Advance Journal of Food Science and Technology 12(1): 31-41, 2016
DOI:10.19026/ajfst.12.2832
ISSN: 2042-4868; e-ISSN: 2042-4876

Submitted: August 17, 2015 Accepted: September 7, 2015 Published: September 05, 2016

Research Article

Study on Fluorescence Spectrum of Pesticide and Serum in Interaction

[1, 2]Wang Lexin, [1]Gao Tianyi, [1]Chen Danping, [1]Wang Chang and [1]Zhang Jinyan
[1]Heilongjiang Bayi Agricultural University, Heilongjiang, Daqing, 163319,
[2]Nanjing Aeronautics and Astronautics University, Jiangsu, Nanjing, 210016, China

Abstract: The fluorescence spectrum of different serum samples added at the same concentrations with different pesticides (seed coated agent, herbicide and the flapping lice spirit) has been studied when 260, 280 and 320 nm, respectively exciting lights are used to excite the serum. It is found by experiment that when the same pesticide is added to the serum in different biochemical indexes, the fluorescence spectrum peak changes differently. The red shift occurs for the position of the fluorescence peak when the different pesticides are added to the serum in the same biochemical index. For the serum in different biochemical indexes added with the pesticides, the fluorescence intensity changes differently. The aim of study was to determine the type of pesticides based on the change of the serum fluorescence intensity. The experimental results show that pesticides have a great influence on the fluorescence of the serum in different biochemical indexes. The further research results will provide an experimental basis for first aid and drug use for patients who are suffered from pesticide or herbicide poisoning in different biochemical indexes.

Keywords: Fluorescence spectrum, herbicide, seed coated with pesticide, serum, the flapping lice spirit

INTRODUCTION

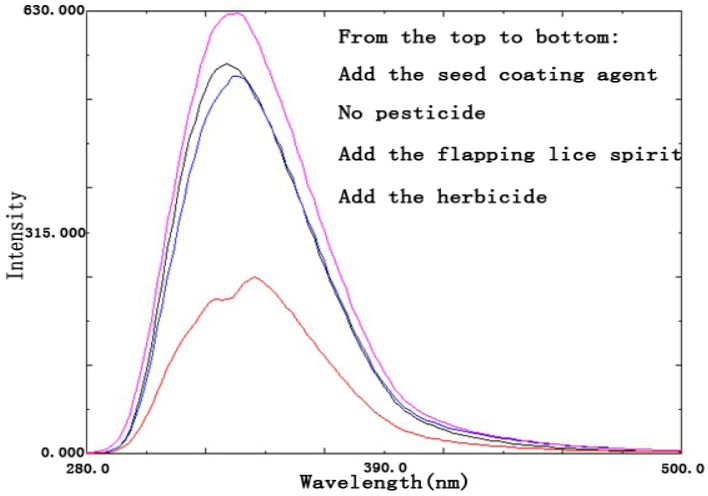
The fluorescence spectrometry as a kind of analysis method is widely used in various fields of industry, agriculture, medicine, health, judicial authentication and scientific research. Detecting pesticide residues pollution in combination with optical fiber sensing and computer signal processing technology is becoming a research focus in the world in this field (Xie *et al*., 2010; Wang *et al*., 2010; Ma *et al*., 2006). The research on fluorescence analysis of pesticides was started by foreign scholars in the 1990s. They had successfully detected fluorescence spectrum of several pesticides by making use of natural fluorescence properties of some pesticides with the help of some fluorescent dyes. Meanwhile, they developed various analyzers based on optical fiber sensing technology (Tanojo *et al*., 2000; Armenta *et al*., 2007). Harris *et al*. (1998) determined the pesticide residues of dichloro ben oxyacetic acid in water using the optical fiber fluorescence immunoassay method. Hassoon and Schechter (2000) developed a fluorescence probe for measurement of DDT pesticides and determination of DDT residues in soil. JiJi *et al*. (1999) applied the Excitation Emission Matrix (EEM) fluorescence spectrum for the quantitative detection of trace PAHs and carbamate pesticide residues. At present, the rapid detection technology of pesticide residues based on laser-induced fluorescence spectrum analysis is just in a stage of principle and experimental research at home and abroad. Sun *et al*. (2003) used laser-induced fluorescence technology to measure the fluorescence spectrum of five pesticides such as tetrachlorvinphos and conducted a preliminary qualitative research on pesticide fluorescence detection. Che and Wang (2004) has developed a fiber optic spectrometer to monitor imidacloprid pesticide. Wang *et al*. (2006) detected the carbamate pesticide residues using the three-dimensional fluorescence technology. Zhang *et al*. (2006) studied the carbaryl spectral characteristics by the synchronous fluorescence spectrum analysis and determined its concentration. Most of these pesticides in question are organic compounds. As molecular structure of different pesticides is different, each fluorescent material can give off specific fluorescence spectrum. In the same excitation, different pesticides give off the fluorescence spectrum in different intensity and show different shapes. Because the fluorescence spectrum of different pesticides is significantly different in distribution, time delay and formation efficiency, the kind and concentration of pesticides can be detected by studying photo-induced fluorescence characteristics (Yang and Gong, 2006; Li *et al*., 2004). This research provides a good foundation for fluorescence spectrum analysis applied for real-time and rapid detection of pesticide residues as well as further development of fluorescence spectrum technology. Reports of pesticide and serum

Corresponding Author: Wang Lexin, Heilongjiang Bayi Agricultural University, Heilongjiang, Daqing, 163319, China

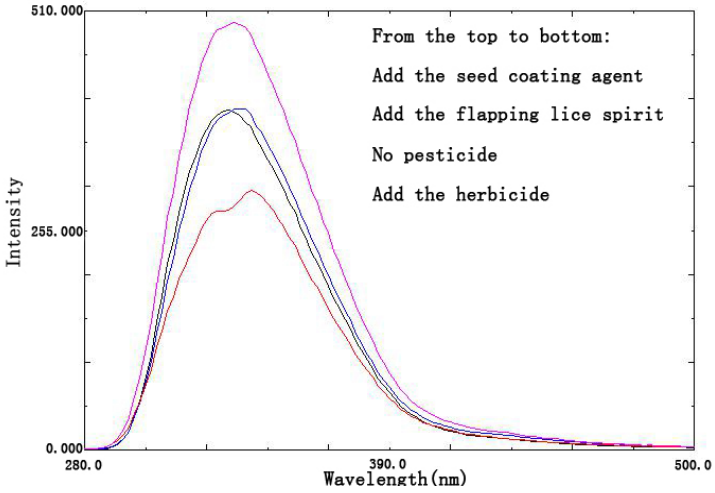
interaction are rare though many spectrum detections related to pesticides have been reported. This study deals with the spectrum law of interaction between pesticides and serums in different biochemical indexes.

SAMPLES AND METHOD

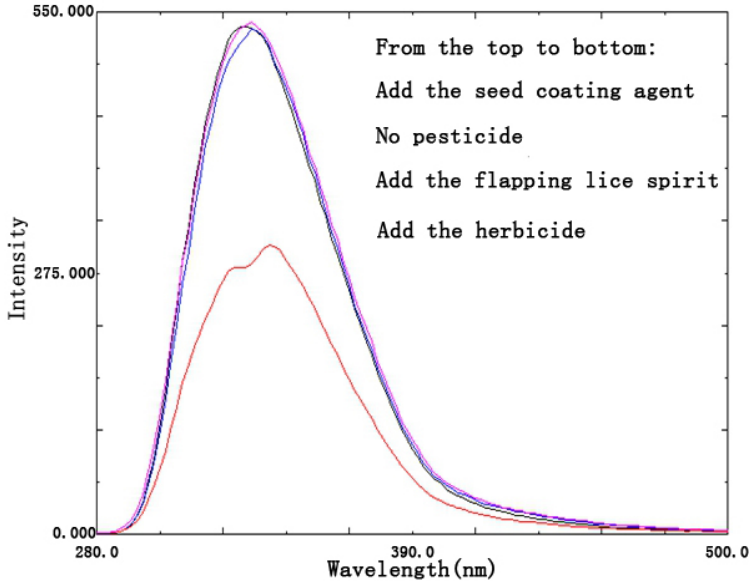
- Experimental serum samples were taken from healthy male patients suffered from high blood lipid and high blood glucose a total of 54 subjects by the vein blood sampling when they were in an empty stomach in early morning. Take the upper serum and prepare the 1:20 serum solution with pure water for testing use.
- The pesticides for the experiment were: Imidacloprid: 10% wettable powder (made by Jiangsu Changqing Agrochemical Co., Ltd.); dichloro•Benzyl: 36% wettable powder (Jiangsu Changqing Agrochemical Co., Ltd.); and 35% seed coating agent (EN Industry Corporation). In the experiment, dissolve 0.2 g active compound by weighing pesticides with distilled water and dilute the solution to 100 mL. After that, take the supernatant liquid and put it into a water bath at a constant temperature of 25°C for use.
- In the experiment, the Shimadzu RF5301 fluorescence spectrophotometer was used to detect the fluorescence spectrum of selected serum samples and serum samples containing pesticides. Take a 3 mL tested sample with a cuvette in the test. According to the early stage of the experiment, studied the effect of wavelength on serum fluorescence excitation (Published in 2008, the tenth period《spectroscopy and spectral analysis》). Therefore, this experimental selected excitation wavelengths respectively were 260, 280 and 320 nm, respectively the accepted wavelength ranges from 280 to 600 nm and the slit width of excitation and emission was 3 nm. The sampling interval was 0.5 nm, with a medium speed automatic scanning every 5 nm.



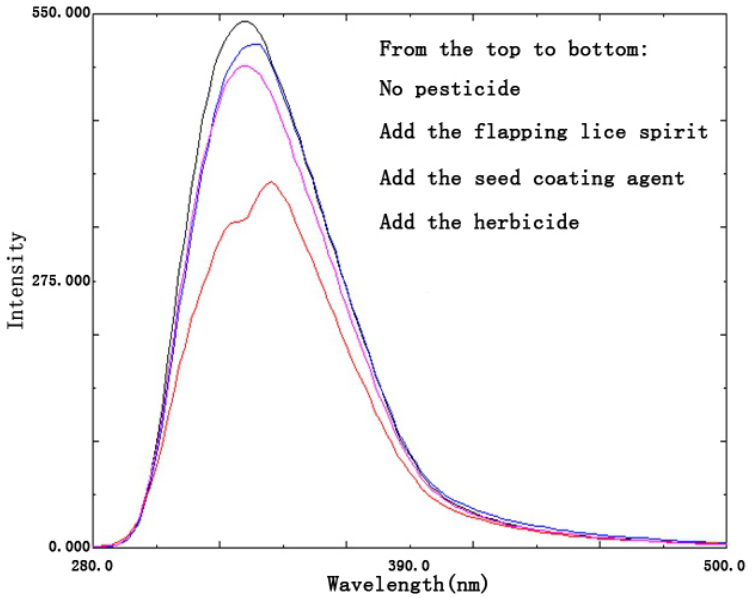
(a) High blood glucose



(b) Normal serum



(c) High cholesterol



(d) High blood lipid

Fig. 1: The fluorescence spectrum of the serum samples added with pesticides at the same concentration (Ex: 260 nm)

RESULTS AND DISCUSSION

In order to study effect of pesticides in different biochemical indexes of serum, the fluorescence spectrum of samples in normal biochemical indexes, high blood glucose, high cholesterol and high triglycerides was measured separately by spectrometer. Serum samples respectively were added to pesticides at the same concentration and the fluorescence spectrum was measured. The experimental results are shown in Fig. 1 to 3. According to the variation of the fluorescence intensity of the spectral lines, the fluorescence intensity changes of the different serum samples added with different pesticides at the same concentration was listed after the spectral analysis as shown in Table 1 to 3. The experimental results are analyzed as follows:

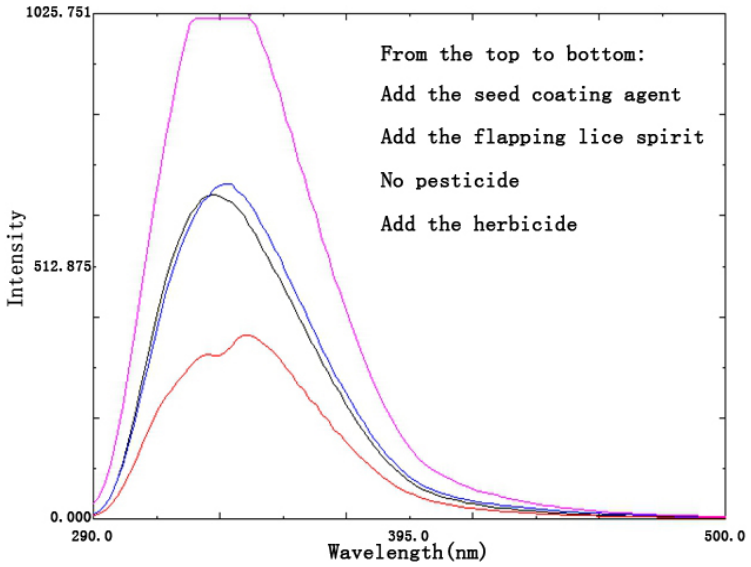
- Figure 1 shows that the fluorescence spectra change differently when 260 nm light excitation serum and the same pesticides are added to the serum in different biochemical indexes. When the different pesticides are added to the same serum, the fluorescence spectrum changes also differently.

Table 1: (a) The fluorescence intensity changes of the serum samples added with pesticides at the same concentration (Ex: 260 nm)

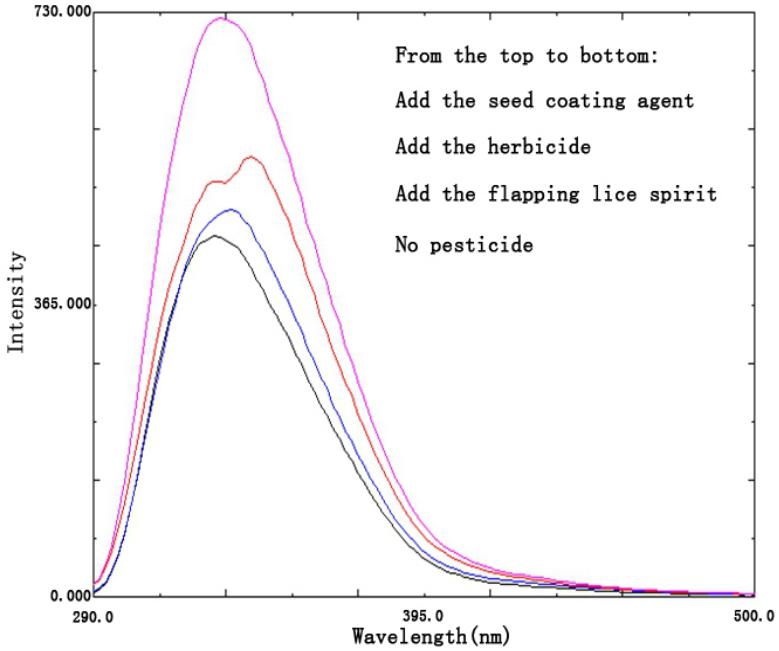
	Normal serum				High blood glucose			
Samples	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit
Peak position (nm)	332	334	340	336	332	336	342	336
Fluorescence intensity	394.4	496.9	301.8	396.7	555	626	250	537
Intensity difference		102.5	-92.6	2.3		71	-305	-18

(b) The fluorescence intensity changes of the serum samples added with pesticides at the same concentration (Ex: 260 nm)

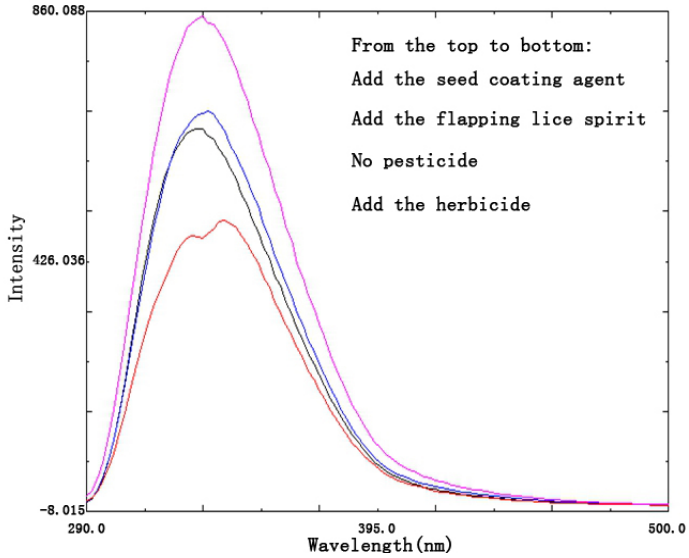
	High cholesterol				High blood lipid			
Samples	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit
Peak position (nm)	332	334	340	334	334	332	342	336
Fluorescence intensity	534.7	540.1	305	532	542.7	496.6	377.3	518.3
Intensity difference		5.4	-229.7	-2.7		-46.1	-165.4	-24.4



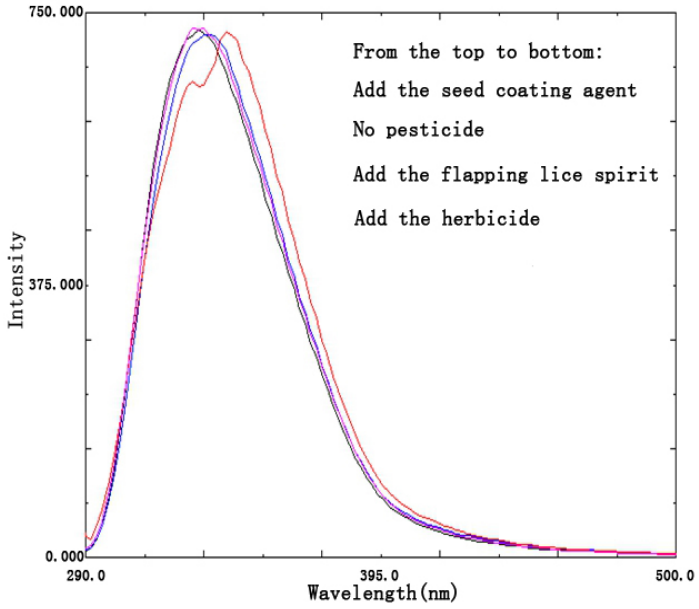
(a) High blood glucose



(b) Normal serum



(c) High cholesterol



(d) High blood lipid

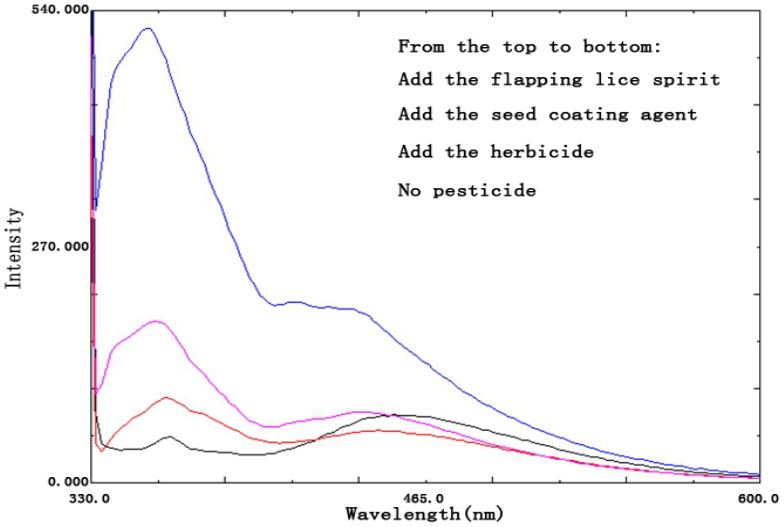
Fig. 2: The fluorescence spectrum of the serum samples added with pesticides at the same concentration (Ex: 280 nm)

Table 2: (a) The fluorescence intensity changes of the serum samples added with pesticides at the same concentration (Ex: 280 nm)

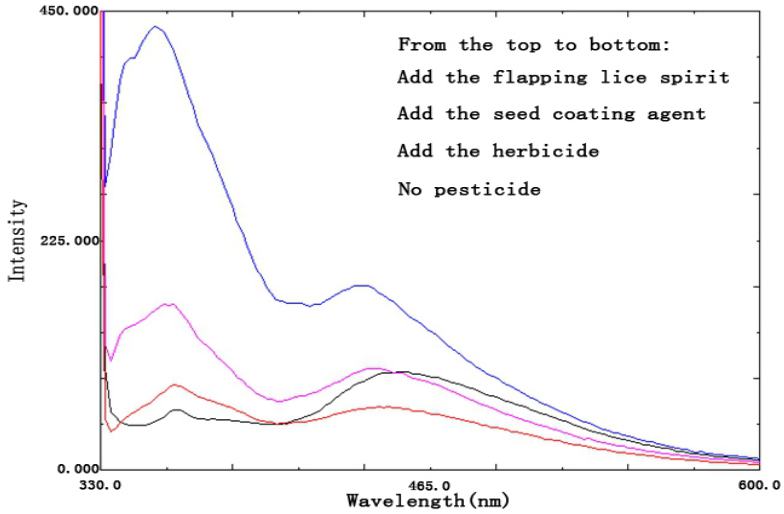
	Normal serum				High blood glucose			
Samples	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit
Peak position (nm)	328	330	340	334	330	342	340	336
Fluorescence intensity	450.2	722.8	549.8	484.4	657.5	1015	373.1	679.8
Intensity difference		272.6	99.6	34.2			-284.4	22.3

(b) The fluorescence intensity changes of the serum samples added with pesticides at the same concentration (Ex: 280 nm)

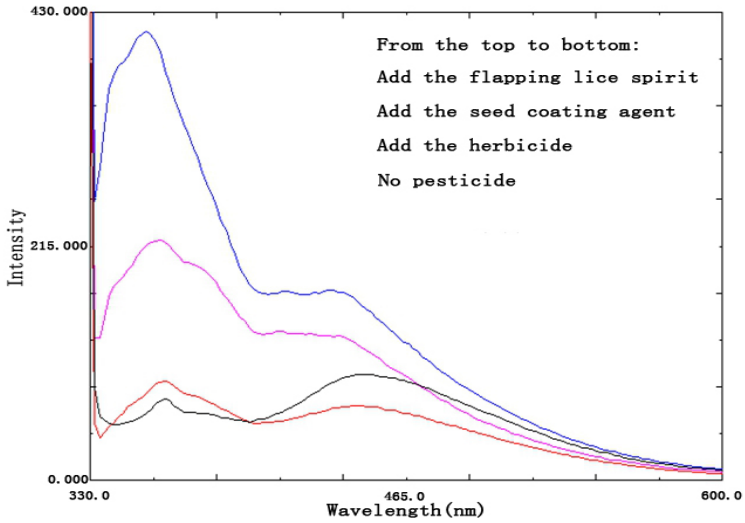
	High cholesterol				High blood lipid			
Samples	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit
Peak position (nm)	332	332	340	334	330	332	340	334
Fluorescence intensity	656	851.6	497	686	727	728	722	720
Intensity difference		195.6	-159	30		1	-5	-7



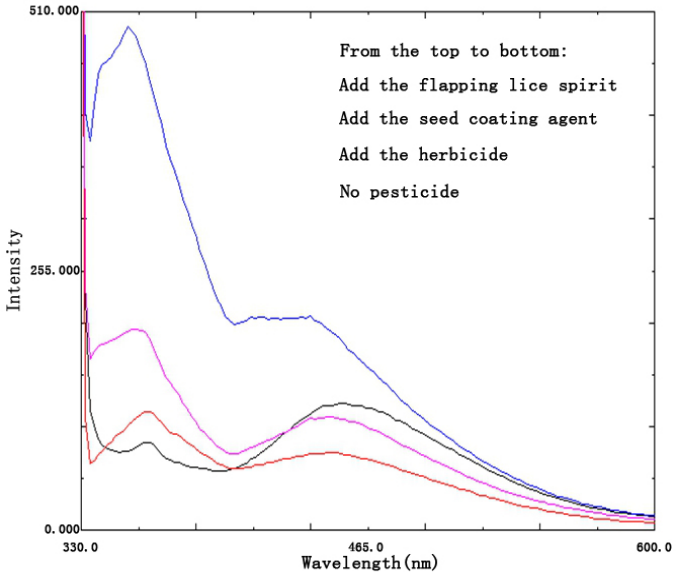
(a) High blood glucose



(b) Normal serum



(c) High cholesterol



(d) High blood lipid

Fig. 3: The fluorescence spectrum of the serum samples added with pesticides at the same concentration (Ex: 320 nm)

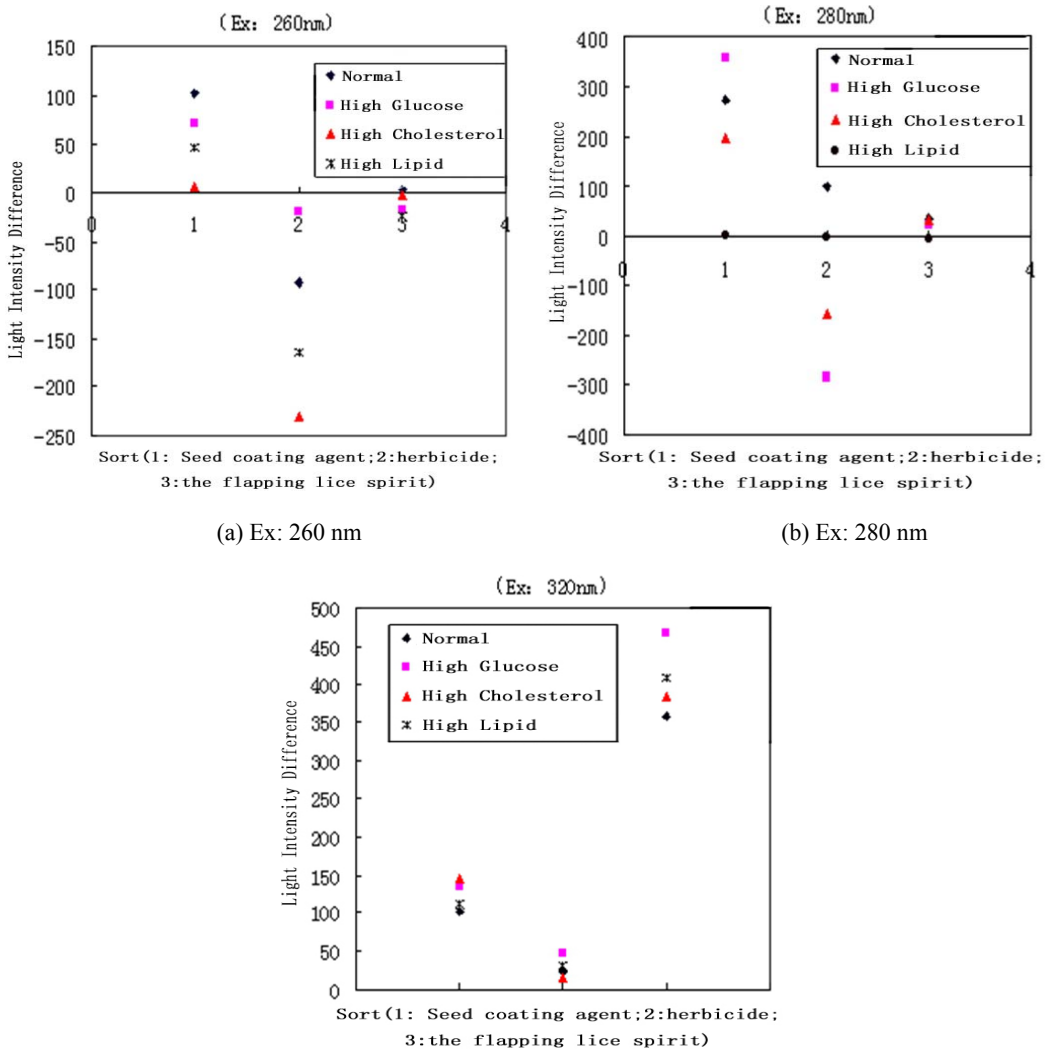
Table 3: (a) The fluorescence intensity changes of the serum samples added with pesticides at the same concentration (Ex: 320 nm)

	Normal serum				High blood glucose			
Samples	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit
Peak position (nm)	362; 452	358	360; 448	352; 438	362; 452	355; 440	360; 446	353
Fluorescence intensity	59.3; 96.3	162.5	83.4; 61.6	435; 180.7	52; 77.6	186; 81.4	98.7; 60	520
Intensity difference		103.2	24.1; -34.7	375.7; 84.4		134; 3.8	46.7; -17.6	468

(b) The fluorescence intensity changes of the serum samples added with pesticides at the same concentration (Ex: 320 nm)

	High cholesterol				High blood lipid			
Samples	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit	No pesticide	Seed coating agent	Herbicide	The flapping lice spirit
Peak position (nm)	362; 446	360; 412	362; 444	354	360; 452	354; 448	360; 450	352
Fluorescence intensity	74.3; 97.1	220.6; 137	91; 68.7	412.6	86.5; 124.4	198.7; 111.4	116.3; 76.3	495.5
Intensity difference		146.3; 39.9	16.7; -28.4	338.3		112.2; -13	29.8; -48.1	409

The red shift occurs for fluorescence peak and the amount of motion for the serum in different biochemical indexes is different after the serum is added with pesticides as shown in Table 1. Especially 2 nm blue shift occurs for high triglyceride serum after seed coating agent is added. After added with herbicides, the high glucose serum has a smaller peak at 327 nm than that of normal serum, while the high blood lipid (including high cholesterol and triglyceride) serum has a bigger peak than that of normal serum. After different pesticides are added, the fluorescence intensity is different as shown in Fig. 2a. Figure 3a shows, the fluorescence intensity increases at near 330 nm after the seed coating agent is added in serum. This is because when the seed coating agent is excited with 260 nm excitation light, the fluorescence also occurs at near 330 nm, resulting in an increase of fluorescence intensity rather than quenching at wavelength. After seed coating agent is added in serum in different biochemical indexes, there is a different increase of fluorescence intensity. Compared with the normal serum, the serum of patients with hyperglycemia and high cholesterol has a smaller increasing intensity, indicating that added seed coating agent has a great impact. After herbicides and the flapping lice spirit are added, the fluorescence intensity is weakened



(a) Ex: 260 nm

(b) Ex: 280 nm

(c) Ex: 320 nm

Fig. 4: The fluorescence intensity changes of different serum samples added with different pesticides

at near 330 nm in different degrees. Obviously, herbicides have a big impact and the flapping lice spirit has a small impact on serum. For serum in different biochemical indexes after added herbicides, the fluorescence intensity decreases in different degrees. Compared with normal serum, hyperglycemia is weakened in a smaller degree than that of normal one and high cholesterol serum decreases in a larger degree than normal one, indicating the serum in different biochemical indexes is impacted by herbicides to different extents. It may be because pesticide molecules are reacted with serum protein molecules after they come into the serum, so that the fluorescence quenching occurs. Compared with normal serum, patients' serum molecular structure and environment change a little, resulting in fluorescence quenching to some extent when pesticides are interacted with serum molecules. After the serum in different biochemical indexes is added to the flapping lice spirit, there is a little impact on normal serum and the fluorescence intensity of hyperglycemia and high cholesterol decreases a little, indicating there is little fluorescence quenching for the flapping lice spirit and serum molecules.

- It is seen from Fig. 2 that the fluorescence spectrum changes differently when 280 nm light excitation serum and the same pesticide are added to the serum in different biochemical indexes. The serum fluorescence spectrum changes differently when different pesticides are added to the same serum. The red shift occurs for fluorescent peak. The quantity of serum motion is different when the serum in different biochemical indexes is added with pesticides. As shown in Table 2, a 10 nm blue

shift longer occurs than other serums after the high blood glucose serum is added with seed coating agent. It is known from Fig. 3b that the fluorescence intensity changes differently after the serum is added with different pesticides. The fluorescence intensity increases at near 330 nm for normal and high blood glucose and high cholesterol serums after they are added with seed coating agent, while the fluorescence intensity for high triglyceride serum is nearly unchanged. This may be because the fluorescence occurs at near 330 nm when the seed coating agent is excited by 280 nm exciting light. It is offset with the fluorescence quenching caused by the interaction of seed coating agent and serum molecules. This indicates further the triglyceride is stronger reacted with seed coating molecule than other molecules, with a great influence on fluorescence intensity. The fluorescence intensity at near 330 nm increases for normal serum after the herbicide is added, while the fluorescence intensity decreases to different extents for high blood glucose and high blood lipid. The fluorescence intensity changes greatest for high blood glucose and it is followed by the high cholesterol; the fluorescence intensity changes smallest for high triglycerides. This further indicates the serum in different biochemical indexes is impacted by herbicides differently. Compared with 260 nm excitation light, the fluorescence intensity increases for the normal and high blood glucose and high cholesterol serums after the serum in different biochemical indexes is added with the flapping lice spirit, but the fluorescence intensity decreases a little for high triglyceride serum. This suggests that the fluorescence intensity of the flapping lice spirit exerts a strong impact on serum.

- It is seen from Fig. 3 that the fluorescence spectrum changes differently when 320 nm light excitation serum and different pesticides are added into the same serum and the fluorescence intensity at 360 nm increases. The blue shift occurs for the fluorescent peak and the quantity of motion is different after the serum in different biochemical indexes is added to pesticides as shown in Table 3. The fluorescence spectrum changes after the same pesticide is added to the serum in different biochemical indexes. After normal serum is added in the flapping lice spirit, there is a peak at 448 nm, but no peak for high blood glucose and high blood lipid serums, instead, forming a wide shoulder peak of 404-440 nm. After the high cholesterol is added in seed coating agent, the shape of its spectral lines changes significantly compared with that of normal serum, forming a wide shoulder peak of 400-436 nm. There is an intersection point between the spectra of the serum added and seed coating agent, but the position of the intersection is different for fluorescence spectrum of the serum in different biochemical indexes. It is intersected at 450 nm for the fluorescence spectrum of normal serum, 450 nm for high blood glucose serum, 470 nm for, high cholesterol serum and 427 nm for high triglyceride serum. The fluorescence spectrum of the serum added with herbicide is intersected with that of normal serum at 404 nm, 421 nm for high blood glucose serum, 398 nm for high cholesterol serum and 402 nm for high triglycerides.

  It is known from Fig. 3c that the serum fluorescence intensity increases at 362 nm when 320 nm light excitation serum is added with different pesticides. The degree of enhancement is different for different pesticides. The fluorescence intensity increases greatest after the serum is added with the flapping lice spirit. This is mainly because when the 320 nm light is excited, the pesticide has the strongest fluorescence itself and the degree of fluorescence intensity quenching is far less than the glow of pesticides after the serum is added with pesticides. The fluorescence intensity increases to some extent when the serum is added with seed coating agent and the intensity is minimum for the herbicide added. The fluorescence intensity increases greatest for high blood glucose serum added with the flapping lice spirit; it is followed by the high triglycerides serum and the last one is high serum cholesterol serum. It suggests that the biochemical indexes of the serum added with pesticide still exert a great impact on the fluorescence of serum.
- It is seen from Fig. 4 that the type of pesticides added in the serum can be judged based on changes of the serum fluorescence strength. When the 260 nm excitation light is used to excite the serum, the seed coating agent has the strongest fluorescence intensity after the pesticide is added. The herbicide has the strongest fluorescence quenching to the greatest extent, but the flapping lice spirit has a little fluorescence quenching. Especially when the herbicide is added, the high blood glucose serum has the less fluorescence strength quenching than normal serum and the high blood lipid serum has the greater fluorescence strength quenching. When the serum is excited with the 280 nm light, the fluorescence strength increases greatest for seed coating agent when the pesticide is added, the fluorescence strength increases less for the flapping lice spirit, but the strength increases weakly for herbicide. When the serum is excited with the 320 nm, the fluorescence strength increases greatest for seed coating agent when the pesticide is added, the fluorescence strength increases less for the flapping lice spirit, followed by seed coating agent and least strength for herbicide.

- When join pesticides into the serum, the molecule of serum and pesticides will interact each other and bring fluorescence quenching, so the strength of serum fluorescence will be lower. Since the different serum (normal, high blood glucose, high cholesterol and high blood lipid) has different molecular concentration, so the same pesticides interact with different serum, the degree of fluorescence quenching will be different; the law of the fluorescence spectrum is also different. The fluorescence will be stronger after put pesticides into the serum, perhaps because seed coating agent, herbicide and the flapping lice spirit these three kinds of pesticides all have benzene ring and conjugated double bond, in some condition will be excited and bring fluorescence, lead by the superposition of two fluorescence. That is to say after pesticides join into serum the degree of the fluorescence strength quenching will be smaller than pesticides luminescence.

CONCLUSION

It is known from the experimental results that seed coating agent, herbicide and the flapping lice spirit which contain carbonates pesticide have strongly fluorescent characteristics, which have a greater influence on blood fluorescence. In this study, the 260, 280 and 320 nm excitation lights are used to excite the serum and the fluorescence spectrum is studied for different serum samples added with different pesticides at the same concentration. The fluorescence spectrum changes differently when the same pesticide is added to the serum in different biochemical indexes. At the same time, the fluorescence spectrum is also different when different pesticides are added to the same serum sample. The red shift occurs for the position of the fluorescence peak. The variation of serum in different biochemical indexes is different after the serum is added to pesticides. According to changes of the serum fluorescence strength, the type of pesticides added to the serum can be judged. This experiment indicates that the biochemical indexes of the serum have a great influence on its fluorescence after the serum is added in the pesticides. However its mechanism of action should be further studied. Its deeper research will provide experiment basis for the first-aid and drug use for patients who are suffered pesticide poisoning in different biochemical indexes.

ACKNOWLEDGMENT

This study has been sponsored by natural foundation of Heilongjiang Province (Grant No: F201427) and Doctoral Fund of Ministry of Education of People's Republic of China (Grant No: 20093218110024) and Department of Education of Heilongjiang Province of China (Grant No: 10541155; 12521376) and Bureau of Daqing city science and technology of China (Grant No: SGG2008-041) and Doctor Priming Foundation of Heilongjiang Bayi Agricultural University of China and Farming cultivate bureau science and Technology Bureau (Grant No: HNK11A-06-09) and University Students' innovative entrepreneurship training program of Heilongjiang Province (Grant No: 20141022313).

REFERENCES

Armenta, S., S. Garrigues and M. de la Guardia, 2007. Partial least squares-near infrared determination of pesticides in commercial formulations. Vib. Spectrosc., 44(2): 273-278.

Che, R.S. and S.T. Wang, 2004. Study of fiber fluorescence spectrometer for monitoring carbamate imidacloprid insecticide. J. Optoelectron. Laser, 15(5): 541-544.

Harris, R.D., G.R. Quigley, J.S. Wilkinson, A. Klotz, C. Barzen, A. Brecht and G. Gauglitz, 1998. Waveguide immunofluorescence sensor for water pollution analysis. Proc. SPIE, 3539: 27-35.

Hassoon, S. and I. Schechter, 2000. In situ fluorimetric determination of pesticides on vegetables. Anal. Chim. Acta, 405(1-2): 9-15.

JiJi, R.D., G.A. Cooper and K.S. Booksh, 1999. Excitation-emission matrix fluorescence based determination of carbamate pesticides and polycyclic aromatic hydrocarbons. Anal. Chim. Acta, 397(1-3): 61-72.

Li, W.X., K.X. Xu, Y. Wang, Z.L. Lei and Z.H. Zhang, 2004. Investigation on the detection of pesticide residue in vegetable based on infrared spectroscopy. Spectrosc. Spect. Anal., 24(10): 1202-1204.

Ma, G.X., C.L. Wang, D.W. Fan, D. Xing, L. Qian, J.H. Wang and S.H. Liu, 2006. Quantitative determination of imidacloprid by infrared absorption spectrometry. Spectrosc. Spect. Anal., 26(3): 434-437.

Sun, L., S.H. Zhou, J.X. Yan, D.S. Jiang and J.Z. Li, 2003. Application of laser induced fluorescence technique on detection of pesticide leftover. Laser Infrared, 33(6): 417-418.

Tanojo, H., H.E. Junginger and H.E. Boddé, 2000. Influence of pH on the intensity and stability of the fluorescence of p-aminobenzoic acid in aqueous solutions. Eur. J. Pharm. Sci., 5(1): 31-35.

Wang, T.H., Z.M. Zhao, B.Z. Wei, L. Zhang and L. Ji, 2010. Spectroscopic investigations on the binding of dibazol to bovine serum albumin. J. Mol. Struct., 970(1-3): 128-133.

Wang, Y.T., L.C. Chui, Y.C. Li *et al*., 2006. Using three-dimensional fluorescence research of detecting carbamate lipid pesticide residue. Meas. Technol., 3: 24-28.

Xie, X.Y., X.R. Wang, X.M. Xu, H.J. Sun and X.G. Chen, 2010. Investigation of the interaction between endocrine disruptor bisphenol A and human serum albumin. Chemosphere, 80(9): 1075-1080.

Yang, Q.Y. and Q.Z. Gong, 2006. Study and analysis on rapid determination of pesticides residue. Agric. Mech. Res., 6: 33-36.

Zhang, G.W., J.H. Pan and Y.N. Ni, 2006. Resolution of synchronous fluorimetric spectrum for carbaryl and coumaphos and its application study. Spectrosc. Spect. Anal., 26(6): 1092-1095.