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DETERMINATION OF ORGANOCHLORINE PESTICIDES RESIDUES IN BROKEN RICE FOR PHARMACEUTICAL GLUCOSE BY GC

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Objective. To examine rice organochlorine pesticides in several districts in Hunan so as to provide security reference for production of pharmaceutical glucose which was made with broken rice and a reasonable range for the application of organic pesticides.

Method. A modified method of Chinese Pharmacopoeia was used to extract and prepare the samples which were analyzed by gas chromatography equipped with SE-54 fused silica capillary column (3.0 m × 0.25 mm × 0.32 mm) and electronic capture detector, the nine organic chlorine pesticides were separated by column temperature program and their contents could be measured and calculated by external standard method.

Result. The results showed that the nine organic chlorine pesticides could be accurately determined by the proposed method. The content of organic chlorine conformed to Chinese Government Standards for rice.

Conclusion. The proposed method was so fast, simple and accurate that it could be used to determine organic chlorine pesticide in rice. The rice in these districts can be used for rice glucose production.

Key words: organic chlorine pesticides, residue, rice, gas chromatography.

1. Introduction

Some organochlorine pesticides (HCH and DDT) have been prohibited to produce and use as early as 1983 in China [1], but they are still widely and globally used today, because of their lower market prices coupled with strong effects in the control of pests and diseases. In addition, the structural stability of organic chlorine makes it difficult to degrade, so there are different levels of residues in the soil, animals and plants, causing great harm to the organism itself and future generations [2]. The damage caused by pesticides mainly present in three forms: acute intoxication, chronic injury and mutagenesis, carcinogenesis or teratogenesis [3]. BHC, DDT and other organochlorine pesticides enter the body through food, and mainly accumulate in fat, followed in liver, kidney, spleen and brain, what is worse, they can affect the fetus' health through mothers' milk [4]. Therefore, in order to ensure food safety and protection of human health, it is critical to determinate organochlorine pesticides residues in broken rice.

The method was reference to the Chinese Pharmacopoeia (2010 version) and the relevant literatures, through collecting broken rice from several areas in Hunan province and determining organochlorine pesticides residues by GC, we could know the conditions of organochlorine pesticides residues in Hunan Province. The determination provided security for production of regular pharmaceutical glucose and a reasonable reference for the intelligent use of organic pesticides.

2. Material and Methods

2.1. Reagents

α-BHC(GBW(E)080725), β-BHC(GBW(E)080727), γ-BHC (GBW(E)0807229), δ-BHC (GBW(E)080731), PP'-DDE (GBW(E)080735), OP'-DDT (GBW(E)080739), PP'-DDD (GBW(E)080733), PP'-DDT (GBW(E)080737), PCNB(GBW (E)060617). All of these were purchased from National Research Center for Certified Reference Materials.

2.2. Apparatus

Gas chromatography (Shimadzu GC-2010) was equipped with a SE-54 fused silica capillary column (3.0 m × 0.25 mm × × 0.32 mm), and an electron capture detector. Universal high-speed micro-mill was purchased from Tianjin Taisite Instrument Co., Ltd., KQ-250B ultrasonic cleaner was a product of Kunshan Ultrasonic Instrument Co., Ltd., RE-5250 Vacuum rotary evaporator was from Shenzhen Sanli Chemical Co., Ltd., TDC-40B Centrifuge , N-EVAPTM112 Nitrogen analyzer were made in USA.

2.3. Sample Collection

The broken rice was taken from six areas (Changde, Changsha, Yueyang, Zhuzhou, Chenzhou, Yiyang) in Hunan using random sampling method, nine samples for each region.

2.4. Method

2.4.1. Preparation of Standard Solution

It was weighed BHC(α-BHC, β-BHC, γ-BHC, δ-BHC), DDT (DDT) (PP'-DDE, PP'-DDD, OP'-DDT, PP'-DDT) and pentachloronitrobenzene (PCNB) pesticides standard solution respectively, then diluted to each milliliter containing 4–5 μg pesticides with petroleum ether (60–90°C). It was taken precisely amount of reference stock solution 0.5 mL into a 10 mL volumetric flask, petroleum ether (69–90°C) diluted to the mark, shook. It was taken precisely amount of reference mixed stock solution and diluted petroleum ether (69–90°C) to each liter contains 0 μg, 1 μg, 5 μg, 10 μg, 50 μg, 100 μg, 250 μg pesticides respectively.

2.4.2. Preparation of Sample Solution

It was taken 2.0000 g broken rice powder extracted by water (20 mL) for 30 min ultrasonic extraction, added acetone 40 mL and weighed, ultrasonic treatment 30 min, let it cool, used acetone to supply losing weight after weighing, add 6 g sodium chloride and 30 mL dichloromethane, weighed, then 15 min ultrasonic treatment, used dichloromethane to supply losing weight after weighing again, put it aside (stratification), moved the organic phase into a 100 mL flask with anhydrous sodium sulfate rapidly, placed 4 hours. It was taken 35 mL extracted solution and dried in the water bath at 40°C on a rotary evaporator, concentrated to nearly dry, addded a small amount of petroleum ether (60–90°C), repeated at least three times until dichloromethane and acetone were eliminated, dissolved with petroleum ether (60–90°C) in a 10 mL centrifuge tube and diluted to 5 mL, added sulfuric acid 1 mL carefully, shake 1 min and centrifuged (3000 r/min) 10 minutes. It was taken 2 mL supernatant into a test tube, concentrated with Nitrogen analyzer and diluted to 1 mL.

2.4.3. Chromatographic Conditions

Operating conditions were as follows: initial temperature 120°C (6 min), increased at a rate of 5°C min^{-1} to 220°C, held for 10 min, then increased at a rate of 8°C min^{-1} to 250°C and finally held at 250°C for 15 min; injector temperature: 260°C ; carrier gas: N_2(99.999%); column flow-rate: 1 mL/min; detector temperature: 300°C injection volume: 1 uL; External standard quantitative method.

3. Results and Discussion

3.1. System Suitability

The mixed standard stock solution was injected into chromatograph under the chromatographic conditions mentioned above. The results showed that, adjacent peaks' efficiency of separation was greater than 1.5 [5], number of theoretical plates was higher than 1×10^6 calculated ac-

cording to α-BHC, and made sure the retention time (RT) of nine organochlorine pesticides (Table 1).

3.2. Reproducibility

Took 2.0 g broken rice powders from Changde for 9 groups and treated the powder by the same method according to sample preparation. Then determined each sample solution continuously according to the above chromatograph condition took the peak area into the regression equation and then calculated the pesticides content. The results show that, PCNB was detected only and RSD value was 0.97% the best is less than 1% [6], the method repeatability was good.

3.3. Stability

Took the sample solution (Yueyang) and inject to the chromatograph for four times during a single day (0 h, 2 h, 4 h, and 6 h) according to the above chromatographic condition. The α-BHC, β-BHC, γ-BHC, δ-BHC, PP'-DDE, PP'-DDD were detected and RSD value were 0.8%, 0.5%, 1.1%, 0.3%, 0.6%, 0.8% respectively (less than 5%), indicating that the preparation of the sample was stable within 6 hours.

3.4. Limit of Detection (LOD)

The mixed standard stock solution were injected into chromatograph respectively under the chromatographic conditions mentioned above, then calculated the instrument limit of detection (IDL) (three times the value of the instrument background signal) and the method limit of detection (MDL). The results showed that the IDL of nine organochlorine pesticides were all lower than 1 μg/L, indicating that the sensitivity was good (Table 2).

3.5. Linearity

1 μL of the mixed standard solution was injected into the chromatograph respectively, the peak area as the vertical axis and the concentration as the abscissa (μg/L), calculated the regression equation (Table 3). The results showed that all tested components from 0 μg/L to 250 μg/L appeared a good linear relationship between the peak area and concentration.

Table 1

The Retention Time of 9 Organochlorine Pesticides

Species	t(RT)/min
α-BHC	15.56
β-BHC	16.75
γ-BHC	17.02
PCNB	17.24
δ-BHC	18.07
PP'-DDE	25.77
PP'-DDD	27.72
OP'-DDT	27.91
PP'-DDT	29.91

Table 2

The Device Limit Determination and Method Limit Determination of 9 Organochlorine Pesticides

Species	IDL, ng/L	MDL, mg/kg
α-BHC	100	0.00025
β-BHC	128	0.00032
γ-BHC	180	0.00045
PCNB	140	0.00035
δ-BHC	200	0.0005
PP'-DDE	368	0.00092
PP'-DDD	298	0.00075
OP'-DDT	275	0.00069
PP'-DDT	336	0.00084

Table 3

Regression Equation of 9 Organochlorine Pesticides

Standard	T (RT)	Regression equation	R^2
α-BHC	15.56	y = 2228.1x – 8818.3	0.9987
β-BHC	16.75	y = 1299.6x + 4282.3	0.9988
γ-BHC	17.02	y = 2026.6x – 5099	0.9993
PCNB	17.24	y = 2154.1x + 7490	0.9993
δ-BHC	18.07	y = 2256.3x – 9267.9	0.9989
PP'-DDE	25.77	y = 1616.3x – 5766.3	0.9985
PP'-DDD	27.72	y = 1108.3x – 268.92	0.9969
OP'-DDT	27.91	y = 959.9x – 1899.3	0.9996

3.6. Recovery

Took the broken rice powder from Changde into 9 portions, each 2.0 g, precisely weighed, and divided them into three groups. Add 0.1 ml, 0.5 ml, 0.9 ml 250 μg/L mixed standard solutions to each group respectively, and then air. Prepare the test sample solution according to the method of 2.3. Each portion determines 3 times, and calculated the recovery; the results can be seen in Table 4.

From the table above we can learn that under different amount of pesticides, the recovery and RSD of 9 types of organochlorine pesticides meet the requirements of pesticide residues detection.

3.7. Determination of Samples

To weigh accurately broken rice powder from different areas, and prepared the test sample solution according to the method of 2.3, then injected them into the gas chromatograph with the above chromatographic condi-

Table 4

Results of Recovery

Species	The amount of adding		
	25 ng	125 ng RSD/%	225 ng
α-BHC	1.28	0.97	0.65
β-BHC	1.92	0.38	0.87
γ-BHC	1.05	0.97	2.32
PCNB	1.21	0.52	0.30
δ-BHC	0.40	0.85	1.06
PP'-DDE	1.47	1.40	0.59
PP'-DDD	1.26	1.57	0.95
OP'-DDT	1.02	1.63	1.27
PP'-DDT	1.02	1.14	1.44

Table 5

The Organochlorine Determination Results in the Rice Sample (w/mg·Kg^{-1})

	α-BHC	β-BHC	γ-BHC	PCNB	δ-BHC	PP'-DDE	PP'-DDD	OP'-DDT	PP'-DDT
Changde	—	—	—	0.1147	—	—	—	—	—
Yueyang	0.0116	0.0256	0.012	—	0.0272	0.0147	0.0387	—	—
Zhuzhou	—	—	—	—	0.013	—	—	—	—
Yiyang	—	0.002	0.008	—	0.01	0	0.013	—	—
Changsha	—	—	—	—	—	—	0.006	—	—
Chenzhou	—	—	—	—	—	—	0.007	—	—

tions. At last, calculated the content of pesticide residues by peak area through external standard method, the results can be seen in table 5.

According to the Chinese national standard of pesticide residues: the maximum limits of DDT, BHC and PCNB was 0.2 mg/Kg, 0.3 mg/Kg and 0.1 mg/Kg in agricultural products [7]. From the above table, it can be known that only the sample of Changde exceeds the limits, the content of PCNB was 0.1147 mg/Kg slightly greater than the limit of 0.1 mg/Kg. The result can be seen in fig. 1.

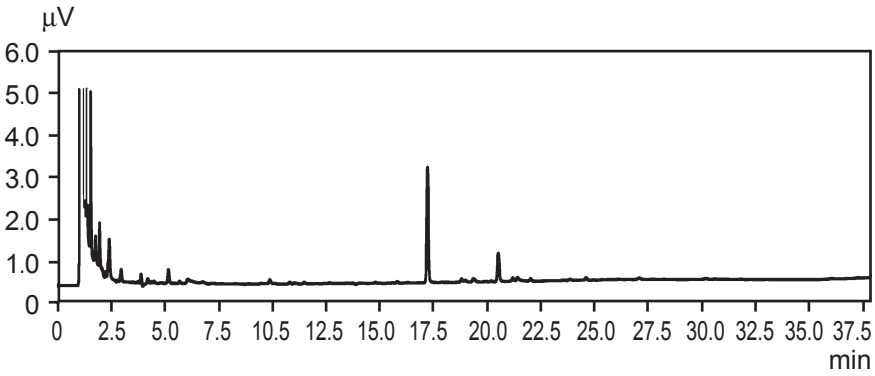

Fig. 1. GC chromatogram of Changde's rice

4. Discussion

It can be seen from the methodology that, firstly, the extraction method we used was modified according to the Pharmacopoeia, for our method was more simple and efficient, time-saving and consuming less reagents than the Pharmacopoeia. Compared to the pre-treatment technology developed in recent years, such as solid-phase extraction, solid phase micro-extraction, supercritical fluid extraction, microwave-assisted extraction, accelerated solvent extraction, gel permeation extraction and so on [8–11], our method was simple and low cost. Secondly, the retention time of nine kinds of organic chlorine, through gas chromatography we established, was shorter than Zeming Guan's [12] literature reported by nearly half. So it can not only saved a lot of time, but also reduced the loss of the instruments. Finally, after the methodological verification, it can be seen that this method had high recovery, good reproducibility, high sensitivity, and can be used to test large quantities of samples. It provides a reliable the method to detection the residues of organochlorine pesticide.

From the results we can find that there were more types of organochlorine pesticide residues in Yiyang and Yueyang, mainly BHC and DDT, but not exceeded. So we can learn BHC and DDT were over used in these two places, and strict controls should be implemented on the amount, otherwise it will have serious consequences beyond the safe limits. Although the rice sample of Changde had only one type of residues-PCNB, it exceeds the safe range. So the pesticides that farmers used were relatively simple, but the amount was large, or abusive. The results of Zhuzhou, Chenzhou and Changsha was good, only have one kind of trace pesticide residues. And the pesticide residues of broken rice in Yiyang have many types but the amount was small. In summary, different places use different pesticides, but generally speaking BHC and DDT were still used frequently. So the government should strengthen restrictions on the two types of pesticides to ensure food security.

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ONE-DAY SURGERY (SHORT TERM DEPARTMENT) IN THE TREATMENT OF PATIENTS SUFFERING FROM CHOLELITHIASIS

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ХИРУРГИЯ ОДНОГО ДНЯ (СТАЦИОНАР КОРОТКОГО ПРЕБЫВАНИЯ) В ЛЕЧЕНИИ БОЛЬНЫХ ЖЕЛЧНОКАМЕННОЙ БОЛЕЗНЬЮ

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В 2006–2011 гг. в нашей клинике выполнено 4533 лапароскопических холецистэктомий. 557 (12,3 %) пациентов находились в стационаре короткого пребывания. Совместно с анестезиологом проводился тщательный отбор пациентов. Выписывали больных при полной уверенности в благополучном течении послеоперационного периода — через 24 ч после поступления с последующим медицинским «сопровождением» в амбулаторных условиях. Из запланированных в стационаре короткого пребывания 678 больных операция и лечение в предполагаемые сроки состоялись у 557. Пациенты наблюдались оперирующим хирургом на протяжении 4–7 дней. При тщательном отборе больных с хроническим калькулезным холециститом возможно их успешное лечение в стационаре короткого пребывания (до 20 %).

Ключевые слова: желчнокаменная болезнь, хронический калькулезный холецистит, стационар короткого пребывания, хирургия одного дня.

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Introduction. Laparoscopic cholecystectomy was recognized as operation of choice in treatment of cholelithiasis. The terms of hospital treatment were maximum decreased with a widespread introduction of laparoscopic technologies in treatment of cholelithiasis. In USA, Japan, European countries the hospitalization after elective laparoscopic cholecystectomy may be no more than 24 hours. From 15 to 30% of laparoscopic cholecystectomies are carried out by “one day surgery” principles.

Materials and methods. In our hospital within the period from 2006 to 2011 there were carried out 4,533 laparoscopic cholecystectomies. There were operated 2,800 (61.8%) patients suffering from chronic cholelithiasis. 557 (12.3%) patients suffering from cholelithiasis were discharged from the hospital in 24 hours.

Results and discussion. There were planned 678 patients with cholelithiasis to be operated by “one day surgery”, but operation and treatment within this term took place in 557 patients. With raising experience the number of patients has grown from 39 (9,9%) in 2006 to 156 (19.5%) in 2011. There were no severe complications. Umbilicitis was formed in 14 patients in periomphalic area, which didn’t require hospital treatment. From 2007 to 2010 in our hospital 102 (2.3%) patients, which were hospitalized with acute cholecystitis, were treated in a short term department.