Determination of 11 kinds of ultraviolet absorbents in plastic food contact materials by high performance liquid chromatography

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Abstract: A method for the determination of 11 kinds of ultraviolet (UV) absorbents in food contact materials was established based on high performance liquid chromatography. The target compounds were separated on an Agilent SB C18 (250 mm×4.6 mm×5 μm) column with gradient elution using acetonitrile and water as mobile phase, which analyzed by diode array detector and quantified by external standard method. The results showed that the linear range of the 11 UV absorbents was between 0.2 and 50.0 mg/L (R²≥0.9985), the limits of detection (LODs) were in the range of 0.01-0.05 mg/L, and the limits of quantification (LOQs) were 0.05-0.20 mg/L. The recoveries (R) of blank samples at 0.2, 10 and 50 mg/kg were from 80.2% to 97.5%, and the relative standard deviations (RSD) were ranged from 0.37% to 4.15%. The method had low detection limit, high stability and good repeatability, which could be used for daily detection of UV absorbents in food contact materials.

1. Introduction
UV absorbents can absorb ultraviolet light in the range of 280-400 nm and release light energy in the form of heat energy to reduce the photo degradation and aging of plastic products and extend the service life, so they are widely used in the manufacturing process of food contact materials [1,2]. Studies have shown that UV absorbents have certain toxicity, which can cause dermatitis and skin allergy. Moreover, UV absorbents have bio-accumulation, which will affect the length and quality of organisms and result in imbalance of thyroid system and fatty acid metabolism imbalance [3,4].

Currently, GB 9685-2016 standard [5] stipulates the safe usage and migration amount of some UV absorbents, but there is no corresponding standard method to detect UV absorbents, which will not guarantee the quality of food contact materials and seriously threaten food safety and human health. Therefore, it is urgent to establish a method for the detection of UV absorbent in food contact materials.

In this paper, a method for the determination of 11 common UV absorbents in food contact materials was established based on high performance liquid chromatography. The method has low detection limit, high stability and good repeatability, which can be used for daily detection of UV absorbents in food contact materials.
2. Materials and Methods

2.1 Materials and reagents. 11 UV absorbents were all supplied from Dr. Ehrenstorfer (Augsburg, Germany). HPLC grade acetonitrile was purchased from Merck (Darmstadt, Germany). The water used in the experiment was ultra-pure water.

2.2 Instruments and equipment. Agilent 1260 HPLC with DAD detector; MS105DU electronic analysis balance (Mettler Toledo International TradeCo., LTD.); N-evap-112 Nitrogen Blower (Organomation, USA); KH3200DE Ultrasonic cleaner (Kunshan Hechuang Ultrasonic Instrument Co., LTD.)

2.3 Preparation of standard solution. Each standard substance of 10 mg was accurately weighed (accurate to 0.1 mg) and placed in a 10 mL volumetric flask, and then dissolved with mixed organic solvent (methanol: acetone: n-hexane =2:1:1) to prepared into UV absorbent single standard reserve liquid of 1000 mg/L. An appropriate amount of 11 kinds of UV absorbent single standard reserve solution was removed and diluted with mixed solvent into standard working solutions with concentrations of 0.2 mg/L, 1.0 mg/L, 10.0 mg/L, 25.0 mg/L and 50.0 mg/L.

2.4 Sample pretreatment. The sample was cut into small pieces with an area no larger than 5 mm×5 mm. The 1.0 g sample was accurately weighed into a 25 mL colorimetric tube, and mixed solvent of 10 mL was added for ultrasonic extraction for 40 min. The sample was cooled to room temperature, and the supernatant was filtered through 0.45 μm organic system filter membrane for UPLC analysis.

2.5 Chromatographic condition. The separation was performed on an Agilent SB C18 column (250 mm ×4.6 mm, 5 μm) with water as mobile phase A and acetonitrile as mobile phase B. Column temperature was 30 ℃, sample volume was 10 μL, the detection wavelength was 300 nm, and the flow rate was 1.0 mL/min. The gradient elution procedure was shown in Table 1.

| Time (min) | A (%) | B (%) |
|-----------|-------|-------|
| 0         | 15    | 85    |
| 3.0       | 15    | 85    |
| 12.0      | 75    | 25    |
| 17.0      | 100   | 0     |
| 30        | 100   | 0     |
| 31        | 15    | 85    |
| 35        | 15    | 85    |

3. Results and Discussions

3.1 Optimization of chromatographic conditions
The effects of chromatographic column, mobile phase, column temperature, elution gradient and wavelength on the separation of UV absorbent were investigated. The results showed that Agilent SB C18 column had high response strength, good chromatographic peak shape and complete baseline separation. Through the comparison of acetonitrile-water, methanol-water and ethyl acetate-water, it was found that acetonitrile-water obtained better separation effect. In addition, the experiment also optimized the column temperature and other influencing factors, finally Agilent SB C18 column in acetonitrile-water system was selected for the separation of UV absorbent. The best column temperature was set at 30 ℃, and absorption wavelength was 300 nm with flow rate of 1.0 mL/min. Under the optimal conditions, 11 UV absorbents could achieve good baseline separation within 30 min, as shown in Figure 1.
3.2 Standard curve and detection limit
The mixed standard solution series were determined under the optimal chromatographic conditions, and the mass concentration of each compound was linearly regression based on the peak area. The results showed that the 11 target compounds showed good linear relationship in the range of 0.2-50 mg/L, and the linear correlation coefficients $R^2$ were all greater than 0.9985. The method limit of detection (LOD) and method limit of quantification (LOQ) of 11 UV absorbents were calculated using 3 times signal-to-noise ratio ($S/N \geq 3$) and 10 times signal-to-noise ratio ($S/N \geq 10$), and the results were shown in Table 2. This indicates that the method can be used for the qualitative and quantitative analysis of 11 kinds of UV absorbents in plastic food contact materials.

| No. | Compound | linear regression equation | $R^2$ | LOD (mg/L) | LOQ (mg/L) |
|-----|----------|----------------------------|-------|------------|------------|
| 1   | UV-2     | $Y=5.2804X-2.5916$         | 0.9994| 0.02       | 0.10       |
| 2   | UV-24    | $Y=10.8880X-5.9158$        | 0.9993| 0.01       | 0.05       |
| 3   | UV-71    | $Y=15.0832X-6.9006$        | 0.9994| 0.01       | 0.05       |
| 4   | TBS      | $Y=6.1525X-3.5478$         | 0.9988| 0.02       | 0.10       |
| 5   | UV-3030  | $Y=15.8745X-8.4015$        | 0.9993| 0.01       | 0.05       |
| 6   | UV-1579  | $Y=19.9449X-9.3209$        | 0.9995| 0.01       | 0.05       |
| 7   | UV-329   | $Y=11.1812X-6.7698$        | 0.9991| 0.02       | 0.10       |
| 8   | UV-234   | $Y=8.3885X-5.3021$         | 0.9990| 0.02       | 0.10       |
| 9   | UV-326   | $Y=4.3749X-2.8150$         | 0.9989| 0.05       | 0.20       |
| 10  | UV-327   | $Y=4.2273X-2.0482$         | 0.9993| 0.05       | 0.20       |
| 11  | UV-1577  | $Y=14.8150X-7.0527$        | 0.9994| 0.02       | 0.10       |
3.3 Precision and recovery
The standard solution with different concentrations was added to the negative sample, and the recovery and precision were tested. The standard addition levels were 0.2, 10.0 and 50.0 mg/L, and each concentration level was tested 6 times in parallel. The experimental results (Table 3) showed that the recoveries (R) of 11 UV absorbents at high, medium and low spiked levels were 80.2%-97.5%, and the relative standard deviations (RSD) were 0.37%-4.15%, which could meet the actual detection requirements.

Table 3 Recoveries (R) and relative standard deviations (RSD) (n=6) for 11 UV absorbents

| No. | Compound | Spiked levels (mg/kg) | R (%) | RSD (%) |
|-----|----------|-----------------------|-------|---------|
| 1   | UV-2     | 0.2                   | 83.6  | 1.45    |
|     |          | 10.0                  | 88.6  | 0.80    |
|     |          | 50.0                  | 95.9  | 0.50    |
|     |          | 0.2                   | 83.3  | 1.53    |
| 2   | UV-24    | 10.0                  | 88.3  | 1.05    |
|     |          | 50.0                  | 95.6  | 0.63    |
|     |          | 0.2                   | 82.5  | 4.15    |
| 3   | UV-71    | 10.0                  | 89.9  | 1.95    |
|     |          | 50.0                  | 96.9  | 0.80    |
|     |          | 0.2                   | 80.2  | 3.18    |
| 4   | TBS      | 10.0                  | 81.6  | 1.61    |
|     |          | 50.0                  | 90.4  | 0.43    |
|     |          | 0.2                   | 82.9  | 2.51    |
| 5   | UV-3030  | 10.0                  | 87.9  | 0.86    |
|     |          | 50.0                  | 95.2  | 0.44    |
|     |          | 0.2                   | 80.4  | 1.43    |
| 6   | UV-1579  | 10.0                  | 90.4  | 1.00    |
|     |          | 50.0                  | 96.6  | 0.59    |
|     |          | 0.2                   | 83.2  | 1.25    |
| 7   | UV-329   | 10.0                  | 87.2  | 1.63    |
|     |          | 50.0                  | 96.0  | 0.51    |
|     |          | 0.2                   | 81.5  | 1.46    |
| 8   | UV-234   | 10.0                  | 88.4  | 2.23    |
|     |          | 50.0                  | 97.5  | 0.60    |
|     |          | 0.2                   | 80.6  | 1.00    |
| 9   | UV-326   | 10.0                  | 88.1  | 2.13    |
|     |          | 50.0                  | 97.0  | 0.65    |
|     |          | 0.2                   | 81.7  | 1.69    |
| 10  | UV-327   | 10.0                  | 89.3  | 0.93    |
|     |          | 50.0                  | 96.4  | 0.63    |
|     |          | 0.2                   | 81.3  | 0.37    |
| 11  | UV-1577  | 10.0                  | 89.1  | 0.81    |
|     |          | 50.0                  | 96.4  | 0.57    |

3.4 Actual sample testing
The method was applied to the determination of 10 kinds of plastic packaging materials, and the results showed that three kinds of them detected a certain amount of UV absorbent residues. UV-326 residue was found in Sample 1, UV-329 and TBS were detected in sample 2, and UV-24 was detected in Sample 3. The content was ranged from 0.28 to 2.14 mg/kg.

4. Conclusions
A HPLC method was established for the determination of 11 kinds of UV absorbents in plastic food contact materials through the investigation and optimization of chromatographic conditions. The
method could effectively separate 11 kinds of UV absorbents within 30 min with the limits of detection (LODs) of 0.01-0.05 mg/L and quantification (LOQs) of 0.05-0.20 mg/L. The method had the advantages of simple operation, high sensitivity, high separation and high accuracy, which could provide technical reference for the analysis and detection of UV absorbent in plastic food contact materials in the future.

Acknowledgments
This research was funded by Scientific Research Project of Chongqing Market Supervision Administration [CQSJKJ2019005].

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