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Detection and identification of Colon Cancer and Rectum Cancer Using Fluorescence and Raman Spectrum

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ABSTRACT-Laser-induced fluorescence spectroscopy (LIF) and Raman spectrum of serum for detection of colon cancer and rectum cancer were investigated in this paper. The aim of this study was that using Raman spectrum and LIF analysis the serum of colon cancer and rectum cancer for found the difference compared to normal, the difference was found. For example: the peaks intensity and red shift both different In this paper, we investigated 82 colon cancers, 69 rectum cancers and obtained 80.7%, 82.5% accuracy separately compared to clinical diagnostic. It is exploding that use Raman spectrum and LIF to detection of cancer .

Keyword: fluorescence, Raman spectrum, serum, rectum cancer ,colon cancer

1. INTRODUCTION

Raman spectrum and LIF got it's star as weapon against cancer and research are developing theory. LIF is more and more important for diagnosis disease recently. Human being is exploring an easy way to check cancer and obtained notable progress on diagnosis of colon cancer using Raman spectrum and LIF[1-8].,and Raman spectroscopy is a powerful new tool for Single molecule detection and identification of analysis[9-12]. LIF can reflect the chemical structure and the state information of its micro-environment, while Raman spectrum provided the high sensitivity. The malignant tumor tissue cell's state metabolize, the chemical structure and micro-environment were changed. For example the PH, oxidation-reduction, chemical bond, polar and ion concentration and so on have been changed, which makes it possible that we use Raman spectrum and LIF to analysis human serum for discovery the difference.

Through use LIF and Raman spectrum to analysis serum sample cases, we found the colon cancer and rectum cancer's metabolite was difference. It provided the criteria for diagnosis cancer. Further research for rectum cancer and colon cancer we use the parameter β α $\Delta\lambda$ to identification them. The parameter I_λ is relative intensity of C Raman peak (absolute intensity divided by maximum intensity). The parameter β is I_{5145}/I_{4880} . The parameter $\Delta\lambda$ is the red shift, while the parameter α is the ratio of start point and end point in fluorescence-Raman spectrum. We use it in clinical diagnostic and obtained 80.7% and 82.5% accuracy compared to pathological section diagnosis.

2. MATERIAL AND METHODS

It may become more difficult to extract valuable information because some chemical components would violently influent the Raman spectra, so we must process the samples beforehand. To minimize the interference, subjects were phlebotomized before breakfast in the morning. The vein blood obtained was separated in segregator at 3000 rot/min for 10 min. Then upper serum was sucked and made into samples. Samples were kept in refrigerator (temperature 4^oc) hermetically for latter investigation but not exceeding four day.

Spectra were collected with a double spectrometer equipped with a PMT. After amplified by a lock-in amplifier, for excitation. What we recorded was relative intensity (absolute intensity divided by maximum intensity) Spectral data were input into computer and transacted The spectral range scanned was from520nm to 640nm and from 510nm to 530nm, from 540nm to 560nm and from 520nm-640nm at a spectral resolution of $2cm^{-1}$ .the frequency of chopper was 700HZ.Fig.1 shows the main parts of our instrument: Ar-ion laser (made in USA), PMT(R928 model),. lock-in amplifier

(SRS-830 model), double spectrometer (HRD-1model). The wavelengths of 488.0nm and 514.5nm were chosen in order to reduce such interference as the undulation of laser power. And in the process of original data transaction, method of least squares was used to smooth spectra.

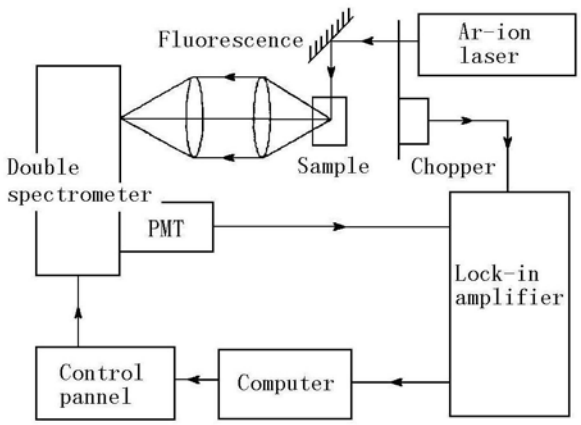
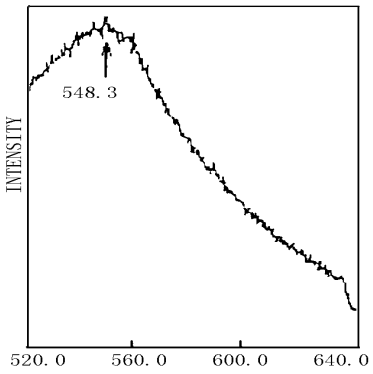


Fig 1 instrument

Described spectrometer collected the needed spectra and transformed them, so we could observe a spectral band from the fluorescence spectrum in the PMT system. In the band, region of fluorescence spectrum was wider than Raman spectrum. The noise mainly composed by system shot noise. After frequency calibration and spectral correlation in spectrometer system detection, we could get a relative intensive-wavelength graph.

3. RESULTS AND DISCUSSION

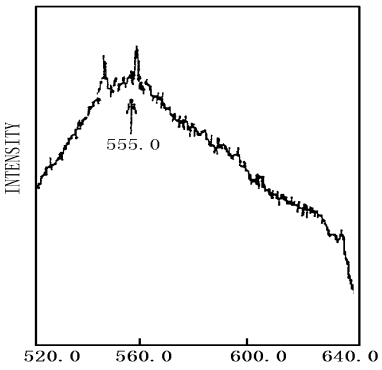
We performed a series of research and found much useful data for diagnosis colon cancer and rectum cancer. After sample (normal, colon cancer and rectum cancer) excited by 488.0 nm, 514.5 nm of laser, we found the native fluorescence spectroscopy of serum be produced. It proved that there were some fluorescence materials. If sample was induced it can radiated the fluorescence in visible region. Fig 2-1, Fig 2-2 and Fig2-3 show it. There is obvious native fluorescence peak in Fig 3-1 of normal. There is big native fluorescence peak in Fig 2-2 obviously of colon cancer. There is no obvious fluorescence peak in Fig 2-3 of rectum cancer. If sample (normal and cancers) was exited some time by 514.5 nm only, the normal and the cancer have a little of difference on producing fluorescence. It proved that the fluorescence material in serum didn't decompose obviously. Samples (normal and cancers) were excited some time by 488.0 nm, then excited by 514.5 nm. We found the fluorescence intensity decreased obviously. It proved that the fluorescence in serum could be decomposed by 488.0nm. It's showed on Fig 2-4, Fig2-5 Fig 2-6. There are red shift of colon cancer and rectum cancer obviously, but there is little red shift of 1.7nm to normal. The red shift ($\Delta\lambda$) is 11.1nm of colon cancer, but the red shift ($\Delta\lambda$) is 34nm of colon cancer. We select $\Delta\lambda>0.84$nm to identification colon cancer, use $\Delta\lambda>16.4$nm distinguish rectum cancer. We use more than 200 instances for research. And from the average quantity, we know that, generally speaking, the red shift of normal is below 12nm, but the red shift of cancer is beyond 12nm. So we can use $\Delta\lambda$ to distinguish cancer. At the same time, we found that the change of native fluorescence is obvious in range 600nm-640nm of rectum cancer, but there is a little of change of native fluorescence to colon cancer. It's proved that the metabolite content of 600nm-640nm is abundant in rectum. The parameter $\alpha>1$ obviously of normal, but $\alpha<1$ obvious of rectum cancer. All it was showed in Fig 2-5 and Fig 2-6. After sample was excited by 488.0nm and 514.5nm the Raman spectrum all was product. It proved that the metabolite in serum can product Raman emission. Raman spectrum all was added to fluorescence, there is strong fluorescence background, the normal and the cancer's Raman frequency shift are same. The average is 1009cm^{-1}, 1164cm^{-1} and 1523cm^{-1}. It proved the same material product the Raman spectrum. Following the time passed the intensity of Raman peak didn't decreased. It proved that the wavelength 488.0nm and 514.5nm can't lead to material decompose obviously which can product Raman spectrum. The Raman spectrum relative intensity (I_λ) is different after sample (normal and cancers) excited by 488.0nm. The parameter I_λ is 62.9‰, 183‰ and 74‰ of normal, colon cancer and rectum cancer separately. When sample was excited by 514.5nnm, there is only change on relative intensity of Raman peak. The parameter I_λ is 80.5‰, 112‰ and 55‰of normal, colon cancer and rectum cancer separately. So we can obtain the parameter β. It is obvious that βnormal>1, βcolon<1, βrectum<1. We often use $\beta<1$ to identification cancer. The parameter β is a major way cancer and the parameter α and $\Delta\lambda$ are auxiliary way.



Wavelength (nm)(normal)

Excited by 514.5nm

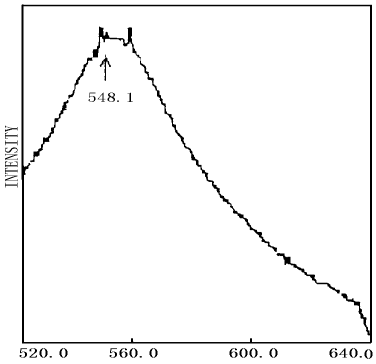
Fig2-1



Wavelength (nm)(normal)

Excited by 514.5nm after sample being radiated by laser

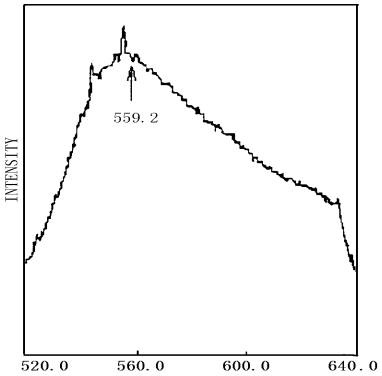
Fig2-4



Wavelength (nm)(colon)

Excited by 514.5nm

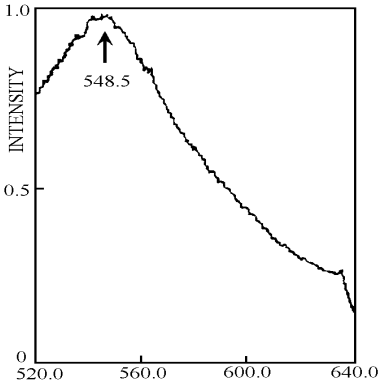
Fig 2-2



Wavelength (nm)(colon)

Excited by 514.5nm after sample being radiated by laser

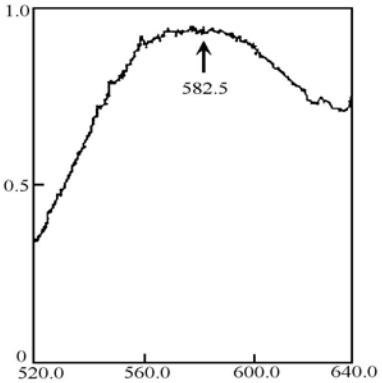
Fig 2-5



Wavelength (nm)(rectum)

Excited by 514.5nm

Fig 2-3



Wavelength (nm)(rectum)

Excited by 514.5nm after sample being radiated by laser

Fig 2-6

Tumor cell's metabolize is exuberant, the malignant tumor is obvious especially. It makes the content of DNA increase, the process of ribotide compose increase but the process of ribotide decompose decreased. The compose process and the decompose process both increased of protein, even the normal tissue cell's decompose material of protein were snatched to compose tumor protein, which make the content of it in serum increased. All the tumor tissue cell's metabolize material will enter human serum. Therefore detect the metabolic change in serum can provide the information of cancer occurred. Results of some studies indicate that porphyrin concentration occurs in cancerous serum[11][12]. Fluorescence ranging between 600nm and 640nm may be derived from the transition of π-electron in porphyrin in heme protein[13]. And riboflavin contributes to the fluorescence ranging between 510nm and 530nm. We assumed all these changed is due to the photochemical reaction of some materials in serum. Research performed in epidemiology showed beta carotene has close relationship with the incidence of cancer. Spectral diagnosis using β parameters that is correlated with beta carotene agreed with clinical diagnosis.

4. CONCLUSION

The sequence of experiments we carried out show the following. There are some differences assuredly of colon cancer, rectum cancer and normal in serum. It's possible through Raman spectrum and LIF to detect the difference. The results indicate that the parameter β, α, Δλ to detect the difference of the cancer and normal.

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