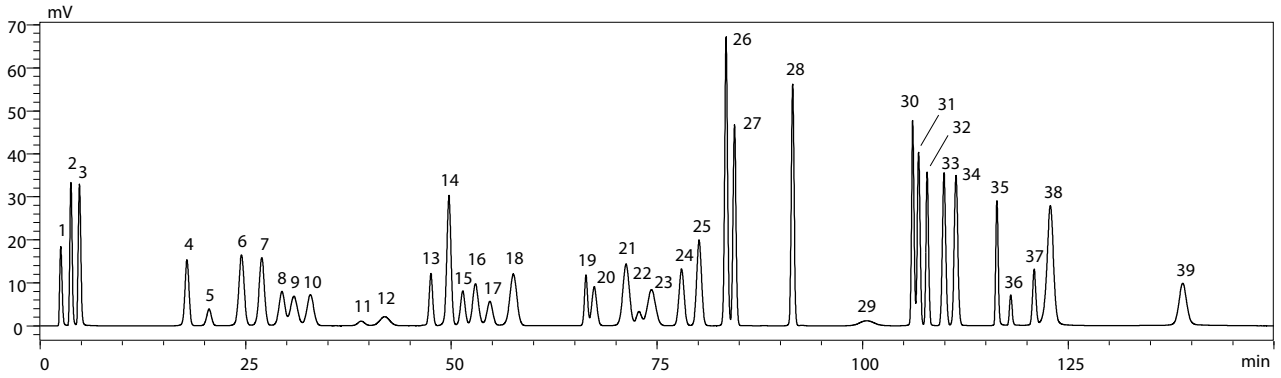
Filtrate 50 µL
- 0.2 mol/L Sodium citrate buffer (pH 2.2) (950 µL)

HPLC

Fig. 32 Pretreatment Protocol for Chopped Chicken Breast Meat (Performic Acid Oxidation-Hydrolysis)

Analysis of Free Amino Acids (Li Type) [Standard Sample, Ham, Black Vinegar, Broccoli Sprout]

In free amino acid analysis (Li type), separation and quantitation of approximately 40 amino acids are possible. If the reagent kit is used, the 150 minutes analysis is conducted easily with good repeatability. Fig. 33 shows a standard analysis by the Li type method, and Table 3 shows the Li type analytical conditions. Figs. 34 to 39 are analyses of free amino acids by the Li type method. High resolution is possible with the Li type method, even with samples such as foods that contain many contaminants.



1. o-Phosphoserine
2. Taurine
3. o-Phosphoethanolamine
4. Aspartic acid
5. Hydroxyproline
6. Threonine
7. Serine
8. Asparagine
9. Glutamic acid
10. Glutamine
11. Sarcosine
12. α-Aminoadipic acid
13. Proline
14. Glycine
15. Alanine
16. Citrulline
17. α-Amino-n-butyric acid
18. Valine
19. Cystine
20. Methionine
21. Isoleucine
22. Cystathionine
23. Leucine
24. Tyrosine
25. Phenylalanine
26. β-Alanine
27. β-Aminoisobutyric acid
28. γ –Aminobutyric acid (GABA)
29. Tryptophan
30. Histidine
31. 3-Methylhistidine
32. 1-Methylhistidine
33. Carnosine
34. Anserine
35. Hydroxylysine
36. Ornithine
37. Lysine
38. Ammonia
39. Arginine

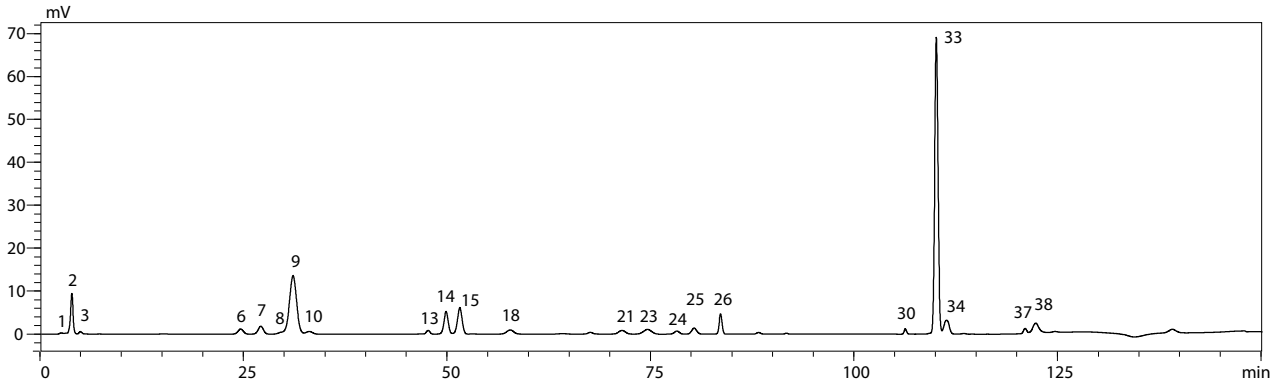
Fig. 33 Analysis of 39 Standard Amino Acids (0.1 mmol/L Each, Li Type Condition)**

** : However, the concentrations are 0.05 mmol/L for components 1-3, 12, 17, and 22, 0.25 mmol/L for component 11, and 0.026 mmol/L for component 29.

Table 3 Analytical Conditions of Li Type Method

Column	: Shim-pack Amino-Li (100 mm × 6.0 mm I.D., 5 µm) *7
Ammonia trap column	: Shim-pack ISC-30/S0504Li (50 mm × 4.0 mm I.D.) *8
Mobile phase	: Amino acid mobile phase kits (Li type) *9 Gradient elution
Flow rate	: 0.6 mL/min
Column temp.	: 39 °C
Injection vol.	: 10 µL
Vial	: Shimadzu Vials, LC, 1.5 mL, Glass
Reaction reagent	: Amino acid reagent kits
Flow rate of reagent	: 0.2 mL/min for each
Reaction temp.	: 39 °C
Detection	: Ex. 350 nm Em. 450 nm

*7 : P/N S228-18837-92, *8 : P/N S228-00821-91, *9 : P/N S228-21195-95



1. o-Phosphoserine
2. Taurine
3. o-Phosphoethanolamine
6. Threonine
7. Serine
8. Asparagine
9. Glutamic acid
10. Glutamine
13. Proline
14. Glycine
15. Alanine
18. Valine
21. Isoleucine
23. Leucine
24. Tyrosine
25. Phenylalanine
26. β-Alanine
30. Histidine
33. Carnosine
34. Anserine
37. Lysine
38. Ammonia

Fig. 34 Analysis of Ham (Li Type Condition)

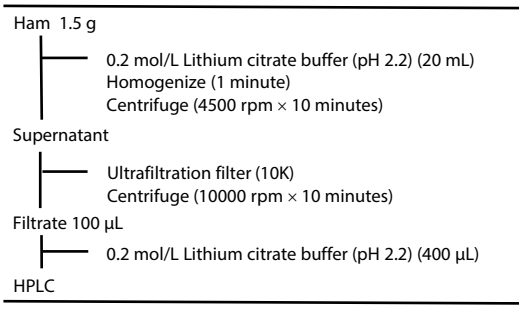
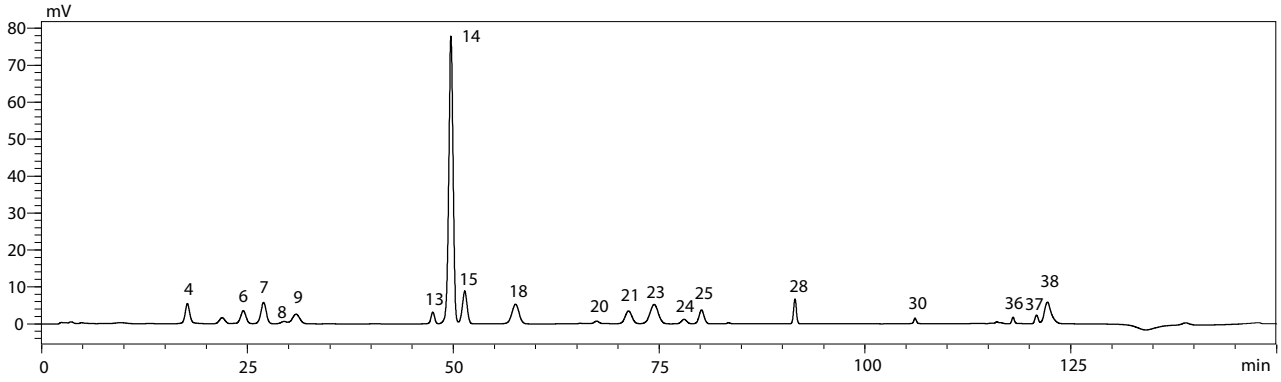


Fig. 35 Pretreatment Protocol for Ham



4. Aspartic acid
6. Threonine
7. Serine
8. Asparagine
9. Glutamic acid
13. Proline
14. Glycine
15. Alanine
18. Valine
20. Methionine
21. Isoleucine
23. Leucine
24. Tyrosine
25. Phenylalanine
28. γ –Aminobutyric acid (GABA)
30. Histidine
30. Histidine
36. Ornithine
37. Lysine
38. Ammonia

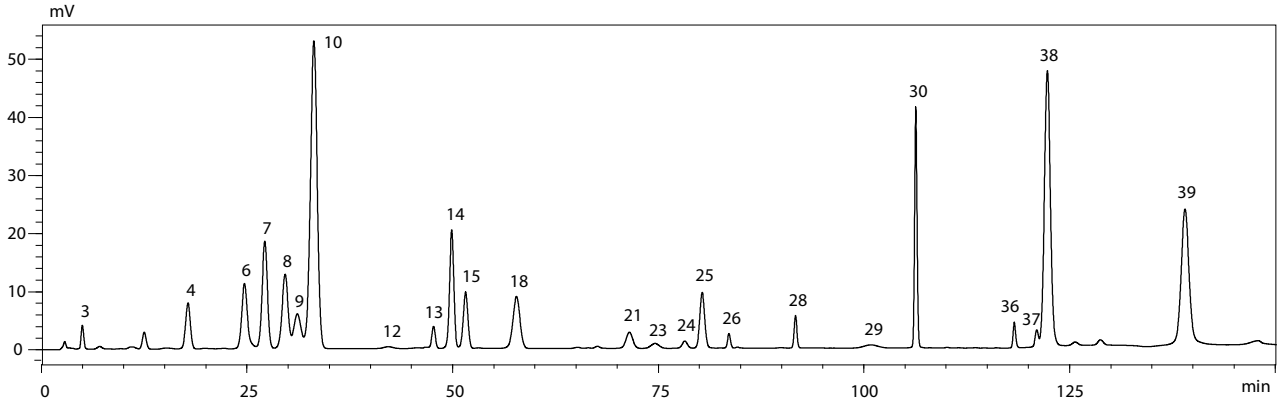
Fig. 36 Analysis of Black Vinegar (Li Type Condition)

Black vinegar 50 µL
- 0.2 mol/L Lithium citrate buffer (pH 2.2) (4.95 mL)
- Ultrafiltration filter (10K)
- Centrifuge (10000 rpm × 10 minutes)

Filtrate

HPLC

Fig. 37 Pretreatment Protocol for Black Vinegar



3. o-Phosphoethanolamine
4. Aspartic acid
6. Threonine
7. Serine
8. Asparagine
9. Glutamic acid
10. Glutamine
12. α-Aminoadipic acid
13. Proline
14. Glycine
15. Alanine
18. Valine
21. Isoleucine
23. Leucine
24. Tyrosine
25. Phenylalanine
26. β-Alanine
28. γ –Aminobutyric acid (GABA)
29. Tryptophan
30. Histidine
36. Ornithine
37. Lysine
38. Ammonia
39. Arginine

Fig. 38 Analysis of Broccoli Sprout (Li Type Condition)

Broccoli sprout 0.548 g
- 0.2 mol/L Lithium citrate buffer (pH 2.2) (15 mL)
- Homogenize (1 minute)
- Centrifuge (4500 rpm × 1 minute)

Supernatant

Filtration (0.45 µm)

HPLC

Fig. 39 Pretreatment Protocol for Broccoli Sprout

Conclusion

Analyses of various food products using the Nexera post-column amino acid analysis system were introduced.

It should be noted that the pretreatment protocols used here are just examples. When actually conducting a quantitative analysis, confirmation of the recovery rate by a recovery test prior to the analysis is recommended.

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