Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2014, 6(3):1356-1360

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Spectral discrimination of gastric cancer and atrophic gastritis by the comparison of three parameters based on Raman spectroscopy

Xiaozhou Li[a*], Su Zhang[b], Tianyue Yang[a] and Siqi Li[c]

[a]*School of Science, Shenyang Ligong University, Shenyang, China*
[b]*Section of Physiology and Biochemistry, Shenyang Sport University, Shenyang, China*
[c]*Integrated Life Sciences, Kent State University, Kent, America*

ABSTRACT

The feasibility of discriminating atrophic gastritis and gastric cancer from healthy controls and gastric cancer after operation by serum based on Raman spectroscopy was explored. Fifteen atrophic gastritis, 16 gastric cancer, 12 gastric cancer after operation and 14 controls were selected in our experiment. Comparison between different Raman spectra demonstrated that there are changes occurred with the deterioration of diseases. Three parameters were calculated and compared among groups. This exploratory study demonstrated potential for developing serum Raman spectroscopy analysis into clinical use for gastric diseases.

Keywords: gastric cancer, atrophic gastritis, serum, Raman spectroscopy

---

INTRODUCTION

American cancer society estimates that about 21 thousand new cases of gastric cancer will be diagnosed, and 11 thousand related deaths will occur in 2013 in USA[1]. Curing cancer depends upon early diagnosis and prompt treatment[2]. Biomedical imaging such as magnetic resonance imaging (MRI), positron emission tomography (PET), and computed tomography (CT) for gastric cancer and gastritis are the main methods for gastric disease diagnosis[3]. However, these conventional methods have drawbacks such as tedious and easily influenced by human factors[4].

Raman spectroscopy is caused by inelastic light scattering and can provide 'fingerprint' spectra for particular molecular structure[5]. It can provide quantitative information about the biochemical and morphological states of tissues in a noninvasive manner[6]. Over the last decade, the use of Raman spectroscopy has grown significantly. Raman spectroscopy has been applied to a number of diseases detection[7]. Applications include lung, breast and cervical cancers, etc[8–10].

In this study, Raman spectroscopy of serum taken from gastric cancer were measured and compared. Trends and characteristic parameters of the Raman spectra were analyzed among groups. Our research aims to find an effective way for differentiations among atrophic gastritis, gastric cancer, gastric cancer after operation and normal.

EXPERIMENTAL SECTION

Forty-five selected samples were studied, and were composed of seven 14 normal cases, 16 gastric cancers (before operation), 12 gastric cancers (after operation), and 15 atrophic gastritis. All specimens were obtained from Tumor Hospital of Liaoning Province and have been exactly diagnosed in clinic.

Subjects were phlebotomized before breakfast in morning in view of the interference of the intake of variant food. The vein blood obtained was separated in segregator at a speed of 3000 rot/min for ten minutes. Then upper serum was

sucked and made into samples. All samples were mounted, stored in refrigerator (temperature 4°C) until study but not exceeding three weeks, and were partitioned as normal cases, gastric cancer (before operation), gastric cancer (after operation), and atrophic gastritis. For spectroscopic study, specimen about 2 mL was injected into a quartz cuvette with a one-off sucker.

The experimental setup is shown in

Figure **1**. An Ar-ion laser is used as the source of excitation light (488.0 nm and 514.5 nm). After modulation by a chopper (modulation frequency is 700 Hz), the laser beam is reflected into the sample (pass through cuvette lengthways from the mouth of cuvette to the bottom). In the vertical direction of the beam, fluorescence and Raman spectrum was focused by a lens into a double spectrometer (it can be precisely controlled by a computer) equipped with a PMT. And after amplified by a lock-in amplifier, spectral data were input into a computer and transacted.

The fluorescence and Raman spectra cover the range of 520 - 640 nm with a resolution of $2cm^{-1}$. Three spectra were recorded for each sample. The first is excited by 514.5 nm, and the second is excited by 514.5 nm after samples are radiated by laser.

We calculated the values of three parameters as we did in our former studies. But the most important in this study is to find the best criteria for spectral diagnosis of gastric cancer and differentiate gastric cancer from other gastric diseases. In order to improve the diagnostic accuracy, many values were tested and the best one that fit for gastric cancer was discovered.

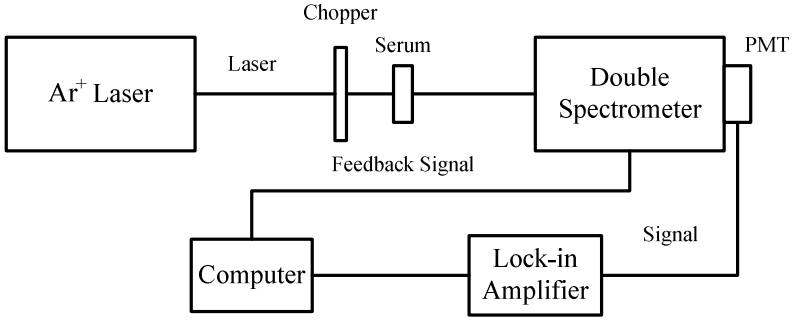

Figure 1. Experimental setup of spectroscopy system

RESULTS AND DISCUSSION

Among the seven normal cases, six exhibit three relatively strong sharp peaks (A, B and C). A typical one is shown in Figure 2(a). And the residual one has not, we thought, accounting for the bad quality of serum. Additionally, there is a broad, featureless fluorescence band centered at 547 nm. Raman peaks A, B and C at the range from 1100 cm^{-1} to 1579 cm^{-1} were generated by biochemical constituents of proline, tyrosine and phenylalanine related to pathological changes of gastric diseases[11].

Spectra of gastric cancer after operation and the one of normal people both have three apparent Raman peaks (see Figure 2 and Figure 3). It indicated that two kinds of samples had similar components and the abnormal metabolism of tumor cells was refrained after operation. Latter examinations in clinic proved that the operation was very successful.

For atrophic gastritis and gastric cancer after operation, three sharp Raman peaks were observed in the spectra excited by 514.5 nm (Figure 2). The intensity of them is lower in contrast to normal cases, but higher than gastric cancer before operation. But the spectrum of gastric cancer before operation only has a broad fluorescence band centered at 550 nm, without any sharp Raman peaks. However, a notable difference is that the red shift of fluorescence peak in atrophic gastritis and gastric cancer are often bigger than 12nm, whereas are less than 12 nm in normal cases and gastric cancer (after operation). Just as normal cases, spectra excited by 514.5 nm after radiated by laser (Figure 3) of atrophic gastritis, gastric cancer before and after operation all changed remarkably, both spectral shape and intensity distribution compared with spectra excited by 514.5 nm (Figure 2).

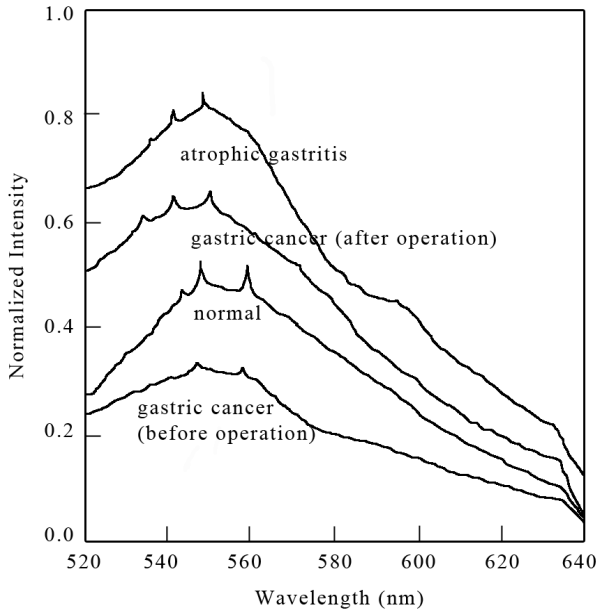

Figure 2. Raman spectra excited by 514.5 nm of the four groups

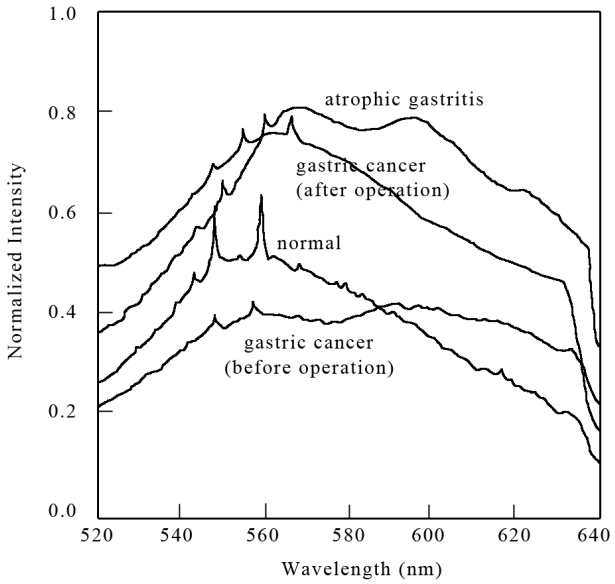

Figure 3 Raman spectra excited by 514.5 nm after radiated by laser of the four groups

For quantitative evaluation of the spectra, three parameters were designed. The three parameters are Δλ, α and β. Among them, Δλ is the red shift of fluorescence peak. α is the ratio between the fluorescence peak intensity at 520nm and the intensity at 634 nm – I_{520}/I_{634}. β is the ratio between the relative intensity of Raman peak C radiated by 514.5nm and the one excited by 488.0 nm – $I_{514.5}/I_{488.0}$.

Three parameters of the four groups (normal, atrophic gastritis, gastric cancer and gastric cancer after operation) were calculated and the results were shown in Table 1. From the table we can see that, all the three parameters have statistically significant difference between groups (p value<0.001).

Experimental data showed that β was an applicable parameter for demarcation between stomach cancer and atrophic gastritis. Comparison showed the best values were 12 nm, 0.8 and 1.0 respectively. But for the diagnosis of colon cancer, researchers indicated that the best values were 11.3 nm, 0.94 and 1.02 respectively. We utilized β < 1.0 as a criterion and got an accuracy of 81.5% in stomach cancer detection. In clinical medicine, atrophic gastritis is viewed as

a kind of pathological change before gastric cancer. And the similarities of them in spectrum after samples are radiated by laser, to some extent, indicate that there are many identical components in their serum.

Though it is observed that the serum native fluorescence spectra of stomach cancer cases are distinctly different from that of atrophic gastritis cases, it should be noted that the fluorescence spectra of other types of stomach diseases had not been investigated, and whether β is the real distinction between stomach cancer and other stomach diseases should be testified by more researches on the serum native fluorescence spectra of other stomach diseases.

Another distinction is the ratio (α) between relative intensity at 520 nm and at 634 nm in spectrum excited by 514.5 nm after radiated by laser. In normal cases and stomach cancer (after operation), α usually bigger than 0.8, and in others are less than 0.8. Using Δλ or α, we can only differentiate most stomach cancer from normal cases and stomach cancer (after operation). Other features should be found to distinguish stomach cancer from atrophic gastritis.

Figure 4 is the three parameters of all the samples from three groups. From the figure we can see that, parameters belonging to different groups have apparent different values. The results show that the three parameters are useful in the prediction of groups.

Table 1. Three parameters and the SD value of three groups

Parameters	Value (mean ± SD)				*p*-value
	Normal	Atrophic gastritis	Gastric cancer	Gastric cancer after operation	
Δλ	10.571(0.495)	12.867(0.806)	13.062(0.747)	10.250(0.433)	<0.001
α	0.957(0.049)	0.707(0.025)	0.706(0.024)	0.721(0.001)	<0.001
β	1.064(0.048)	1.080(0.040)	0.844(0.049)	1.058(0.049)	<0.001

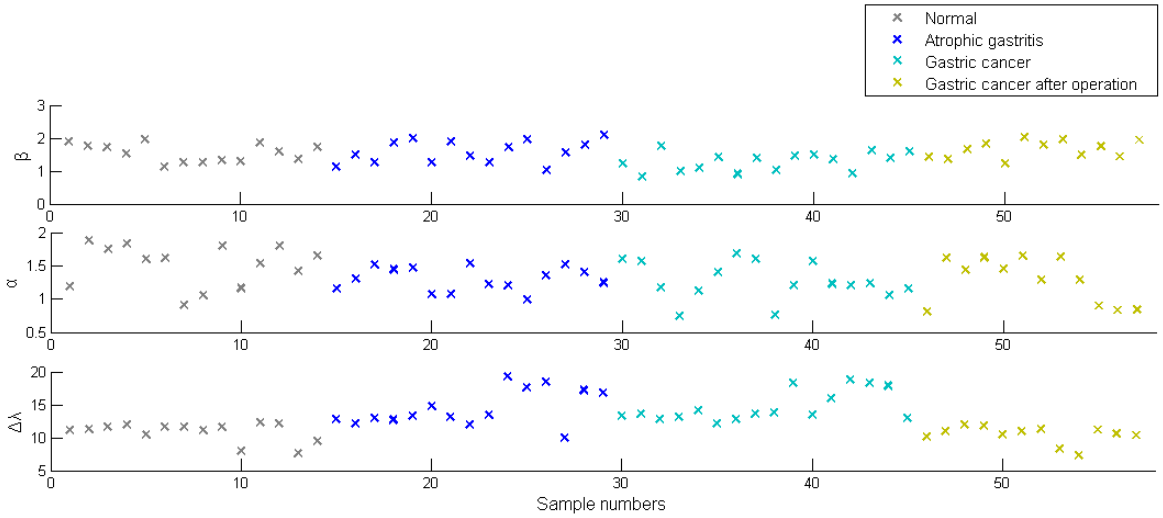

Figure 4. Values of the three parameters of the three groups

CONCLUSION

We investigated the spectra of normal, stomach cancer (both before and after operation), esophagus cancer and atrophic gastritis sera for stomach cancer detection. Results demonstrate several points. First, spectra of different samples exhibit different features, but we cannot differentiate them from each other at once; second, of the three parameters we have introduced, β is the best one that can differentiate stomach cancer from other cases, especially from atrophic gastritis. Therefore, we suggest that native fluorescence spectroscopy of blood plasma may be used as a potential method to discriminate stomach cancer subjects from normal and other stomach diseases group. However, spectra of other stomach diseases have not been measured, and extensive studies with more number of cases have to be carried out to examine the repetition of such difference features between different samples.

REFERENCES

[1] R Siegel; D Naishadham; A Jemal. *CA: A Cancer Journal for Clinicians*, **2013**, 63(1), 11–30.

[2] International Agency for Research on Cancer. World Cancer Report 2014, World Health Organization: Geneva, **2014**.
[3] Gastric cancer, Contemporary issues in cancer imaging, Cambridge University Press: Cambridge, UK ; New York, **2010**.
[4] JC Layke; PP Lopez. *Am Fam Physician*, **2004**, 69(5), 1133–1140.
[5] JR Baena; B Lendl. *Current opinion in chemical biology*, **2004**, 8(5), 534–539.
[6] LM Moreira; L Silveira Jr.; FV Santos; JP Lyon; R Rocha; RA Zângaro; AB Villaverde; MTT Pacheco. *Spectroscopy*, **2008**, 22(1), 1–19.
[7] DI Ellis; R Goodacre. *Analyst*, **2006**, 131(8), 875–85.
[8] H Yamazaki; S Kaminaka; E Kohda; M Mukai; H Hamaguchi. *Radiat Med*, **2003**, 21(1), 1–6.
[9] YX Ci; TY Gao; J Feng; ZQ Guo. *Applied spectroscopy*, **1999**, 53(3), 312–315.
[10] A Mahadevan-Jansen; MF Mitchell; N Ramanujamf; A Malpica; S Thomsen; U Utzinger; R Richards-Kortumt. *Photochemistry and photobiology*, **1998**, 68(1), 123–132.
[11] S Luo; C Chen; H Mao; S Jin. *J. Biomed. Opt*, **2013**, 18(6), 067004–067004.