SUMMARY AND RECOMMENDATIONS

Summary

Presence of pesticide residues in food is one of the most important food safety hazards, which are known to cause adverse health effect in consumers. As a result, the amount of pesticide residues in food are strictly regulated by many countries across the world by issuing maximum residue limits (MRLs). In India, the MRLs for pesticides in various foods are fixed by the Food Safety and Standards Authority of India (FSSAI), taking into account various factors, including legality of usage of the pesticides based on good agricultural practices (GAP), toxicological evaluation of the pesticides and consumption pattern of the food in which the MRL is fixed. Typically, the MRLs are in the ranges of low parts per millions (mg kg^{-1}). To evaluate compliance of a particular food with respect to pesticide residues, the food has to be analysed with a sensitivity that enables direct comparison with the MRLs.

Spices, which are used extensively in India for culinary preparations, are prone to incidence of pesticide residues and the associated health hazards. Spices are generally considered difficult matrices to analyse owing to their complex chemical nature. Besides being low moisture commodities, spices contain active compounds that impart colour, flavour and aroma to foods, and these compounds can pose interferences in high sensitivity analysis of pesticide residues in these matrices. Since spices belong to different classes like dried roots, fruits and berries, seeds, bark, floral parts etc., applying a single analytical method for pesticide residues to different classes of spices is not practical.

In the present work, an effective and structured analytical framework for analysis of residues of 78 pesticides commonly used for cultivation of spices in India was developed and validated for six spices belonging to different classes, *viz.* chillies,

cardamom, cumin, ginger, cinnamon and curry leaves. The methods employed high sensitivity chromatography and tandem mass spectrometry techniques, *viz.* UPLC-MS/MS and GC-MS/MS. The issue of matrix effects observed in mass spectrometric analysis of pesticide residues in spices, which introduce qualitative and quantitative errors, were addressed in this work. Two novel strategies to mitigate these matrix effects, *viz.* the use of analyte protectants in GC-MS/MS, and the use of active components in spices as surrogate matrix compounds in solvent-based reference standards in UPLC-MS/MS, were successfully implemented.

Dithiocarbamates (DTC) are a class of broad-spectrum fungicides extensively employed in the cultivation of spices. A sensitive analytical method in which DTC residues were quantitatively converted to carbon disulphide, absorbed into isooctane and detected in GC-MS using selected ion monitoring technique was successfully validated in two spices, cardamom and black pepper.

The developed residue analysis methods using UPLC-MS/MS and GC-MS/MS were successfully applied to real spice samples collected from retail markets for performing compliance evaluation of these samples with the national MRL regulations in India. Characterization of food safety hazards associated with presence of pesticide residues in spices, based on the results of these analyses, were also performed.

Recommendations

Spices typically contain active chemical compounds which contribute to their special properties and which are present in relatively high concentrations. Many of these active compounds, or chemical analogues of such compounds, have been synthesised are readily available as reference standards. The novel strategy developed in the present work for using synthetic capsaicin as surrogate matrix compound in solvent-based standards to mitigate matrix effects in chillies can be extended to many other spices (e.g., curcumin for

analysis of residues in turmeric, piperine for analysis of residues in black pepper, etc.). The analytical method for DTC compounds validated in cardamom and black pepper also affords possibility for extending to other classes of spices. In analysis of samples from retail markets, the residues detected were mostly organophosphates, pyrethroids and carbamates. This showed that the adoption of new generation pesticides in India for cultivation of spices is still not widespread. This is an avenue of improvement which can potentially result in considerably lowering food safety hazards in Indian spices.