

SUBJECT: ANALYSIS OF DESMOSINE AND ISODESMOSINE

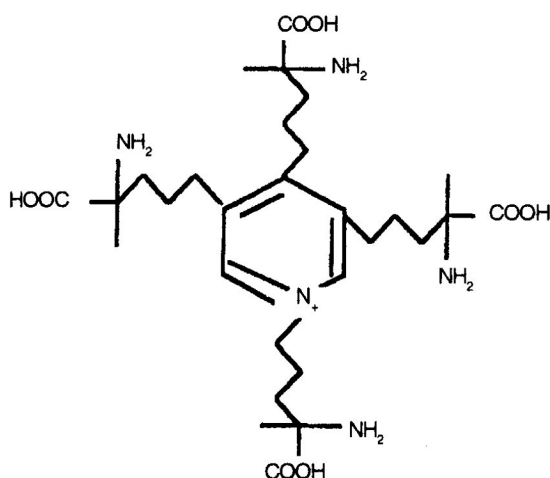
INSTRUMENT: HITACHI MODEL L-8800 AMINO ACID ANALYZER

1. Introduction

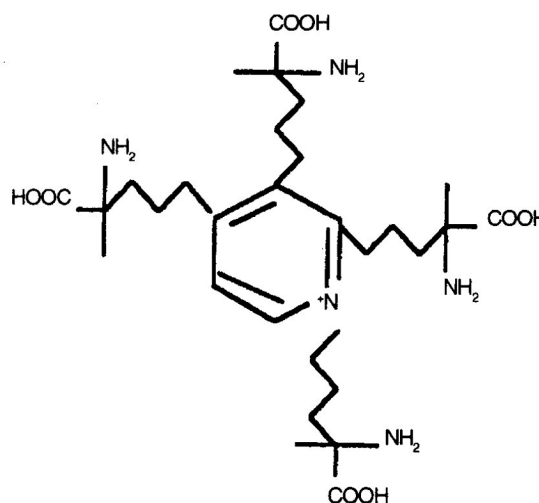
Desmosine and isodesmosine are known as unique amino acids present in elastin (protein which composes skin, ligament, etc.), and they are said to play a role of crosslink in collagen, thereby affecting the elasticity of skin. In the conventional physiological-fluid amino acid analysis, their retention times would overlap that of ornithine (Orn), disabling analysts from obtaining an adequate separation for quantitative analysis. Therefore, desmosine and isodesmosine have been analyzed by the OPA pre-column

labeling method using a reversed phase column.

However, the new Model L-8800 amino acid analyzer equipped with a semi-micro pump and a high-accuracy gradient system is now capable of separating these amino acids from other physiological-fluid amino acids through a revision of the eluent changeover time program, thus allowing their simultaneous quantitative analysis. An example of this analysis is introduced here.



Desmosine



Isodesmosine

Fig. 1 Structure of Desmosine and Isodesmosine

Sheet No.	Measured Substance	Field
59	Desmosine and isodesmosine	Biochemistry

Features

- Desmosine and isodesmosine are separable from other physiological-fluid amino acids.
- The commercially available buffer solution for L-8500 is usable as is.

Example of Measurement

Analytical conditions

1. Column : #2622 (4.6 × 80 mm)
2. Ammonia filter column : 2650L (4.6 × 60 mm)
3. Pump flow rate p1 : 0.35 mL/min
p2 : 0.30 mL/min
4. Reaction method : Reaction column (4.6 × 40 mm)

Program

Time (min)	%B1	%B2	%B3	%B4	%B5	Temperature (°C)
0.0	100	0	0	0	0	38
3.0	100	0	0	0	0	30
25.4	100	0	0	0	0	
25.5	80	20	0	0	0	60
40.5	60	40	0	0	0	
40.6	10	90	0	0	0	
48.6	10	90	0	0	0	40
58.0	10	90	0	0	0	
58.1	0	100	0	0	0	
64.5	0	100	0	0	0	70
64.6	0	0	100	0	0	
91.0	0	0	100	0	0	45
91.1	0	0	100	0	0	
91.2	60	0	0	40	0	
98.5	60	0	0	40	0	
98.6	0	0	0	100	0	
114.5	0	0	0	100	0	
114.6	0	20	0	80	0	
123.0	0	20	0	80	0	70
132.9	0	20	0	80	0	
133.0	0	0	0	100	0	
146.0	0	0	0	100	0	
146.1	0	0	0	0	100	
158.0	0	0	0	0	100	
158.1	100	0	0	0	0	
167.0	100	0	0	0	0	38
200.0	100	0	0	0	0	

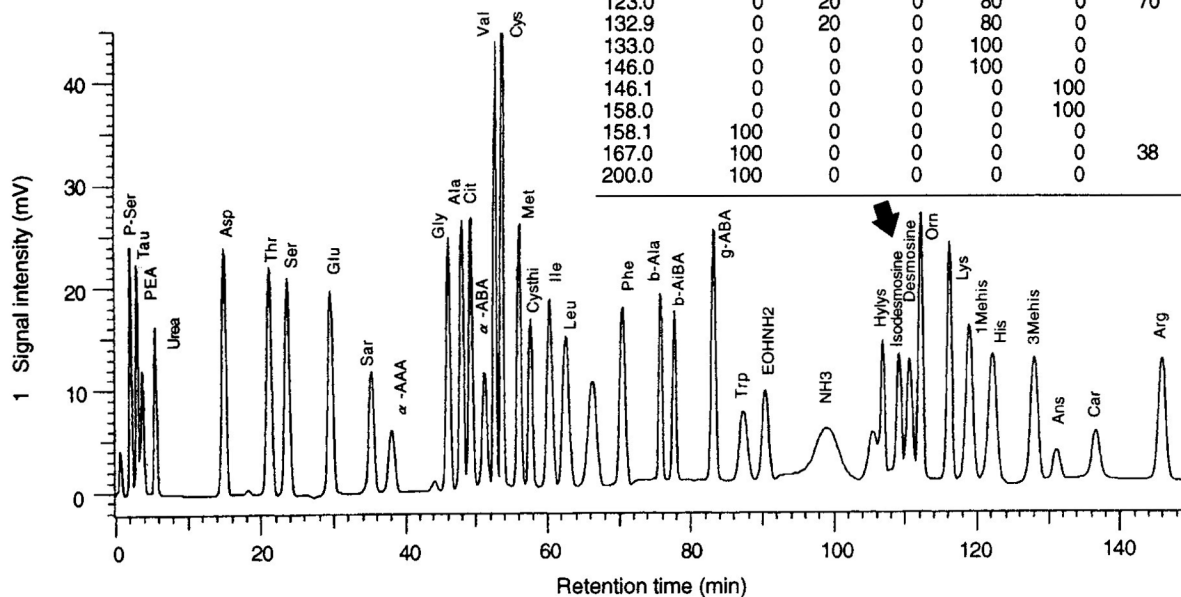


Fig. 2 Chromatogram of Desmosine and Isodesmosine

Keywords : Physiological fluid analysis, Free amino acid, Desmosine, Isodesmosine

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