Raman Spectra from Pesticides on the Surface of Fruits

P.X. Zhang\textsuperscript{1,2} Xiaofang Zhou\textsuperscript{1,2} Andrew Y. S. Cheng\textsuperscript{3} Yan Fang\textsuperscript{2,1}

\textsuperscript{1} IAMPE, Kunming University of Science and Technology, Kunming, Yunnan, China 650051
\textsuperscript{2} NPNS, Beijing Key Lab, Capital Normal University, Beijing, China 100037
\textsuperscript{3} Department of Physics and Materials Science, City University of Hong Kong, Hong Kong, SAR China

Abstract. Raman spectra of several vegetables and fruits were studied by micro-Raman spectrometer (514.5 nm) and Near-infrared Fourier Transform Raman spectrometer (FT-Raman). It is shown that at 514.5 nm excitation, most of the spectra are from that of carotene with some very strong fluorescence in some cases. While at 1064 nm wavelength excitation, the spectra from the different samples demonstrate different characteristic Raman spectra without fluorescence. We discuss the spectroscopic difference by the two excitation wavelengths, and the application of Raman spectra for detection of pesticides left on the surface of vegetables and fruits. Raman spectra of fruits and pesticides were successfully recorded, and using the FT-Raman spectra the pesticides left on the surface of the fruits can be detected conveniently.

1. Introduction

Along with improvements in living standards, the qualities of food are of more concerns today. Large quantities of pesticides are used in fruits production making it unsafe and endangering our health. The currently analyze methods used to detect the residual pesticides on the surface of fruits include Gas Chromatography (GC), High Performance Liquid Chromatography (HPLC), Gas Chromatography-Mass Spectrum (GC-MS), Capillary Electrophoresis (CE), Enzyme Linked Immunity Saturation Adsorption (ELISA) and etc. However, these methods have a lot of weaknesses. One prime inconvenience is that for each kind of examined pesticide usually a specific method has to be adopted. As a result, one has to have prior knowledge of the kind of pesticide to be examined in advance. Secondly, the organic solution used in the manipulation is usually also toxic, which is also not environmentally friendly. Moreover, the process of sample preparation is inconvenience. The samples under examination are damaged and have to be treated, due to the poor sensitivity or separations required for several pesticides. Thus these methods usually requires a much longer time. On the other hand, Raman technique demonstrates wonderful features in trace amount analyses \cite{1-6}: high sensitivity, short time, multi-species, non-destructive and does not required much sample preparation etc. The most important advantage is the examination can be performed on the spot and at the same time several pesticide species can be detected.

In this paper we report a new attempt using Raman technique to detect the residual pesticides on the surface of fruits. For that, two aspects of research work are involved. Firstly, one has to establish a
unique database to store the Raman spectra of the vegetables and fruits, as well as all the frequently used pesticides, respectively. In order to study the suitability of such method, we studied the Raman spectra of several clean fruits as well as fruits with pesticides at 514.5 and 1064 nm excitation wavelengths. It turns out that Raman spectral features of vegetables and fruits depend on the excitation wavelength. At 514.5 nm excitation, most of Raman spectra of fruits exhibit the features of carotene and with strong fluorescence. But at 1064 nm excitation, the Raman spectra recorded have characteristic peaks that can be used for pesticides identifications and without strong interference from fluorescence.

2. Experiments
We first measure Raman spectra from clean fruits and pesticides respectively, and these spectra are used as standards in the comparison database. Simulation of field spray of pesticides on fruits is performed by plating certain amount of pesticides on the fruits. These fruits with pesticides are measured and the Raman spectra obtained are compared with those in the database to identify the trace amount of pesticides on the fruit surfaces.

2.1. Samples
The fruit samples in the experiment are purchased from the market directly. The fruits are pear, grapes, apple, longan, spicy-pear, Chinese gooseberry (kivi), orange, banana etc. The pesticides used in the experiment are Abameclin Petroleumoil, Chongmanjue, Cypermethrin and Zhibaoboshi.

Before making the Raman spectrum, we clean every sample carefully. Firstly, flushing several times with the fresh water to get rid of the dirt left on the surface, and then wipe with cotton and alcohol.

Taking small pieces of peels from the cleaned fruits, and add a sort of pesticide on its surface with the same amount as in field spray. Afterwards filter papers are used to wipe off any left over pesticides from the peels, and let the samples to dry naturally before taking the Raman spectra.

2.2. Instruments and Operation
The major instruments used in this paper are RM2000 micro-Raman spectrometer and an 514.5 nm Ar$^+$ ion laser for Raman excitation. The laser power is set to its minimum of approximately 2 mW, so that the heating effect to the fruits from the laser can be kept to a negligible level. Signals are integrated for 10 s, and 5 individual measured spectra are co-added to give the final spectrum.

Another set of apparatus used in the experiment is RFS100 FT-Raman spectrometer together with a laser at the excitation wavelength of 1064 nm with a power of 100 mW.

3. Results
3.1. Raman spectra of fruits at excitation wavelengths of 514.5 and 1064 nm
Under the same conditions mentioned above, we measured Raman spectra of different fruits at several surface points. The typical spectra are shown in Fig. 1 and Fig. 2. The fluorescence background is very strong for most of the fruits, and the spectra demonstrate no obvious characteristic peaks among various fruits. Most of these fruits show the Raman spectra of carotene at 514.5 nm excitation. The strongest Raman modes of carotene are located at 1521, 1155 and 1003 cm$^{-1}$. Several previous papers have reported the Raman spectra of leaves and other plants, which also mainly demonstrate the carotene vibration modes $^{[5]}$. The reason is due to the strong resonance Raman effect between the incident wavelength and the energy level of carotene. The resonance effect not only enhances the scattering intensity of carotene and produces strong fluorescence, but also consumes the incident energy so the scatterings from other constituents are very weak. In the measurements, the fluorescence background in the spectra of pear, apple, and grape are stronger than that of orange.

Raman spectra of fruits obtained by FT-Raman with 1064 nm excitation show little fluorescence,
and there is also no resonant Raman effect taken place, they are different from excitation at 514.5 nm. There are characteristic Raman modes for each fruits in the Raman spectra excited at 1064 nm. One can distinguish various different types of vegetables and fruits clearly at 1064 nm excitation. Fig. 2 demonstrates the measured results at this excitation.

3.2. FT-Raman spectra of four kinds of pesticides at 1064 nm excitation

We obtained Raman spectra of pesticides easily using the FT-Raman spectrometer with 1064 nm excitation. Although the chemical compositions of the pesticides are complex, the Raman spectra exhibit characteristic features for each of the different pesticides. The results are shown in Fig. 3. It can be seen from the figure that the most pronounced modes of the four pesticides are very characteristic and different and can be easily identified (refer to details in Table 1 below).

An interesting observation from Fig. 3 is that there are several Raman modes recorded at the same wavenumber for Abameclin Petroleumoil (a) and Chongmanjue (b). According to this observation, one can conclude that these two pesticides may well have contained a similar component.

Table 1

| Pesticides                      | Vibration modes (cm⁻¹)                          |
|---------------------------------|------------------------------------------------|
| Abameclin Petroleumoil (a)      | 3056, 2922, 2872, 2732, 1607, 1449 and 1003    |
| Chongmanjue (b)                 | 3056, 2922, 1605, 1003                         |
| Cypermethrin(c)                 | 3071, 1591, 1348 and 993                        |
| Zhibaoboshi(d)                  | 1000 ~1600, 1363, 1517 and 1575                 |

Fig. 3. FT-Raman spectra of four kinds of pesticides (A: 500 to 1800 cm⁻¹, B: 2600 to 3200 cm⁻¹) with a: Abameclin Petroleumoil, b: Chongmanjue solution, c: Zhibaoboshi solution and d: Cypermethrin solution.
3.3. FT-Raman spectra of several fruits with pesticides

Measurements of Raman spectra of several fruits with pesticides are performed with the same experimental condition as before. Some sample results are reported in this section. For example, the FT-Raman spectra of the pure peels differ from that with sprayed Zhibaoboshi pesticides in the region between $800 \sim 1800 \text{ cm}^{-1}$, which is depicted in Fig. 4. The FT-Raman spectra of the pesticide Zhibaoboshi left on the four kinds of peels are shown in Fig. 4. Obviously, the characteristic modes of Zhibaoboshi are presented in each spectrum, which are 993, 1348 and 1591 cm$^{-1}$. In similar experiments, FT-Raman spectra obtained for four kinds of pesticides on the same fruit are shown in Fig. 5. We have chosen to report on Chinese goosebeery as an example. In Fig. 5, we can observe all the characteristic modes of the different pesticides as well as those of the Chinese goosebeery. From the spectra the locations of characteristic modes of pesticides recorded from 500 to 2000 cm$^{-1}$ have been identified: Chongmanjue at 624, 1003 and 1065 cm$^{-1}$, Zhibaoboshi at 993, 1348 and 1591 cm$^{-1}$, Abameclin Petroleumoil at 786, 1449, 1607 cm$^{-1}$, and Cypermethrin at 1363 cm$^{-1}$. For comparison, the Raman spectrum of pure Chinese goosebeery without any pesticide is also presented in the Fig. 5. From these results, definitely, one should be convinced that the method presented can be used for automatic and intelligent identification of trace amount of pesticides left on the surface of fruits.

4. Discussions

The compositions of fruits are very complicated as there are usually different amount of water, sugar, carotene protein, fat, vitamin, as well as other components and elements such as calcium, iron, phosphorus etc. At 514.5 nm excitation, the fluorescence background is very strong and the Raman spectra of fruits are mainly from carotene due to the strong resonance Raman effect. Carotene absorbs green light strongly at 514.5nm excitation. Resonant enhanced scattering intensity of carotene dominates the vibration spectrum. The incident energy at 514.5 nm, is consumed by the resonance absorption and fluorescence, hence the interaction with other components are very weak. Therefore, the Raman spectra from other components do not show up in the recorded spectra. It is impossible to characterize and analyze the components on the vegetables and fruits by Raman technique at such excitation wavelength. However, the condition will be totally different if the sample is excited at a different wavelength. In such a case it is effective and convenient to detect pesticides left on the
surface of fruits. Any small amount of pesticides can be identified easily by Raman spectrometer according to their characteristic Raman peaks if they are not covered by the strong resonance peaks from carotene. While at 1064 nm excitation, not only the fluorescent interference is strongly suppressed, but also more Raman modes besides carotene were obtained. So one can use FT-Raman technique to measure the effective components in the fruits and vegetables as well as the pesticides left on the surface.

Near infrared Fourier transform Raman spectrometer (FT-Raman) is a useful apparatus for detecting pesticide residuals. One can obtain the Raman spectra of fruits and pesticides simultaneously. It is thus clear that the pesticide on the surface of fruit can be detected by Raman spectrometer conveniently. The results presented here clarified the issues and doubts of such application. Raman technique has been employed here to analyze pesticides residue as a non-destructive, fast, convenient and precise method.

5. Conclusions
The main conclusions that can be drawn from this paper are:
(1) Raman spectra can be used to measure the vegetables and fruits in situ and without special preparation. Present Raman instrumentation technology enables the recording of reliable and repeatable spectra in this application.
(2) With 514.5 nm excitation, the main spectrum from vegetables and fruits is that from carotene, and the fluorescence background is fairly strong.
(3) With 1064 nm excitation, the resonance Raman effect of carotene is much reduced, and at the same time strong fluorescence is also suppressed. Hence it is very convenient to obtain the characteristic Raman spectrum of each vegetables and fruits, as well as to identify individual components. At this excitation wavelength, one can also detect the pesticides left on the surfaces easily.

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