Determination of pyraoxystrobin in technical material and formulations by HPLC

ABSTRACT: A high performance liquid chromatography method has been established for the analysis of pyraoxystrobin in technical material and formulations. The separation was carried out on a reverse phase C_{18} column(150mm×4.6 μ m i.d.) The analysis was complete in less than 8 min in Mobile phase: ψ (CH_3CN+H_2O)=60+40 at 280 nm of detection wavelength. A good linearity was obtained for pyraoxystrobin in the range from 200 to 2500 mg/L with correlation coefficient were 1.0000. The average recoveries were in the range of 99.3-100.4 and 99.2-101.2, relative standard deviations (RSDs) of pyraoxystrobin were 0.40% .

Key words: HPLC; pyraoxystrobin; analysis

The IUPAC name of pyraoxystrobin is (E)-2-(2-(((3-(4-chlorophenyl)-1-methyl-1H-pyrazole-5-yl)-oxo)-methyl) phenyl)-3-methoxy-methacrylate. Pyraoxystrobin is developed by Shenyang Research Institute of Chemical Industry, P.R. China. Its patent was filed in 2004 by Shenyang Research Institute of Chemical Industry (patent No. ZL200410021172.3). It is a fungicide developed for control of Erysiphe graminis on wheat and Pseudoperonospora cubensis on cucurbitaceae.

1. Experiment

1.1. Apparatus and Reagents

Liquid chromatograph: Waters 2489 liquid chromatograph, with DAD UV detector Computer for data treatment;

HPLC- Column: 150 mm × 4.6 mm(id) stainless steel column packed with Agilent SB-C_{18} , 5 μm;

Acetonitrile: HPLC grade, Sinopharm Chemical Reagent Shenyang Co., Ltd;

Water: New second distilled water;

Pyraoxystrobin std: 99.7% (State Key Laboratory of the Discovery and Development of Novel Pesticide);

1.2. HPLC operating conditions

Mobile phase: $\psi(CH_3CN+H_2O)=60+40$, filtered and degassed;

Flow rate: 1.3 mL/min;

Column temperature: room temperature;

Detector wavelength: 280 nm;

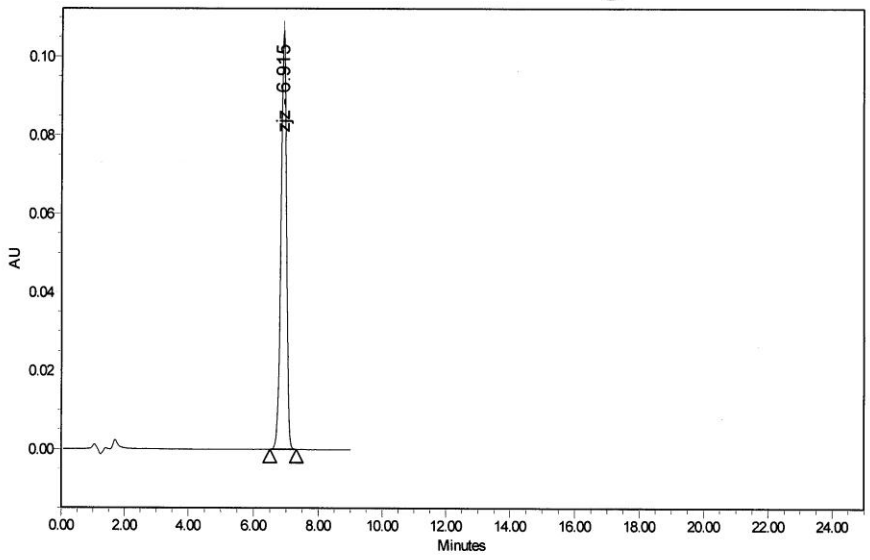
Injection volume: 5 μL;

Retention time: pyraoxystrobin 6.915 min (The typical liquid chromatogram for pyraoxystrobin TC is shown in Fig.1.)

SAMPLE INFORMATION

Sample Name:	b2	Acquired By:	chemist
Sample Type:	Unknown	Sample Set Name:	20120315zjzscjmdhsl
Vial:	6	Acq. Method Set:	yl
Injection #:	1	Processing Method:	zjz
Injection Volume:	5.00 ul	Channel Name:	W2489 ChA
Run Time:	9.0 Minutes	Proc. Chnl. Descr.:	W2489 ChA 280nm
Date Acquired:	2012-3-15 15:04:51 CST		
Date Processed:	2012-3-23 15:58:35 CST		

Auto-Scaled Chromatogram



Peak Results

	SampleName	Name	RT	Injection	Vial	Area	Height	% Area
1	b2	zjz	6.915	1	6	1263734	106754	100.00

Fig.1 HPLC chromatogram of pyraoxystrobin

1.3. Operating procedure

1.3.1. Preparation of calibration solution

Weigh 0.1 g (to the nearest 0.00002 g) of pyraoxystrobin standard material into a 50 mL-volumetric flask, add acetonitrile, Ultrasonic to dissolve it, then dilute to mark with acetonitrile and mix well. Transfer by pipette 10.0 mL of these solutions into separate volumetric flasks (50 mL), dilute with acetonitrile and fill to the mark.

1.3.2. Preparation of sample solution

Weigh sufficient sample to contain 0.1 g (to the nearest 0.00002 g) of pyraoxystrobin into a 50 mL-volumetric flask, add acetonitrile, Ultrasonic to dissolve it. Transfer by pipette 10.0 mL of these solutions into separate volumetric flasks (50 mL), dilute with acetonitrile and fill to the mark.

1.3.3. Determination

Under the operating conditions mentioned above, successively inject calibration solution for a few times. Then carry out the determination according to the order: calibration solution, sample solution, sample solution, calibration solution..

1.3.4. Calculation

Mass fraction X_1(%) of pyraoxystrobin in the sample is calculated by formula (1):

$$X_1 = \frac{A_2 \times m_1 \times p}{A_1 \times m_2} \quad \cdots\cdots (1)$$

Where:

A_2=mean value of peak areas of pyraoxystrobin in sample solutions;

A_1=mean value of peak areas of pyraoxystrobin in calibration solutions;

m_1=mass of pyraoxystrobin standard material taken, g;

m_2=mass of sample taken, g;

P= purity of pyraoxystrobin standard material, %.

2. Result and Conclusion

2.1. Linearity range test

Prepare 6 sample known content, the range of the concentrations for the samples is 0.1~6.2 times of that of calibration solution above.

The results obtained are given in Fig.2,

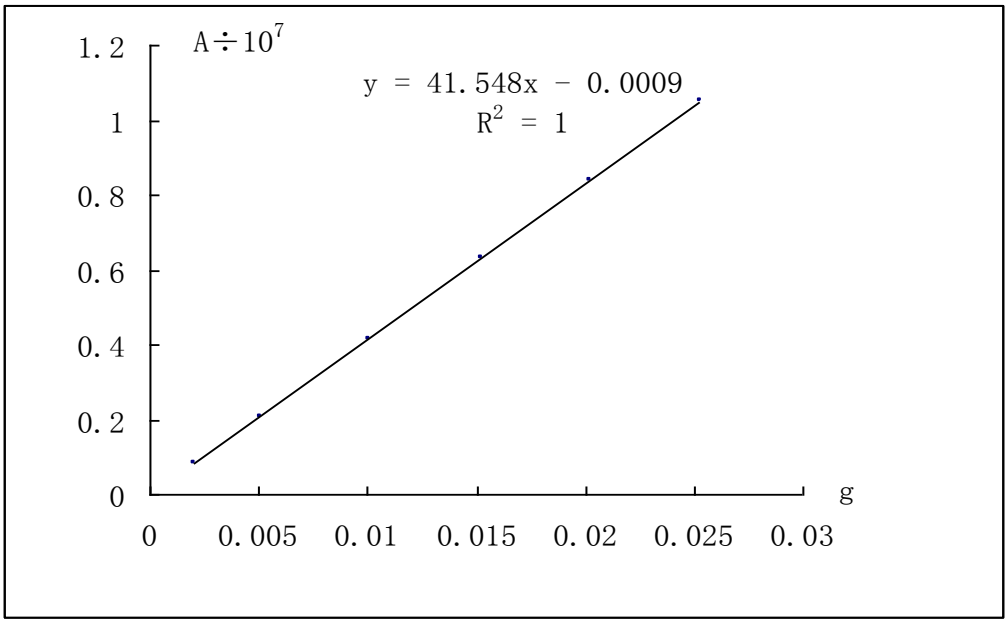


Fig.2 pyraoxystrobin linearity relativity on UV detector

The relation coefficient is 1.0000 based on the calculation, therefore the linearity between the mass and peak area is good in the concentration range above.

2.2. Consistency of System Performance

Prepared a solution with pyraoxystrobin sample, dilute the solution with acetonitrile, Under the operating conditions mentioned above, successively inject solution for 6 times.

2.3. Precision of the method

Determinate the contents of the TC samples, the results of pyraoxystrobin content in the sample (The precision test) obtained are given in table 1.

Table 1 the results of determination of pyraoxystrobin in the pyraoxystrobin TC

(The precision test)

No. of sample	1	2	3	4	5	6
Content，%	97.71	97.11	98.11	97.30	98.01	97.38
Average ,%	97.6					
Standard deviation	0.39					
Relative standard deviation，%	0.40					

2.4. Accuracy of the method

Prepare 6 samples by means of mixing a certain amount of pyraoxystrobin. Material with the sample known content. Determine the

total amount of pyraoxystrobin by the HPLC method and calculate the recovery. The results obtained of pyraoxystrobin in technical material are given in table 2, The results obtained of pyraoxystrobin in suspension concentrates are given in table 3.

Table 2 the results of the recovery test (pyraoxystrobin material Std 99.7%, sample 97.6%)

No.	Mass in the sample，g	Mass in the std. Material，g	pyraoxystrobin mass in the sample, g	Total mass found，g	Recovery，%	Mean value，%
1	0.01367	0.04991	0.02094	0.02102	100.38	99.9
2	0.02157	0.04052	0.02118	0.02020	100.09	
3	0.03351	0.03132	0.02120	0.02114	99.72	
4	0.04382	0.02188	0.02068	0.02063	99.76	
5	0.05335	0.01059	0.02047	0.02029	99.12	
6	0.05951	0.00574	0.01940	0.01942	100.10	

Table 3 the results of the recovery test (pyraoxystrobin Std 99.7%, sample 20.3%)

No.	Mass in the sample，g	Mass in the std. Material，g	pyraoxystrobin mass in the sample, g	Total mass found，g	Recovery，%	Mean value，%
1	0.01612	0.00234	0.01846	0.01844	99.89	99.9
2	0.01796	0.00491	0.02287	0.02275	99.48	
3	0.01189	0.00853	0.02042	0.02047	100.24	
4	0.00950	0.01244	0.02194	0.02184	99.54	
5	0.00537	0.01735	0.02272	0.02300	101.23	
6	0.00291	0.01828	0.02119	0.02101	99.15	

2.5. Method evaluation

2.5.1. Determination of the specificity for quantitation of pyraoxystrobin

The specificity of pyraoxystrobin was determined by testing peak purity using diode array detector. Prepare a pyraoxystrobin TC sample

solutions, determination the chromatogram in sample solutions according to analysis condition of the HPLC-DAD for determining the specificity of pyraoxystrobin TC. The purity angle of pyraoxystrobin is 2.553 the threshold of the noise is 2.916, the purity angle is lower than the threshold of the noise, the specificity pass.

2.5.2. System suitability tests for quantitation of pyraoxystrobin

2.5.2.1.Chromatographic suitability test

Prepared a solution with pyraoxystrobin TC sample , Determination according to analysis condition of determination pyraoxystrobin in pyraoxystrobin TC samples. The determined pyraoxystrobin should be evaluated for tailing factor, theoretical plates and resolution from the nearest eluting peaks. Results obtain are given in table 4.

Table 4 The result of chromatographic suitability test of pyraoxystrobin

item	result	criteria	Pass /fail
theoretical plates, *N*	1.4×10^4	>2000	pass
tailing factor, *T*	1.1	<2	pass
Resolution, *R*	4.9	>1.5	pass

2.5.3. Consistency of system performance of pyraoxystrobin

Consistency of system performance was determined by replicating injections a typical standard solution 6 times, Calculated the relative standard deviation of the analytical results. The RSD％ for area of pyraoxystrobin should be less than 1.2 %, results obtain are given in table

5.

Table 5 The result of consistency of system performance test of pyraoxystrobin

No.	Peak Area of component A	Average of peak	RSD,%
1	6050299	6087687	0.4
2	6083921		
3	6093378		
4	6082929		
5	6102482		
6	6113115		

3. Conclusion

The precision, the evaluation and accuracy of the method is good based on 2.1~2.5, The method is considered appropriate for the determination of pyraoxystrobin in technical and suspension concentrates product.