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Research Article

Quantitative Determination of Germinability of *Puccinia striiformis* f. sp. *tritici* Urediospores Using Near Infrared Spectroscopy Technology

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Stripe rust caused by *Puccinia striiformis* f. sp. *tritici* (*Pst*) is an important disease on wheat. In this study, quantitative determination of germinability of *Pst* urediospores was investigated by using near infrared reflectance spectroscopy (NIRS) combined with quantitative partial least squares (QPLS) and support vector regression (SVR). The near infrared spectra of the urediospore samples were acquired using FT-NIR MPA spectrometer and the germination rate of each sample was measured using traditional spore germination method. The best QPLS model was obtained with vector correction as the preprocessing method of the original spectra and 4000–12000 cm^{-1} as the modeling spectral region while the modeling ratio of the training set to the testing set was 4 : 1. The best SVR model was built when vector normalization was used as the preprocessing method, the modeling ratio was 5 : 1 and the modeling spectral region was 8000–11000 cm^{-1}. The results showed that the effect of the best model built using QPLS or SVR was satisfactory. This indicated that quantitative determination of germinability of *Pst* urediospores using near infrared spectroscopy technology is feasible. A new method based on NIRS was provided for rapid, automatic, and nondestructive determination of germinability of *Pst* urediospores.

1. Introduction

Stripe rust caused by *Puccinia striiformis* f. sp. *tritici* (*Pst*) is an important air-borne disease of wheat worldwide [1–6]. This disease can occur in most wheat growing areas of China [2, 4–6]. Some devastating epidemics have occurred in China since the 1950s. Especially, in 1950, 1964, 1990, and 2002, wheat yield losses caused by this disease were as high as 6.0, 3.2, 1.8, and 1.3 million tons, respectively [2, 4]. Being an obligate parasitic pathogen, the causing agent of this disease expands in wheat host and has impacts on wheat growth and wheat production. A large number of urediniospores are released after rupture of the uredinia on wheat leaves. Generally, the long distance dispersal of wheat stripe rust relies on the transport of *Pst* urediniospores by airflow [2]. Up to date, isolation, purification, cultivation, and preservation of wheat stripe rust pathogen could not be performed on artificial medium. The spores of *Pst* used for scientific research are usually collected from diseased wheat leaves. Sometimes, the pathogen is acquired by using a spore trap. Due to the effects of the various environmental factors, the survival time of the *Pst* urediniospores is short under natural conditions. Thus the spores of *Pst* are usually preserved at low temperature. Only after germination of the *Pst* urediniospores landing on wheat leaves, it is possible for the pathogen to further penetrate the leaves and induce symptom appearance. The viable *Pst* urediniospores are very important for the effective spreads of wheat stripe rust. It is very helpful to precisely measure the germination rate of *Pst* urediospores for accurate prediction of the disease. Therefore, it is of great significance to investigate the methods for determining the germinability of *Pst* urediospores for the studies on the characteristics of pathogenic biology, pathogen monitoring, and disease prediction.

At the present time, the determination of the germinability of *Pst* urediospores is generally conducted by using the spore germination method [7, 8]. As a destructive measurement method, this method is time-consuming and laborious, and its measurement accuracy is easily influenced by environmental factors and human factors. Qiao et al. [9] evaluated the viability of *Pst* urediospores by detecting the integrity of RNA from the urediospore samples. The results showed that the samples with high RNA integrity had high viability. However, this method could not quantitatively evaluate the viability. Therefore, it is necessary to explore a nondestructive, simple, rapid, and accurate method for the determination of the germinability of *Pst* urediospores.

As a nondestructive, nonpolluting, and rapid analysis technology, near infrared reflectance spectroscopy (NIRS) has been widely applied in agriculture, chemical industries, pharmaceutical industries, and many other fields [10–12]. Studies on detecting plant diseases and nondestructive identification of plant pathogens based on NIRS have been reported [13–19]. Based on NIRS, Li et al. [18] qualitatively identified *Pst* and wheat leaf rust pathogen (*Puccinia recondita* f. sp. *tritici*, *Prt*) using distinguished partial least squares (DPLS) and made the quantitative determination of *Pst* and *Prt* using quantitative partial least squares (QPLS). Based on NIRS, Cheng et al. [19] identified three physiological races of *Pst* (CYR32, CYR31, and CYR33) using support vector machine (SVM) with high accuracy. At this present time, there are no reports on the quantitative determination of germinability of *Pst* urediospores using near infrared spectroscopy technology.

In this study, based on the germination rates of *Pst* urediospores obtained by using the traditional spore germination method and the obtained near infrared spectra of the corresponding samples, quantitative determination of germinability of *Pst* urediospores was implemented by using near infrared spectroscopy technology combined with QPLS and support vector regression (SVR). The effects of spectral data preprocessing methods, spectral regions, and the ratios between training sets and testing sets on modeling were investigated. The aim of this study was to provide a method for rapid and nondestructive determination of the germinability of *Pst* urediospores and to provide a reference for the nondestructive measurement of the spore germinability of other pathogens.

2. Materials and Methods

2.1. Materials. Three currently predominant physiological races of *Pst* including CYR31, CYR32, and CYR33 in China were used in this study. The races were multiplied on seedlings of wheat cultivar Mingxian 169, susceptible to all known physiological races of *Pst*, in the artificial climate chamber in the Lab of Plant Disease Epidemiology, Department of Plant Pathology, China Agricultural University.

2.2. Methods

2.2.1. Multiplication of Pst. After being soaked in sterile water for 24 h, the seeds of Mingxian 169 were sowed in pots (10 cm in diameter) with about 20 seeds per pot and then were incubated in the artificial climate chamber at 11–13°C and 60–70% relative humidity with 12 h light (10000 lux) per day. When the first leaves of the wheat seedlings were fully unfolded, the urediospores of CYR31, CYR32, and CYR33 preserved in the liquid nitrogen container were taken out. After water bath at 40°C for 5 min and hydration in the dark at 4°C for 12 h, the spores were reactivated and were used to make the suspension with 0.2% Tween 80. The leaves of wheat seedlings were sprayed with the spore suspension, and then the seedlings were immediately placed into a moist chamber in dark conditions at 11–13°C for 24 h. Subsequently, the inoculated wheat seedlings were incubated in the artificial climate chamber. When the uredinia appeared on wheat leaves, the urediospores of the three physiological races were individually collected and stored in a dryer at 4°C. To obtain enough urediospores with different germination rates resulting from the different storage times, multiplication and collection of *Pst* urediospores were conducted in different batches.

2.2.2. Acquisition of Near Infrared Spectra. The germination rates of the *Pst* urediospores under different storage times are different. The germination rate decreases with the prolonging of the storage time. Before acquisition of the near infrared spectra, the collected urediospores of the three physiological races of *Pst* were randomly mixed and stored in the dryer at 4°C. Then the urediospores under different storage times were randomly mixed to obtain pathogen samples with the germination rates of 0%~100% in a uniform distribution as far as possible. Totally, 64 *Pst* urediospore samples (160 mg per sample) with different germination rates were obtained. The near infrared spectra of the urediospore samples were acquired by using FT-NIR MPA spectrometer (Bruker, Germany). The measurement parameters were set as follows: measured spectral range, 4000–12000 cm^{-1}; resolution, 8 cm^{-1}; number of scans, 32. Each urediospore sample was equally divided into four parts (40 mg per part). One part of 40 mg *Pst* urediospores was put into a sample cup (4 mm in diameter) and its spectrum was acquired using integrating sphere diffuse reflectance method. Then the spectra of the other three parts were acquired using the same method. The tightness of the spores in the sample cup should be kept in the same consistency to reduce the experimental error caused by different tightness. Thus, four spectra were obtained for each urediospore sample. The average spectrum of the four spectra was regarded as the spectrum of the corresponding sample. A total of 64 near infrared spectra were obtained as shown in Figure 1.

2.2.3. Microscopic Determination of the Germination Rates of Pst Urediospore Samples. The germination rates of the *Pst* urediospore samples were measured using the spore germination method. After acquisition of the near infrared spectra, the spores of each sample were mixed with 0.1% water agar and were incubated in the dark at 9°C for 24 h. Then the germinated spores were checked microscopically by examining 300 spores for each sample. In this study, a spore with a germ tube longer than half of the diameter of the spore was

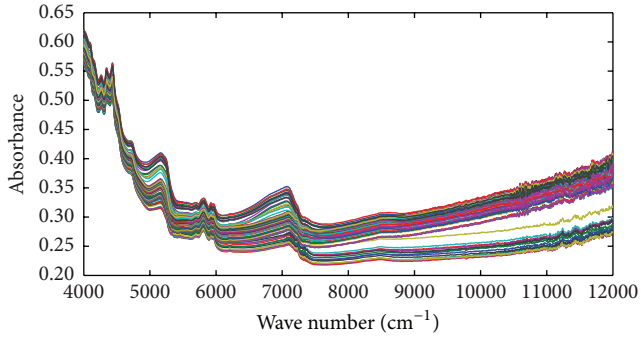

Figure 1: Near infrared spectra of 64 samples of *Puccinia striiformis* f. sp. *tritici* urediospores.

considered germinated. According to the results of microscopic examination, the germination rate of each *Pst* urediospore sample was calculated. Subsequently, data analysis was conducted based on the near infrared spectra of the *Pst* urediospore samples and the corresponding germination rates.

2.2.4. Establishment of the QPLS Models to Determine the Germinability of Pst Urediospores. The specific near infrared analysis software system CAUNIR [20] developed by China Agricultural University was used for the establishment of the QPLS models to determine the germinability of *Pst* urediospores. The obtained near infrared spectra of the *Pst* urediospore samples were randomly divided into training sets and testing sets based on the required ratios. The QPLS models to determine the germinability of *Pst* urediospores were built using internal cross verification method. The effects of six preprocessing methods of the original spectral data including centralization, range normalization, vector correction, scatter correction, first derivative transform, and second derivative transform on modeling in the spectral region of 4000–12000 cm^{-1} were compared. To select the suitable spectral region for modeling, the spectral region of 4000–12000 cm^{-1} was divided into 36 spectral regions including 4000–5000 cm^{-1}, 4000–6000 cm^{-1}, 4000–7000 cm^{-1}, 4000–8000 cm^{-1}, 4000–9000 cm^{-1}, 4000–10000 cm^{-1}, 4000–11000 cm^{-1}, 4000–12000 cm^{-1}, 5000–6000 cm^{-1}, 5000–7000 cm^{-1}, 5000–8000 cm^{-1}, 5000–9000 cm^{-1}, 5000–10000 cm^{-1}, 5000–11000 cm^{-1}, 5000–12000 cm^{-1}, 6000–7000 cm^{-1}, 6000–8000 cm^{-1}, 6000–9000 cm^{-1}, 6000–10000 cm^{-1}, 6000–11000 cm^{-1}, 6000–12000 cm^{-1}, 7000–8000 cm^{-1}, 7000–9000 cm^{-1}, 7000–10000 cm^{-1}, 7000–11000 cm^{-1}, 7000–12000 cm^{-1}, 8000–9000 cm^{-1}, 8000–10000 cm^{-1}, 8000–11000 cm^{-1}, 8000–12000 cm^{-1}, 9000–10000 cm^{-1}, 9000–11000 cm^{-1}, 9000–12000 cm^{-1}, 10000–11000 cm^{-1}, 10000–12000, and 11000–12000 cm^{-1}. The effects of different ratios between training sets and testing sets (1 : 1, 2 : 1, 3 : 1, 4 : 1, and 5 : 1) on modeling were also analyzed. Determination coefficient (R^2), standard error of calibration (SEC), standard error of prediction (SEP), and average absolute relative deviation (AARD) were used to evaluate the established QPLS models.

2.2.5. Establishment of the SVR Models to Determine the Germinability of Pst Urediospores. The obtained near infrared spectra of the *Pst* urediospore samples were preprocessed by using nine methods, respectively. The nine preprocessing methods included db2 level 1 decomposition denoising, db2 level 2 decomposition denoising, db2 level 3 decomposition denoising, Euclidean normalization, multiplication scatter correction, standard normalized variate transform, vector normalization, Savitzky-Golay (S-G) first derivative transform, and S-G second derivative transform [10, 21]. In this study, the soft thresholding method with heursure threshold was selected for denoising using db2 wavelet. When Euclidean normalization was carried out, the data were normalized by the Euclidean norm (2-norm). When S-G first derivative transform or S-G second derivative transform was performed, the degree of the polynomial was set as 3 and the span was set as 7. The calculations above were implemented by using the software MATLAB 7.8.0 (R2009a).

Using the content-grads method [21], the spectra of the *Pst* urediospore samples were divided into training set and testing set based on the ratio of training set to testing set equal to 1 : 1, 2 : 1, 3 : 1, 4 : 1, or 5 : 1, respectively. In the 36 spectral regions above, the SVR models to determine the germinability of *Pst* urediospores were built with radial basis function (RBF) as the kernel function based on the original spectra and the spectral data obtained by using the nine preprocessing methods, respectively. Using the grid search algorithm, the optimal penalty parameter C and the optimal kernel function parameter γ for each SVR model were searched in the range 2^{-8}–2^{8} with the searching step equal to 0.8. Mean squared error (MSE) was calculated at each point within the grid. As the minimum MSE of the training set was achieved, the corresponding values of C and γ were regarded as the optimal parameters. The values of R^2 and MSE of the training set and the testing set were used to evaluate each SVR model. And the best model was chosen to determine the germinability of *Pst* urediospores.

3. Results

3.1. The Effects of Spectral Data Preprocessing Methods, Spectral Regions, and Modeling Ratios on the QPLS Models

3.1.1. The Effects of Different Preprocessing Methods on the QPLS Models. In the spectral region of 4000–12000 cm^{-1}, the effects of different preprocessing methods of the original spectral data on the QPLS models for determination of the germinability of *Pst* urediospores are shown in Table 1. For the QPLS model built based on the original spectra, R^2, SEC and AARD of the training set were 0.9895, 0.02%, and 5.32%, respectively, and R^2, SEP, and AARD of the testing set were 0.8705, 0.08%, and 20.23%, respectively. For the QPLS model built when centralization, first derivative transform or second derivative transform was used as the preprocessing method, the value of R^2 of the training set was relatively low. This indicated that any of these three methods was not the optimal preprocessing method. For the QPLS model built based on the spectra preprocessed by range normalization, R^2, SEC, and

Table 1: Influence of different preprocessing methods of the near infrared spectra on prediction results of QPLS models for quantitative determination of germinability of *Puccinia striiformis* f. sp. *tritici* urediospores.

Preprocessing method	The number of principal components	Training set			Testing set		
		R^2	SEC/%	AARD/%	R^2	SEP/%	AARD/%
No preprocessing	9	0.9895	0.02	5.32	0.8705	0.08	20.23
Centralization	4	0.7032	0.11	22.87	0.7498	0.11	16.76
Range normalization	9	0.9833	0.03	6.93	0.8686	0.08	20.06
Vector correction	8	0.9665	0.04	9.44	0.8961	0.07	17.31
Scatter correction	8	0.9592	0.04	10.71	0.8455	0.08	21.09
First derivative transform	4	0.8081	0.09	19.31	0.8616	0.08	19.41
Second derivative transform	5	0.7851	0.10	20.59	0.8758	0.08	14.34
Average	7	0.8850	0.06	13.40	0.8526	0.08	18.46

AARD of the training set were 0.9833, 0.03%, and 6.93%, respectively, and R^2, SEP, and AARD of the testing set were 0.8686, 0.08%, and 20.06%, respectively. For the QPLS model built when scatter correction was used as the preprocessing method, R^2, SEC, and AARD of the training set were 0.9592, 0.04%, and 10.71%, respectively, and R^2, SEP, and AARD of the testing set were 0.8455, 0.08%, and 21.09%, respectively. The results indicated that the QPLS model built when range normalization or scatter correction was used as the preprocessing method was not better than that built based on the original spectra. For the QPLS model obtained with vector correction as the preprocessing method, R^2, SEC, and AARD of the training set were 0.9665, 0.04%, and 9.44%, respectively, and R^2, SEP, and AARD of the testing set were 0.8961, 0.07%, and 17.31%, respectively. Compared with the QPLS model built based on the original spectra, for the model QPLS model built when vector correction was used as the preprocessing method, R^2 of the training set was slightly lower, and SEC and AARD of the training set were slightly higher; however, the values of R^2, SEP, and AARD of the testing set were all better. Therefore, vector correction was used as the optimal preprocessing method of the original spectra in this study.

3.1.2. The Effects of Different Spectral Regions on the QPLS Models. After the original spectra were preprocessed using vector correction method, the QPLS models for determination of the germinability of *Pst* urediospores were built in the 36 spectral regions above, respectively. The effects of the different spectral regions on the QPLS models are shown in Table 2. The QPLS model built in the spectral region 4000–12000 cm^{-1} and the QPLS model built in the spectral region 5000–12000 cm^{-1} were better than that built in other spectral regions. For the QPLS model built in the spectral region 4000–12000 cm^{-1}, R^2, SEC, and AARD of the training set were 0.9665, 0.04%, and 9.44%, respectively, and R^2, SEP, and AARD of the testing set were 0.8961, 0.07%, and 17.31%, respectively. For the QPLS model built in the spectral region 5000–12000 cm^{-1}, R^2, SEC, and AARD of the training set were 0.9727, 0.03%, and 8.92%, respectively, and R^2, SEP, and AARD of the testing set were 0.8493, 0.08%, and 21.81%, respectively. In comparison with the QPLS model built in the spectral region 5000–12000 cm^{-1}, for the QPLS model built in the spectral region 4000–12000 cm^{-1}, R^2 of the training set was slightly lower, and SEC and AARD of the training set were slightly higher. However, the values of R^2, SEP, and AARD of the testing set for the QPLS model built in the spectral region 4000–12000 cm^{-1} were all better than that for the QPLS model built in the spectral region 5000–12000 cm^{-1}. Therefore, the spectral region of 4000–12000 cm^{-1} was chosen for modeling.

3.1.3. The Effects of Different Modeling Ratios on the QPLS Models. After the original spectra were preprocessed using vector correction method, the QPLS models for determination of the germinability of *Pst* urediospores were built in the spectral region 4000–12000 cm^{-1} based on different modeling ratios. The effects of the different modeling ratios between training sets and testing sets on the QPLS models are shown in Table 3. For the QPLS model built when the ratio of the training set to the testing set was 4 : 1, R^2, SEC, and AARD of the training set were 0.9665, 0.04%, and 9.44%, respectively, and R^2, SEP, and AARD of the testing set were 0.8961, 0.07%, and 17.31%, respectively. The results demonstrated that the QPLS model built when the ratio of the training set to the testing set was 4 : 1 was better than other models. Therefore, the ratio of 4 : 1 was chosen as the optimal modeling ratio for building the QPLS model to determine the germinability of *Pst* urediospores. The best QPLS model was obtained as the number of principal components was 8.

3.2. The Effects of Spectral Data Preprocessing Methods, Modeling Ratios, and Spectral Regions on the SVR Models. The prediction results of the optimal SVR models using different preprocessing methods of the near infrared spectra for quantitative determination of germinability of *Pst* urediospores are shown in Table 4. As shown in Table 4, for the optimal SVR model built based on the original spectra, R^2 and MSE of the training set were 0.9475 and 0.002277, respectively, and those of the testing set were 0.8996 and 0.005421, respectively. In contrast, the SVR model built when each of the three wavelet denoising methods was used as the preprocessing method was worse than that built with the original spectra. When vector normalization was used as the preprocessing method, four SVR models with satisfactory effects

Table 2: Influence of different spectral regions on prediction results of QPLS models for quantitative determination of germinability of *Puccinia striiformis* f. sp. *tritici* urediospores.

Spectral region/cm^{-1}	The number of principal components	Training set			Testing set		
		R^2	SEC/%	AARD/%	R^2	SEP/%	AARD/%
4000–5000	3	0.5836	0.13	26.72	0.6083	0.14	16.89
4000–6000	3	0.5194	0.14	28.96	0.4696	0.16	23.84
4000–7000	3	0.4930	0.15	30.41	0.4259	0.16	30.59
4000–8000	4	0.5618	0.14	28.93	0.6174	0.13	18.56
4000–9000	6	0.7306	0.11	24.91	0.8814	0.07	13.42
4000–10000	5	0.6402	0.12	26.16	0.7436	0.11	17.74
4000–11000	8	0.8850	0.07	15.45	0.8785	0.08	16.72
4000–12000	8	0.9665	0.04	9.44	0.8961	0.07	17.31
5000–6000	5	0.6665	0.12	26.81	0.8143	0.09	18.09
5000–7000	4	0.5494	0.14	29.45	0.8216	0.09	16.80
5000–8000	4	0.5562	0.14	30.00	0.7434	0.11	19.36
5000–9000	7	0.7597	0.10	23.74	0.9382	0.05	11.41
5000–10000	5	0.6424	0.12	26.38	0.8888	0.07	13.70
5000–11000	7	0.8419	0.08	19.29	0.8892	0.07	14.96
5000–12000	8	0.9727	0.03	8.92	0.8493	0.08	21.81
6000–7000	6	0.8378	0.08	18.03	0.8565	0.08	18.46
6000–8000	6	0.7228	0.11	23.87	0.8833	0.07	13.15
6000–9000	7	0.8279	0.09	19.22	0.8632	0.08	16.95
6000–10000	6	0.7538	0.10	23.67	0.9034	0.07	13.51
6000–11000	6	0.8014	0.09	21.91	0.9085	0.07	13.77
6000–12000	5	0.7877	0.10	21.50	0.8924	0.07	14.43
7000–8000	5	0.6788	0.12	25.21	0.8051	0.10	16.77
7000–9000	6	0.8309	0.08	18.77	0.9244	0.06	15.06
7000–10000	5	0.7259	0.11	23.80	0.8707	0.08	15.13
7000–11000	4	0.6979	0.11	23.69	0.8990	0.07	14.60
7000–12000	4	0.7897	0.09	21.82	0.8904	0.07	16.61
8000–9000	4	0.8103	0.09	21.18	0.8874	0.07	17.35
8000–10000	4	0.7189	0.11	26.65	0.8694	0.08	20.78
8000–11000	5	0.8231	0.09	19.75	0.8791	0.08	18.67
8000–12000	4	0.7531	0.10	24.59	0.8478	0.08	22.12
9000–10000	4	0.7938	0.09	21.51	0.8550	0.08	20.46
9000–11000	4	0.8038	0.09	20.88	0.8145	0.09	22.65
9000–12000	3	0.6436	0.12	31.45	0.7053	0.12	24.85
10000–11000	4	0.7863	0.10	21.93	0.7605	0.11	26.12
10000–12000	3	0.6754	0.12	29.52	0.7342	0.11	25.76
11000–12000	2	0.6252	0.13	31.85	0.6024	0.14	34.99
Average	5	0.7294	0.10	23.51	0.8088	0.09	18.70

Table 3: Influence of different modeling ratios on prediction results of QPLS models for quantitative determination of germinability of *Puccinia striiformis* f. sp. *tritici* urediospores.

The ratio of training set to testing set	The number of principal components	Training set			Testing set		
		R^2	SEC/%	AARD/%	R^2	SEP/%	AARD/%
1 : 1	6	0.8919	0.06	12.78	0.7585	0.11	28.80
2 : 1	7	0.9046	0.06	14.03	0.7653	0.11	17.27
3 : 1	7	0.9149	0.06	12.71	0.7727	0.11	26.88
4 : 1	8	0.9665	0.04	9.44	0.8961	0.07	17.31
5 : 1	6	0.8288	0.08	18.48	0.8765	0.09	15.70
Average	7	0.9013	0.06	13.50	0.8138	0.10	21.19

Table 4: The prediction results of the optimal SVR models using different preprocessing methods of the near infrared spectra for quantitative determination of germinability of *Puccinia striiformis* f. sp. *tritici* urediospores.

Preprocessing method	The ratio of training set to testing set	Spectral region/cm^{-1}	Optimal parameters		Training set		Testing set	
			C	γ	R^2	MSE	R^2	MSE
No preprocessing	3 : 1	4000–12000	3.0314	5.278	0.9475	0.002277	0.8996	0.005421
Level 1 decomposition denoising using db2 wavelet	3 : 1	4000–12000	3.0314	5.278	0.9464	0.002322	0.8996	0.005422
Level 2 decomposition denoising using db2 wavelet	3 : 1	4000–12000	3.0314	5.278	0.9429	0.002474	0.8978	0.005508
Level 3 decomposition denoising using db2 wavelet	3 : 1	4000–12000	3.0314	5.278	0.9393	0.002630	0.8954	0.005574
Euclidean normalization	3 : 1	7000–12000	84.4485	147.0334	0.9047	0.004231	0.8956	0.005408
Multiplication scatter correction	3 : 1	7000–12000	48.5029	1	0.8925	0.004805	0.8754	0.007024
Standard normalized variate transform	3 : 1	6000–9000	84.4485	0.0359	0.8531	0.006267	0.9065	0.007758
Vector normalization	2 : 1	4000–12000	147.0334	0.3299	0.9340	0.003024	0.9034	0.004604
	5 : 1	8000–11000	27.8576	16	0.9461	0.002464	0.9262	0.003488
	3 : 1	8000–12000	256	0.3299	0.9320	0.003128	0.9129	0.007432
	5 : 1	8000–12000	147.0334	0.5743	0.9462	0.002639	0.9259	0.004726
S-G first derivative transform	3 : 1	5000–9000	147.0334	256	0.9324	0.002947	0.8817	0.007193
S-G second derivative transform	5 : 1	8000–10000	256	256	0.9099	0.004248	0.8519	0.010270

were picked out and shown in Table 4. For the SVR model built when vector normalization was used as the preprocessing method, the ratio of the training set to the testing set was 2 : 1 and 4000–12000 cm^{-1} was chosen as the modeling spectral region, R^2 and MSE of the training set were 0.9340 and 0.003024, respectively, and those of the testing set were 0.9034 and 0.004604, respectively. For the SVR model built when vector normalization was used as the preprocessing method, the ratio of the training set to the testing set was 5 : 1 and 8000–11000 cm^{-1} was chosen as the modeling spectral region, the values of R^2 and MSE of the training set were 0.9461 and 0.002464, respectively, and those of the testing set were 0.9262 and 0.003488, respectively. For the SVR model built when vector normalization was used as the preprocessing method, the ratio of the training set to the testing set was 3 : 1 and 8000–12000 cm^{-1} was chosen as the modeling spectral region, the values of R^2 and MSE of the training set were 0.9320 and 0.003128, respectively, and those of the testing set were 0.9129 and 0.007432, respectively. For the SVR model built when vector normalization was used as the preprocessing method, the ratio of the training set to the testing set was 5 : 1 and 8000–12000 cm^{-1} was chosen as the modeling spectral region, R^2 and MSE of the training set were 0.9462 and 0.002639, respectively, and those of the testing set were 0.9259 and 0.004726, respectively. The results indicated that the prediction effects of the four SVR models above for the training sets were worse and that for the testing sets were better in comparison with the optimal SVR model built based on the original spectra. Among these four models, the SVR model built when vector normalization was used as the preprocessing method, the ratio of the training set to the testing set was 5 : 1, and 8000–11000 cm^{-1} was chosen as the modeling spectral region was the best. The effect of the optimal SVR model built when Euclidean normalization, multiplication scatter correction, standard normalized variate transform, S-G first derivative transform, or S-G second derivative transform was used as the preprocessing method was worse than that of the optimal SVR model built based on the original spectra, and the optimal SVR model built when S-G second derivative transform was used as the preprocessing method was the worst among them. In contrast with other SVR models, for the SVR model built when vector normalization was used as the preprocessing method, the ratio of the training set to the testing set was 5 : 1 and 8000–11000 cm^{-1} was chosen as the modeling spectral region, R^2 of the training set was relatively high, MSE of the training set was relatively low, R^2 of the testing set was the highest, and MSE of the testing set was the lowest; in addition, the spectral range for modeling was relatively small. Therefore, this SVR model was regarded as the best SVR model for quantitative determination of germinability of *Pst* urediospores.

4. Conclusions and Discussion

In this study, the effect of the optimal QPLS model or the optimal SVM model built for quantitative determination of germinability of *Pst* urediospores using near infrared spectroscopy technology was satisfactory. The results indicated that it is feasible to rapidly and nondestructively determine the germinability of *Pst* urediospores by using near infrared spectroscopy technology. A new method based on NIRS was provided for rapid, automatic, and nondestructive determination of germinability of *Pst* urediospores in this study. Meanwhile, a reference was provided for nondestructive

determination of spore germinability of other kinds of pathogens.

When the germination rate measurement models of *Pst* urediospores were built using QPLS, the effects of different preprocessing methods of the original spectral data, different spectral regions, and different ratios between training sets and testing sets on modeling were analyzed. The results showed that the best QPLS model was obtained when vector correction was used as the preprocessing method, the spectral region of 4000–12000 cm^{-1} was selected as the modeling spectral region, the ratio of the training set to the testing set was 4 : 1, and the number of principal components was 8. For this model, R^2, SEC, and AARD of the training set were 0.9665, 0.04%, and 9.44%, respectively, and R^2, SEP, and AARD of the testing set were 0.8961, 0.07%, and 17.31%, respectively. When the germination rate measurement models of *Pst* urediospores were built using SVR, the effects of different preprocessing methods, different spectral regions, and different ratios between training sets and testing sets on modeling were also analyzed. The results showed that the best SVR model was built when vector normalization was used as the preprocessing method, the ratio of the training set to the testing set was 5 : 1, and the spectral region of 8000–11000 cm^{-1} was selected as the modeling spectral region. For this SVR model, the values of R^2 and MSE of the training set were 0.9461 and 0.002464, respectively, and those of the testing set were 0.9262 and 0.003488, respectively. The results demonstrated that QPLS and SVR could be used to build the model for nondestructive and quantitative determination of germinability of *Pst* urediospores based on near infrared spectra.

SVR is suitable to solve small sample, nonlinearity, high dimension, and other complex problems [22]. Moreover, the spectral range of 8000–11000 cm^{-1} in which the best SVR model was built was smaller than the spectral range of 4000–12000 cm^{-1} in which the best QPLS model was built in this study. Therefore, in the practical application, it is preferred to use SVR to build the model for quantitative determination of germinability of *Pst* urediospores. In addition, there are many factors that can affect the germinability of the *Pst* urediospores. In particular, ultraviolet radiation, temperature, humidity, and many other environmental factors can influence the vitality and the survival of *Pst* urediospores during their long distance dispersal [2, 6, 23]. Further studies are needed to determine whether these factors would affect the relationship between the near infrared spectra and the germinability of *Pst* urediospores.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

Authors' Contribution

Yaqiong Zhao and Feng Qin contributed equally to this paper.

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