Raman Analysis of Fatty Acid Unsaturation

(As appeared on Spectroscopy – The Application Notebook, February, 2005, page 20.)

Rapid analysis by Raman spectroscopy of fatty acid (FA) unsaturation can be employed for quality & process monitoring and biomedical applications.

The unsaturation of FAs and hydrocarbons is pivotal in nutritional, medical, and chemical applications, which makes the analysis of double bond (DB) content critical for quality assurance and monitoring of synthetic and production processes. Evidence that normal tissue and invasive carcinoma cells often differ in membrane fluidity underscores the diagnostic and medical value of FA analysis. When using multiple spectral markers to analyze either simple or complex systems, Raman spectroscopy can provide the speed and specificity needed.

Experimental Methods

C-18 FA standards (stearic acid, oleic, linoleic, and linolenic acids) in vials, two brands of safflower oils (SOs) in glass and plastic bottles, and fish oil FA in gelatin capsules were analyzed using LSI Dimension-P1™ HR Raman System with Vector Raman Probe™ and External Sampling Module (ESM) at 80°C and room temperature. DB values of 1.5 and 2.5 were obtained using 1:1 mixtures of oleic:linoleic and linoleic:linolenic, respectively. Linearity of DB content with different peak intensities was examined at 80°C so as to have stearic acid in a liquid state to obtain zero DB peak values. Raman peak intensities were examined at 1265 and 1655 cm^{-1} for CH ethylene and C=C alkyl stretching, respectively, and the ratio of 1265 and 1301 cm^{-1} (CH_2-alkanes) was also examined. Room temperature studies also included SO and fish oil samples, each analyzed *in situ*. RamanSoft™ was used for data acquisition, background removal, and normalization to laser power and to the peak intensity at 1438 cm^{-1} (CH3, CH2-alkanes).

Results

A linear relationship of peak intensity with DB content is illustrated for the peak at 1655 cm^{-1} (Figure 1). The peak at 1265 cm^{-1} and the ratio of peaks at 1265 and 1301 cm^{-1} also provided a strong relationship with DB content. However, since the alkane peak at 1301 cm^{-1} decreases slightly with increasing DB content, the relationship of this ratio with DB is non-linear. The three quantitative methods were tested with the two brands of SO and fish oil samples. The expected DB contents were 1.07 and 1.7 for the SOs and 1.75 for fish oil; the average DB content ($\pm$ 6%) for SOs was found to be 1.01 & 1.67 and for fish oil, 1.86.

Conclusions

In situ analysis with an external sampling system and multiple Raman spectral markers permits quantification of FA unsaturation. These quantitative models are the basis for algorithms for use in a range of applications by analytical chemists and food scientists. Moreover, the data indicate that differences in Raman spectra in the 1265 and 1301 cm^{-1} regions, associated with tissue lesions, e.g., invasive breast carcinoma[1], reflect, in part, the population of FAs, and that this may be a useful tool for neoplastic tissue classification.

Reference

1. C.J. Frank and R.L. McCreery, Anal. Chem. **67**, 777-783 (1995)

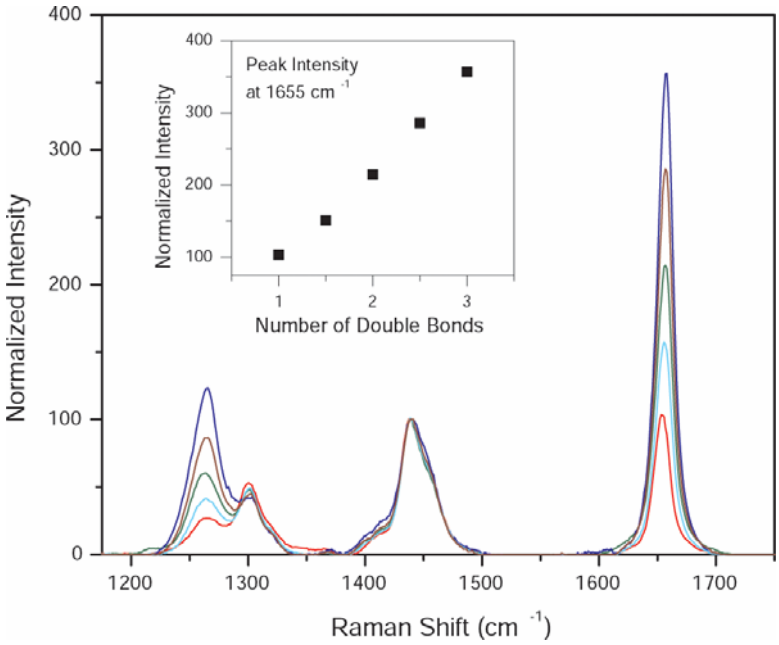

Figure 1. Raman spectra of oleic (red), linoleic (green), linolenic (blue) acids and 1**:**1 mixtures of oleic**:**linoleic (cyan) and linoleic**:**linolenic (maroon). Inset: Quantification of double bond content at 1655 cm^{-1}.