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Application of Curve Fitting and Radial Basis Function in the Quantitative Analysis of Molecular Fluorescence Spectroscopy

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The fluorescence of Lactobacillus plantarum solution was characterized by two-dimensional fluorescence spectroscopy and three-dimensional fluorescence spectroscopy and the quantitative detection of the fluorescence peak intensity of Lactobacillus plantarum solution is performed. The application effect of radial basis function and curve fitting in the quantitative analysis of molecular fluorescence spectroscopy is studied. At the same time, the simulation technology is used for the quantitative detection of dihydroxybenzene isomers. It is found that the detection results are relatively accurate and the final measurement results are satisfactory.

1. Introduction

Optical analysis is an application method in signal change analysis, which can be used to study the wavelength and intensity during the transition process of molecular energy level. Fluorescence analysis is an effective method in optical analysis, which can absorb and activate visible light and detect the fluorescence intensity. However, in the actual application process, ordinary two-dimensional broad-spectrum analysis data can no longer satisfy the requirements of fluorescence measurement. For example, in the detection of environmental pollution, biological and food related data, accurate data cannot be obtained by fluorescence analysis, so the optimization of fluorescence analysis method is necessary. In the detection and analysis process of chemical signals, the problem of signal overlap or interference can be solved by radial basis function and curve fitting, so that the results of fluorescence analysis method is more accurate.
Based on this, this paper mainly introduces the curve fitting method and radial basis function to understand the problems in chemometrics research. Then, the application effect of radial basis function and curve fitting in fluorescence analysis is analyzed through the molecular fluorescence broad-spectrum analytical application of Lactobacillus plantarum solution and urine.

2. Literature review

Optical analysis is based on the analysis of signal changes in wavelength, intensity, or intensity of emission, absorption, or scattering when a substance interacts with radiant energy (Engelhardt et al., 2016). Fluorescence analysis is one of the optical analysis methods (Bauer et al., 2016). The substance is used to absorb the ultraviolet light into an excited state, and the process of deactivation (the process of returning to the original ground state) is performed to detect the fluorescence intensity by the phenomenon of light radiation (Lagerweij et al., 2017). Qualitative and quantitative analysis of substances can be performed by measuring the fluorescence intensity. The method has the characteristics of high sensitivity, selectivity, diversity, and rich information. It has developed into an important and effective means of spectral analysis, and has been widely used in various fields of analysis such as environment, biochemistry, video, and medicine (Du et al., 2017). Urine sampling is simple and harmless to the human body. Fluorescent marker studies are ideal for the diagnosis of various lesions, especially in early studies. Therefore, it has gradually become a new research hotspot. There are few studies on the use of spectroscopy in urine analysis abroad (Zhao et al., 2017). Urine fluorescein is used to study the residual toxic substances in humans and does not

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involve the spectral study of urine (Cranfill et al., 2016). After research, some scholars have found that due to the differences in individuals, the spectral characteristics of the fluorescence emitted by mucus will vary greatly under different physical and chemical environments (Massabò et al., 2016). Moreover, mucus is a multi-component system containing multiple fluorescent chromophores. It is difficult to find the characteristics of lesions in human urine with a simple spectral line. Thus, the accuracy of analysis using urine fluorescence spectroscopy may be affected (Ji et al., 2018).

Urine is a multi-component system containing albumin, porphyrin, riboflavin, bilirubin, glucose, urobilinogen, etc. (Birriel et al., 2018). It has been reported that riboflavin can be subjected to fluorescent photons with a stimulating emission wavelength of 520 nm, and the characteristic fluorescent peaks of coproporphyrin are at 612 nm and 671 nm, respectively. Thus, the contribution of riboflavin and coproporphyrin can be determined by the line in the experiment. It can be seen from the molecular structure of riboflavin that riboflavin contains four electron-donating groups -OH, which greatly enhances the degree of conjugation of π electrons. It emits strong fluorescence (Matsui et al., 2017).

In summary, with the development of modern analytical techniques, the acquisition of huge amounts of analytical data has become easier, and the complexity of data has become stronger. The acquired chemical data contains not only rich chemical information, but also mixed interference information such as noise and background. Traditional information data processing methods have been unable to meet the needs of analysis. Therefore, in order to solve the above problem, "chemical separation" is used instead of "chemical or physical separation" for quantitative analysis of overlapping spectra of complex systems. This approach makes multi-component analysis simple and fast. The curve fitting and radial basis function neural network method are applied to molecular fluorescence analysis, which provides new ideas and new methods for the application of spectroscopy.

3. Method

3.1 Curve Fitting Method

Fluorescence analysis has become an important tool for trace analysis due to its high sensitivity. When the peak positions of different spectra of complex samples are close, the bands may be severely overlapped, which will cause trouble for spectral analysis processing. In recent years, with the development of computer science, people have attached more importance to chemometrics, providing an important research method for multi-component simultaneous detection. Chemometric methods, such as principal component regression, wavelet transform, factor analysis, least square method and neural network model are widely used in the quantitatively analysis of complex systems. The EMG model, namely the modified Gaussian model, is a modification of the Gaussian model that can effectively describes the spectral peak of the tailing phenomenon. Implementation of the code: the genetic algorithm is not a direct discussion of the research object, but to unmarshall the problem into a suitable code. The real-number coding is used in this paper and each overlapping spectrum is regarded as a chromosome. Each chromosome has four 4-dimensional vectors, number of genes (peak number). The 4-dimensional vector corresponds to the four parameters of the EMG model. (The peak area A of the sub-peak, the strongest emission wavelength of the Gaussian peak, the standard deviation of the Gaussian function and the attenuation constant of the Gaussian function). In general, if the initial population with a large number is selected, it can produce more candidate solutions and the algorithm is less likely to fall into local situations. However, it also requires more iteration time, thus increasing the computation amount. If the initial population size is small, the range of the search space will also be small, so it is likely to stop iteration before reaching maturity. Also, it is possible to fall into local optimum, causing immature convergence. Based on experience, the initial group is set to 50 in this paper. The fitness function is also called the object function, which is the measurement function of the problem solving quality. Generally, the model function of the problem can be used as the object function. In this paper, the spectral curve of the molecular fluorescence spectroscopy is expressed by the EMG function. Table 1 is the comparison of relevant parameters of the simulated and fitted overlapping spectrogram, including the peak area of sub-peak, fluctuating failure constant, wavelength corresponding to the strongest emission, and standard deviation. The advantage of the matched grating method is simple structure. Also, it has no absolute requirement for the light intensity of reflection of the final detection, so the noise of all kinds of intensity will not affect the output result. However, this method also has its shortcomings: first, it requires two gratings to be strictly matched; second, limited by the dependent variable of reference grating, the detection range of sensing grating cannot be too large; third, due to the limited response speed of PZT, this method can only be used to detect the physical quantity of static low-frequency variation and it has limited ability for the detection of the physical quantity with high frequency, such as such as acoustic vibration.

Table 1: Comparison of correlation parameters of overlapping spectra of simulated generation and fitting

	Generation/Solution			
	A	r	tg	σg
Urine	38 727.0/38 832.0	31.0/31.0	412.0/412.0	33.0/33.0
CFLX	33 084.0/33 038.0	31.0/30.0	478.0/478.0	26.0/26.0

3.2 Radial Basis Function

The most straightforward method for detecting the wavelength variation of a fiber grating sensor is to use a spectrometer (or monochrome) to detect the ΔλB of the output light, as shown in Figure 1. The advantage of this method is that it has simple structure and is suitable for laboratory use. The disadvantage is that the resolution of conventional spectrometers based on dispersive prisms or diffraction gratings is low and cannot satisfy the requirements. Although the high-resolution fiber optic spectrum analyzers can meet the requirements, such spectrometers are expensive and large in size, so the resulting system lacks necessary compactness and robustness. In a sensor system for practical application, it is extremely unrealistic to detect the wavelength shift of the fiber grating using this type of spectrometer. More importantly, it cannot directly output the electrical signal corresponding to the wavelength variation, which is impossible to achieve the recording, storage and display of the result, the provision of necessary electrical signals to the control loop and the automatic control of industrial production process.

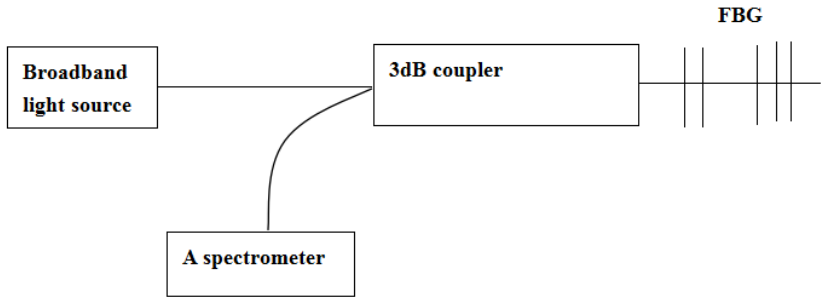

Figure 1: Schematic diagram of wavelength detection of fiber Bragg grating by spectrometer

The demodulation technology of FBG wavelength is one of the keys to the practical application of FBG sensors. The scheme of demodulating the FBG wavelength by using the tunable fiber F-P cavity has the advantages of small volume and low price, and can directly output the electrical signals corresponding to the wavelength variation, which is a good demodulation scheme. The F-P cavity can be used as a narrowband filter. In a certain wavelength range, if parallel light is incident on the F-P cavity, only the light of certain wavelengths satisfying the coherent condition can generate interference, resulting in a large coherence. This characteristic of the FP cavity can be used to detect the reflection wavelength of the FBG sensor. The principle of the detection is shown in Figure 2: The light from the broadband source is transmitted to the FBG sensor through the isolator and the light reflected back by the FBG sensor is introduced into the tunable F-P cavity through a 3dB coupler.

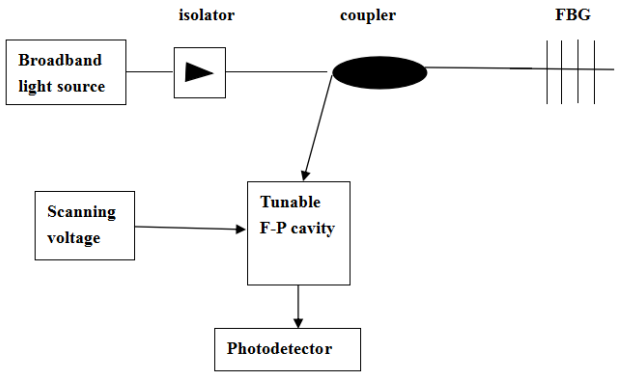

Figure 2: Schematic diagram of demodulation of tunable fiber F-P filter

4. Result Analysis

The experiment shows that the fluorescence intensity enhancement of GFLX in HAc-NaAc buffer system at pH 5.5 in the presence of 0.15 mol/L SDS is the optimal. The sensitization performance of SDS to GFLx may be caused by the formation of SDS micelles, which generates a masking effect fir the GFLX molecules dispersed and bonded to the micelles. On the one hand, it reduces the energy loss of collisions; on the other hand, it can protect GFLX from the fluorescence quenching effect of dissolved oxygen. At the same time, the GFLX molecule is in a more ordered microenvironment, which reduces the rate of non-radiative deactivation process and fluorescence quenching process of GFLX molecules, thereby improving fluorescence quantum efficiency and fluorescence lifetime and eventually leading to fluorescence enhancement. Endogenous substances in the urine can cause different degrees of fluorescence quenching of GFLX.

Table 2: broad-spectrum results of simulated synchronous fluorescence

Analyte	A		tg		g			
	Simulation	Resolution	Simulation	Resolution	Simulation	Resolution	Simulation	Resolution
Catechol	2139	2120	2.0	2.0	273.5	273.7	6.6	6.6
Resorcinol	4592	4580	2.0	2.0	274	274	7.0	7.0
Hydroquinone	5046	5080	4.1	4.1	286.5	268.4	7.1	7.1

Under the optimal experimental conditions, λcm=320 nm is in the wavelength range of 200 to 600 nm. The results of the fluorescence scanning of the blank fresh urine containing 0.04 to 5.0μg·mL-1GFLX show that there is a good linear relationship between the concertation and fluorescence intensity of GFLX in the range of 0.06 to 3.5μg·mL-1. The linear regression equation is F=56.911+733.68c, r=0.999 4 and the regression equation is the result of the blank solution after 11 consecutive detections. (The analytical result of the fitted synchronous fluorescence spectroscopy of adjacent, inter and diphenol are shown in Table 3 below)

Table 3: analytical results of synchronous fluorescence spectra of adjacent, inter, and diphenol

Compound	A		tg	g
Catechol	1911	2.0	275.3	6.6
Resorcinol	4410	2.0	272.3	7.0
Hydroquinone	5800	4.1	291	7.1

30 unknown strains are predicted by radial basis function neural network method. Table 4 is the predicted results of 30 strains. It can be seen from the data in the Table that the predicted results are in good agreement with the actual values, indicating that the results of the radial basis function neural network are very accurate in the identification of strains.

Table 4: The predicted and actual values of the ten sets of unknown data

	Lactobacillus acidophilus		Lactobacillus bulgaricus		S.mutans	
	predicted value	actual value	predicted value	actual value	predicted value	actual value
1	0.0013	0	0.4993	0.5	1.0025	1
2	0.0016	0	0.5001	0.5	1.0025	1
3	0.0014	0	0.5004	0.5	1.0023	1
4	0.0028	0	0.5000	0.5	1.0024	1
5	0.0057	0	0.4994	0.5	1.0032	1
6	0.0038	0	0.4987	0.5	1.0009	1
7	0.0032	0	0.4996	0.5	1.0040	1
8	0.0023	0	0.4989	0.5	0.9997	1
9	0.0046	0	0.4993	0.5	1.0017	1
10	0.0028	0	0.5002	0.5	1.0018	1

5. Conclusion

Molecular fluorescence spectroscopy is an effective means of spectroscopic analysis. This paper applies the radial basis function and curve fitting in the analysis of molecular fluorescence spectroscopy, and the three types of dihydroxybenzene isomers in urine and strains are analyzed, which effectively solves the GFLX interference and overlapping problems existing in the spectroscopic analysis process. In this paper, the RBF neural network method is used for the quantitative detection and analysis of NOR in urine, and a new detection method is proposed, whose results are satisfactory. During the simulation analysis, the error between the optimized simulation data and the experimental data is relatively small.

In the process of continuous updating and development of science and technology, it is easier to obtain the three-dimensional fluorescence spectrum data and related research literature is gradually increasing. In future quantitative detection and analysis of molecular fluorescence spectroscopy, it is necessary to combine three-dimensional data and neural network organically to form a multi-media advantage to promote the development of chemometrics.

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