Analysis on Influencing Factors of QuEChERS Pretreatment Method for Pesticide Residues

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Abstract Pesticide residues are related to environmental pollution, food safety and other issues, and sample processing is a key link in the whole analysis and detection process. Therefore, it is of far-reaching significance to establish a reliable and safe pretreatment technique for pesticide detection. QuEChERS (quick, easy, cheap, effective, rugged and safe), as a pretreatment method for pesticide residue analysis, is widely used in the field of testing and analysis because of its characteristics of rapidity, simplicity, low cost and high efficiency. In this paper, the effects of material selection involving extractants, analyte protectants and purification agents and their ratio optimization in QuEChERS method on the improvement of pretreatment efficiency were analyzed, hoping to provide further technical reference for expanding the pretreatment technique.

Key words Pesticide residue; QuEChERS; Influencing factor
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Pesticides are defined as a class or a variety of chemical substances that can be applied to prevent, control, and eliminate agricultural and forestry pests, diseases, and weeds. They play a significant role in helping to increase agricultural yields and improve crop quality. However, excessive use of pesticides will inevitably lead to problems such as pesticide residues, and 70% - 80% of pesticide substances will remain in the environment during unreasonable use, resulting in environmental pollution, food safety and other adverse events[1]. Combined with the national sampling data, pesticides with excessive residues in vegetables are mainly insecticides, and pyrethroids, nicotinoid imidacloprid and other pesticides have the characteristics of high efficiency and broad spectrum, and their abuse intensifies the detection risk of excessive residues. Some pesticide residue detection methods have complex pretreatment operations, long detection time and low accuracy, and are not conducive to batch, multi-parameter and high-precision detection. Therefore, the problem of pesticide residues is still serious, and it is of great practical significance to establish accurate and reliable analysis and detection techniques[2-3].

QuEChERS is widely used in the field of sample processing as a simple, efficient and environmentally friendly pretreatment technique[4]. In this study, the influence factors of QuEChERS treatment technique were analyzed, and the effects of material selection involving extractants, analyte protectants and purification agents and their ratio optimization in QuEChERS method on the improvement of pretreatment efficiency were analyzed and discussed, aiming to provide further technical reference for improving this detection technique.

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QuEChERS Technique

QuEChERS is based on the combination and derivation of solid-phase extraction and matrix solid-phase dispersion. Under the action of efficient extraction and purification reagents, analytes are separated from sample matrix interferents by vortex centrifugal force[5]. As a relatively advanced pretreatment technique, typical QuEChERS method uses sodium chloride to eliminate matrix interference at the initial stage of application, and extractants are used in a lower proportion. Subsequently, 1% acetic acid-acetonitrile solution and sodium acetate buffer solution are increased to strengthen the extraction efficiency of pH-sensitive compounds. Finally, it evolves into sodium chloride to reduce interference, and buffer reagents are used in a certain proportion to protect alkali-sensitive target analytes. Taking GB 23200.113-2018 *National food safety standard: Determination of* 208 *pesticides and metabolites residues in foods of plant origin* as an example, different emphases of QuEChERS pretreatment under different matrix conditions were elaborated in detail.

Analysis of Influencing Factors in QuEChERS Pretreatment Process

Effects of extractants on extraction effect

In the process of pesticide residue treatment, acetonitrile,

ethyl acetate, acetone and other single extractants or mixed extractants with a certain proportion of acetic acid, formic acid, hydrochloric acid and so on are mainly used. There are differences in the selection of single or mixed solvents, mainly due to the polarity differences of the above substances, among which methanol and acetic acid are the representatives of solvents with higher polarity. In addition, acetone is miscible with water and requires the addition of nonpolar solvents to achieve water layer separation. On the contrary, when ethyl acetate is mixed with water, there is no need to add nonpolar substances to separate it from water, but there are difficulties in separating samples with higher polarity. Therefore, acetonitrile is often used as the extractant in the extraction process, and the obtained solution has relatively less interference, and the organic solvent and the water layer are obviously layered. The most commonly used extraction solvents for pesticide residue analysis by QuEChERS method are acetonitrile, ethyl acetate, acetone or mixtures of several solvents. Deng *et al.* [6] compared and analyzed the extraction efficiency of 21 organochlorine pesticides in fruits and vegetables with single solvents including acetonitrile, ethyl acetate and acetone and a mixed solvent in equal proportion by gas chromatography/mass spectrometry.

Effects of analyte protectants on extraction efficiency

Analyte protectants are single compounds or simple mixtures that imitate the protective effect of matrixes. They can compete for the active sites in the liner, improve the response value of pesticides in pure solvents, and make them reach the same response as pesticides in matrixes, so as to realize multi-residue and high-throughput detection. The analyte protectants mainly have following characteristics: ① they will not react with the analytes in solutions and will not induce degradation. ② They will not affect the performance of chromatographic columns or detectors. ③ They will not interfere with detection. ④ They will not deposited in analytical instruments; ⑤ They can be widely used, and are low in cost and harmless. And ⑥ they are soluble in solvents. Polyethylene glycol, D-sorbitol, 3-ethoxy-1, 2-propanediol, erythritol, triglycerol and so on are commonly used as analyte protectants in QuEChERS pretreatment. These substances are mainly polyhydroxy substances and aliphatic compounds, or have amphoteric characteristics of amino-carboxyl or amino-hydroxyl functions, and the combined analyte protectants are more conducive to meeting the needs of analysis[7].

Effects of dehydrating agents on extraction effect

Dehydrating agents produce salting-out effect in the extraction process of target analytes to promote separation. Dehydrating agents commonly used in QuEChERS method include magnesium sulfate, sodium sulfate and sodium chloride. Among them, the precipitation effect of magnesium sulfate in acetonitrile is the best, and the combination of magnesium sulfate and sodium chloride is the most widely used in QuEChERS pretreatment. Liu[8] analyzed the effects of different dehydrating agents on the recovery rate in the research of QuEChERS extraction. The results showed that anhydrous magnesium sulfate was the best water removal agent, and the experimental recovery rate was between 80% and 110%. Anhydrous sodium sulfate has an average water removal effect, and there is almost no water removal effect when sodium chloride is used alone. However, because sodium chloride has a strong salting-out effect on protein, adding a small amount of sodium chloride before homogeneous extraction can play a better role in protein removal.

Promoting effect of purifying agents on recovery effect

The purifying agents used by QuEChERS have a positive effect on the purification and recovery of pesticide residues. Proper selection and proportion of purifying agents can improve the analytical performance of the whole method, including reducing matrix effect, improving sensitivity and increasing the recovery rate of target analytes in complex matrixes. Purifying agents commonly used in QuEChERS method include PSA, C18 and GCB. PSA has polar function and weak anion exchange function, which can remove polar compounds from nonpolar mixed acetonitrile solutions. C18 is a nonpolar adsorbent, which is mainly used to adsorb long-chain aliphatic compounds, sterols and other nonpolar interfering substances from polar samples, but it will reduce the recovery effect of pesticides such as penconazole. GCB is mostly used to remove pigments such as chlorophyll and carotenoids, steroids and polyphenols with planar structures[9]. Dong *et al.* [10] also added C18, GCB, aminopropyl and other adsorbents, which strengthened the removal ability of pigments and vitamins and improved the purification effect, in addition to PSA in the purification process of QuEChERS method. Shao *et al.* [11] also studied the effects of GCB dosage, adding time and other factors on the recovery rate, and greatly reduced the detection limit of samples on the basis of obtaining a good recovery rate.

Conclusions

Sample pretreatment is an important step in the detection and analysis of pesticide residues. The QuEChERS method has become a mainstream technique because of its simple operation, low cost, high efficiency, rapidity and accuracy, and is widely used in high-throughout detection and analysis. Analysis on the influencing factors of QuEChERS pretreatment process provides technical reference for further improving the detection techniques.

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(Continued on page 68)

Analysis of actual samples

Three kinds of milk tea samples of different brands were randomly purchased, and the PFCs were detected by the established mature detection method. The results showed that the 18 perfluorinated and polyfluorinated compounds were all not detected.

Conclusions and Discussion

In this study, an LC-MS/MS method was established for the simultaneous analysis of 18 PFCs in milk. The samples were first subjected to precipitation of proteins by potassium ferrocyanide solution and zinc acetate solution, and then extracted by acetonitrile for the detection by LC-MS/MS. The detection limits of the method were in the range of 0.001 – 0.05 μg/kg, and the quantitation limits were in the range of 0.03 – 0.20 ng/L. The recovery values ranged from 72.8% to 110.5%. The results showed that the method has high sensitivity and accuracy. This study provides a rapid, accurate and reliable analysis method for the determination of PFCs in milk tea, which has strong practical value.

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(Continued from page 63)

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