Study of Capsicum Frutescens L. by Fourier Transform Infrared Spectroscopy

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Abstract Fourier transform infrared spectroscopy (FTIR) combined with principal component analysis (PCA) and hierarchical cluster analysis (HCA) is used to study different kinds of capsicum frutescens L.. FTIR spectra of 50 samples are obtained from five species of capsicum frutescens L.. The results show that the infrared spectra of capsicum frutescens L. are similar, but tiny differences in wave-numbers and absorption intensities of peaks are observed in the range of 1800～800 cm^{-1}. PCA and HCA are performed on the second derivative infrared spectra in this range. FTIR spectroscopy is found to be efficient in classification of capsicum frutescens L.. 50 samples are well divided into five groups and about 100% classification accuracy for HCA, about 98% accuracy for PCA are yielded. It is proved that FTIR combined with statistical analysis can be used to discriminate capsicum frutescens L..

Key words spectroscopy; Fourier transform infrared spectroscopy; principal component analysis; hierarchical cluster analysis; capsicum frutescens L.

OCIS codes 140.3070; 300.6300; 300.6340; 300.6360

辣椒的傅里叶变换红外光谱研究

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摘要 利用傅里叶变换红外(FTIR)光谱结合主成份分析(PCA)和系统聚类分析(HCA)技术对不同品种辣椒进行鉴别研究。测试了 5 种辣椒共 50 个样品的红外光谱,结果表明,5 个不同品种辣椒的红外光谱相似,但在 1800～800 cm^{-1}范围内红外光谱的峰位、峰形及吸收强度有一些微小差异。对原始光谱作二阶导数处理,发现在该范围内 5 种辣椒的二阶导数光谱差异明显,利用该范围二阶导数光谱数据对 5 个品种 50 个样品进行聚类和主成分分析。聚类分析正确率为 100%,主成分分析正确率达 98%,能把 5 个品种的辣椒分开。研究结果表明:傅里叶变换红外光谱技术结合统计分析的方法可以把不同品种的辣椒区分开。

关键词 光谱学；傅里叶变换红外光谱；主成分分析；聚类分析；辣椒

中图分类号 O657.3 **文献标识码** A **doi**：10.3788/CJL201441.s115001

1 Introduction

Capsicum frutescens L. include chili and pepper, also called chilly, hot pepper, which is a kind of plants belonging to the genus solanaceae chili. Capsicum frutescens L. came from Mexico, and the Ming dynasty introduced into China. Capsicum frutescens L. is one year or perennial herbaceous plants. The fruits are usually oblong, green when immature, mature into bright red and yellow, but red being the most common. Capsicum frutescens L. is piquancy because the fruit peel containing capsaicinoid. Vitamin C content in chili ranks first in the vegetables. The main bio-active components of capsicum frutescens L., range from protein, fat, carbohydrate, calcium, phosphorus, iron, carotene, vitamin C, thiamine, riboflavin, nicotinic acid, malic acid, citric acid and capsicum red pigment, etc. Capsicum frutescens L. has strong bioactivity such as antitumor, modulating immunity, protecting liver and so forth[1-2]. Capsicum frutescens L. is highly regarded in medicinal value, it is believed to promote

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health and reduce the risk of cancer and heart disease. Presently, many extract of capsicum frutescens L. products are widely used, such as paprika and capsaicinoid. Paprika is one of the most widely used food colourants for culinary and industrial purposes. Because of its high colouring capacity, and in some cases its peculiar pungency, paprika is used to modify the colour and flavour of soups, stews, sausage, cheese, snacks, salad dressing, sauces, pizza and confectionary products. Capsaicinoids have strong physiologicaland pharmacological properties, which may be used in treating sensory nerve fibre disorders, including arthritis, cystitis, and human immunodeficiency virus among others. It is widely used in medicine, light industry and food. Therefore, the identification and classification of capsicum frutescens L. are the basis of the development and utilization of hot pepper resources.

There are many different kinds of Capsicum frutescens L.. It is difficult to distinguish them because they have little differences in appearance obviously. Conventional chemical analysis method include high performance liquid chromatography (HPTLC), gas chromatography mass spectrometry (GC-MS), microsatellite marker, supercritical CO_2 extraction method, etc. But it always takes long time, high investment, cause some damages and also need specialist to do it. Infrared (IR) spectroscopy techniques, such as mid-infrared spectroscopy, offer a quick analysis, non-destructive and low cost method for food analysis[3]. Such as identification of goat's milk[4], edible oil[5], ganoderma lucidum[6], etc. There are less studies reported in the literature using infrared spectroscopy to analysis capsicum frutescens L.. In this paper, Fourier transform infrared spectroscopy (FTIR) combined with hierarchical cluster analysis (HCA) and principal component analysis (PCA) is used to study the capsicum frutescens L., which provides a quick and accurate vibration spectrum identification method for different varieties of capsicum frutescens L..

2 Materials and Methods

2.1 Equipment

The IR spectra are detected on a FTIR spectrometer (PerkinElmer, UK), equipped with a DTGS detector in the region of 400～4000 cm^{-1} at a resolution of 4 cm^{-1}. Each spectrum is acquired by adding 16 scans together.

2.2 Samples preparation and spectral data preprocessing

Five different varieties of capsicum frutescens L. (capsicum frutescens var., capsicum annuum L. var. longum Sendt, line pepper, goat horn pepper, screw pepper) are collected from Hunan Academy of Agricultural Science and dried for measuring spectra. The dried samples are mixed with KBr powder and pressed into discs, and then examined by FTIR. The FTIR spetra of 50 samples are collected. Reference spectra are acquired by collecting a spectrum of pure KBr disc. These spectra are subtracted from reference spectrum of KBr. Spectral processing software (OMNIC 8.0) is used for preprocessing, which has been used for baseline correction, normalization and smoothing infrared spectrum preprocessing. To increase the resolution of obtained spectra, second derivatives (Savitzky-Golay algorithm, 7-point smoothing) are calculated. The statistical software (SPSS19.0) is used for HCA and PCA.

3 Results and Analysis

3.1 Spectral characteristics of capsicum frutescens L.

Fig. 1 shows the infrared spectra of capsicum frutescens var., capsicum annuum L. var. longum Sendt, line pepper, goat horn pepper and screw pepper. It can be seen that the main spectral peak of five different capsicum frutescens L. are similar. The strongest broad peak at about 3400 cm^{-1} is attributed to O-H stretching and contribution of N-H stretching; vibration 2925 cm^{-1} is mainly assigned to CH_2 antisymmetric stretching vibration[4,7]; 2857 cm^{-1} is mainly assigned to CH_2 symmetric stretching vibration[7]; near 1735 cm^{-1} come mainly from lipid absorption peak of C=O stretching vibration[8]; around 1635 cm^{-1} is C=C bond stretching vibration in capsaicinoid.

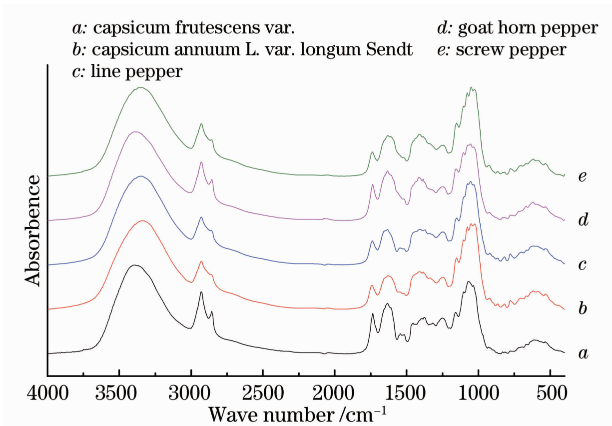

Fig. 1 FTIR spectra of different capsicum frutescens L.

1655 cm^{-1} is assigned to amide band I and 1541 cm^{-1} is assigned to amide band Ⅱ[8]. The bands at 1610 cm^{-1} and 1516 cm^{-1} are benzene ring skeleton stretching vibrations[10]. The range of 1440～1330 cm^{-1} mainly comes from CH_3 and CH_2 symmetric bending vibration absorption in protein, cellulose and lignin affected by oxygen, nitrogen atom[7,11]. Around 1386 cm^{-1} are methyl, methylene and methyl symmetric bending vibrations in protein and cellulose[12]. The range of 1156～950 cm^{-1} are C-O and C-C stretching vibrations of carbohydrate[7,13]. The range of 900～750 cm^{-1} is carbohydrate isomers absorption area, around 895 cm^{-1} C-H is the fiber ring vibration deformation peak[14].

3.2 Analysis of second derivative spectroscopy

The spectra of the samples are similar, only with minor differences in absorption intensity of several peaks. Second derivative spectra of FTIR can obviously enhance the spectral resolution and amplify small differences. The second derivative spectra of the samples are shown in Fig.2. The band at 1635 cm^{-1} in the capsicum frutescens var., capsicum annuum L. var. longum Sendt and goat horn pepper is visible but weak in other spectra. The band at 1034 cm^{-1} is very obvious in the capsicum frutescens var., capsicum annuum L. var. longum Sendt and screw pepper but does not appear in line pepper and almost can't be seen in goat horn pepper.

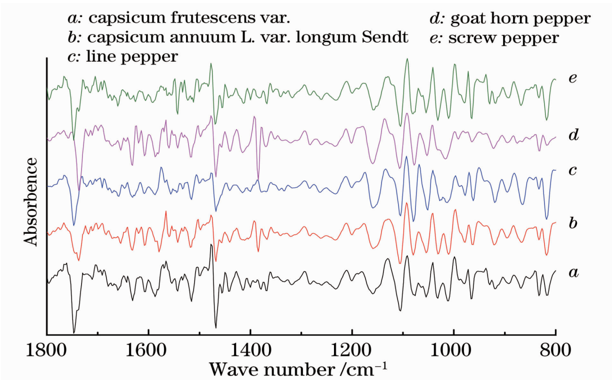

Fig.2 Second derivative FTIR spectra (1800～800 cm^{-1}) of different capsicum frutescens L.

3.3 Hierarchical cluster analysis

Clustering analysis is based on the study of characteristics of object according to certain standards, an analysis method of classifying the research objects, it makes the data objects in the group with the highest similarity, which has great differences between groups. Clustering analysis is performed using Pearson's product moment correlation coefficient as a measure of similarity between the spectra, and Ward's algorithm to draw the dendrogram. The squared Euclidean distance calculation method is used for clustering analysis on second derivative spectrum in the range of 1800～800 cm^{-1}.

The HCA diagram of the samples are shown in Fig.3. Each of the capsicum frutescens L. genera examined in the study formes a separate group. The shortest distances between spectra of capsicum frutescens L. of a given genus are recorded within capsicum pepper and screw pepper, which indicate their highest similarity. All the samples are separated, and the clustering accuracy rate of 50 samples reach 100%.

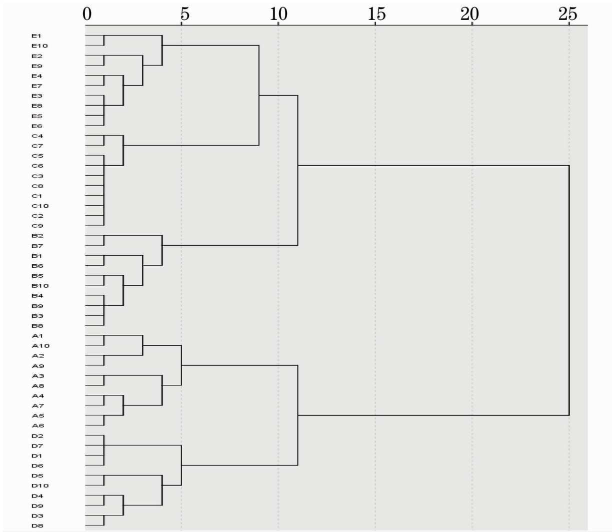

Fig.3 HCA diagram with second derivative infrared spectra of five species of capsicum frutescens L. (1800～800 cm^{-1}, A: capsicum frutescens var., B: capsicum annuum L. var. longum Sendt, C: line pepper, D: goat horn pepper, E: screw pepper)

3.4 Principal component analysis

PCA is an unsupervised pattern recognition and it is often the first step of exploratory data analysis to detect groups in the measured data. PCA models the directions of maximum variations in a data set by projecting as a swarm of points in a space defined by principal components (PCs). PCs describe, in decreasing order, the higher variations among the objects, and because they are calculated to be orthogonal to another one, each PC can be interpreted independently.

The second derivative spectrum in the range of 1800～700 cm^{-1} are selected for principal component analysis. The contribution rate of the frist principal component (PC1) is 84.67%, while that of PC2 is 6.34% and PC3 is 3.45%, the totally contribution rate of these three principal component reaches 96.46%. Therefore, these three principal components could be selected for identification on samples, and the results are shown in Fig.4. 50 capsicum frutescens L. samples are divided into five areas, only one sample is error classified. However, all the samples can be divided into five groups using PC1, PC2 and PC3 with the accuracy of 98%. It demonstrates that the first three principal components have a good clustering effect to five kinds of capsicum frutescens L..

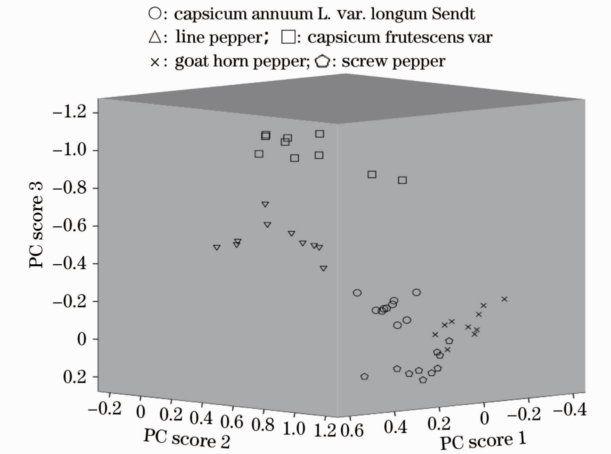

Fig.4 PCA score plots of five capsicum frutescens L. on the second derivative spectra in the range of 1800～800 cm^{-1}

4 Conclusion

FTIR combined with HCA and PCA is used to study five species of capsicum frutescens L. PCA and HCA are performed on the second derivative infrared spectra in the range of 1800～800 cm^{-1}. The clustering accuracy rate of samples is 100% and the principal component analysis accuracy rate of samples is 98%. The results show that FTIR may be used to identify capsicum frutescens L. as it is rapid, nondestructive, convenient and accurate.

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