## **Analysis of Capsaicinoids in Spices**

When absorbed by the body, capsaicins, the spice element found in chili pepper, has the effect of stimulating adrenaline secretion and causing perspiration. Since ancient times, capsaicins have been known for their anti-bacterial, stomachic, and body-warming properties. Furthermore, capsaicins are very stable elements that maintain their spiciness even when used in a variety of cooking methods.

The unit used to indicate the spiciness of a spice is

known as the "Scoville Scale". This scale is indicated in magnitudes based on the amount of time until a taster no longer feels the spiciness of a spice extract dissolved in sugar water. In recent years, to gain more objective indicators, quantitative methods that use HPLC to analyze the capsaicinoids in a spice also are in use.

This issue introduces examples of the analysis of capsaicinoids in spice using HPLC.

## ■ Analysis of Standard Solution

Known capsaicinoids include capsaicin, dihydrocapsaicin, and nordihydrocapsaisin. (Fig.1)

Fig.2 shows the analytical results of capsaisin and dihydrocapsaicin standard solutions (each at 50 mg/L, ethanol solution). Table 1 shows the analytical conditions.

Fig.1 Structure of Capsaicinoids

## Table 1 Analytical Conditions

 $\label{eq:column} \textbf{Column} \qquad : \textbf{Shim-pack} \ \ \textbf{VP-ODS} \ (150 \ \text{mL.} \times 4.6 \ \text{mm I.D.})$ 

Mobile Phase : 1.0% Acetic Acid aq./Acetonitorile

=1/1(v/v)

Flow Rate : 1.2 mL/min Column Temp. : 40 °C

Detection : SPD-20 A at 280 nm

RF-10AxL Ex at 280 nm, Em at 325 nm

Inj. Volume : 1 μL

Detection was conducting using a UV absorption detector and a spectrofluorometric detector.

\*We could not obtain a reference solution of nordihydrocapsaisin, however, based on reference materials, it is assumed this is the peak (arrow) immediately preceding capsaisin by literature data.

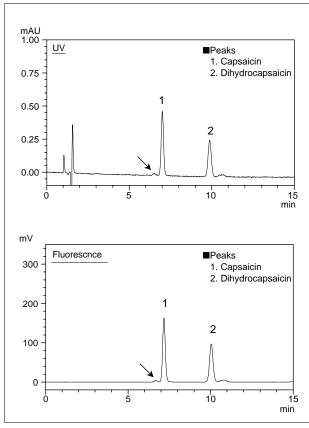


Fig.2 Chromatogram of a Standard Mixture of Capsaicin and Dihydrocapsaicin (50 mg/L, 1  $\mu$ L, injected)

## ■ Analysis of Spice

Fig.3 shows the sample preparation steps used on the commercial spices and Figs.4 – 6 shows the results of the analysis.

With the actual samples, because impurity substances are eluted out after the dihydrocapsaicin, in order to remove those substances from the column we recommend cleansing the column by flowing the mobile phase with increased level of acetonitrile before each analysis.

 $\star \text{The peak}$  indicated by the arrow in the chromatogram is assumed to be nordihydrocapsaicin.

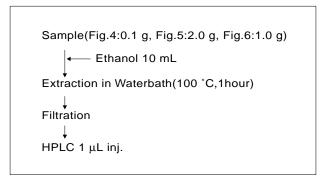


Fig.3 Sample Preparation

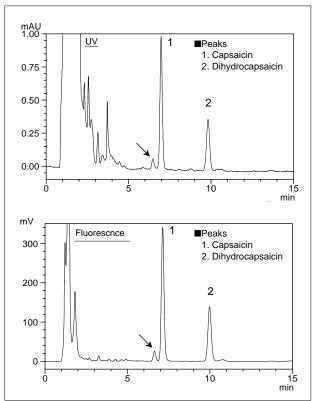


Fig.5 Chromatogram of Pepper Sauce

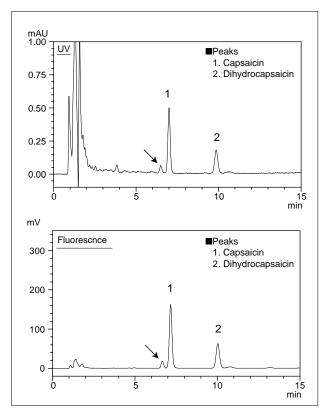


Fig.4 Chromatogram of Red Pepper

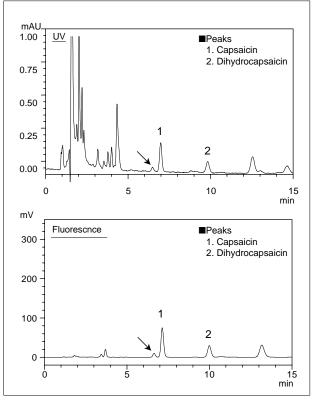


Fig.6 Chromatogram of Spicy Oil

[Reference Literature] AOAC Official Method 955.03

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