High Performance Liquid Chromatographic Method for Determination of Metazosulfuron Residue in Representative Crops

Hyeri Lee,1,2 Eunhye Kim,1 Young Deuk Lee,3 Jeong-Han Kim1*

1Department of Agricultural Biotechnology, Seoul National University, Seoul 151-741, Republic of Korea, 2Environmental Measurement and Analysis Center, National Institute of Environmental Research, Incheon 404-708, Republic of Korea, 3Division of Life and Environmental Science, Daegu University, Gyeongsan 712-714, Republic of Korea

Received: 19 May 2012 / Revised: 1 June 2013 / Accepted: 21 June 2013
ⓒ 2013 The Korean Society of Environmental Agriculture
This is an Open-Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/3.0) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

Abstract

BACKGROUND: This study was performed to develop a single residue analytical method for new herbicide metazosulfuron in crops.

METHODS AND RESULTS: Brown rice, apple, mandarin, Kimchi cabbage and soybean were selected as representative crops, and clean-up system, partition solvent and extraction solvent were optimized. Instrumental limit of quantitation (ILOQ), linearity of calibration curve and method limit of quantitation (MLOQ) were determined based on the chromatography and whole procedures. For recovery tests, brown rice, apple, mandarin, Kimchi cabbage and soybean samples were macerated and fortified with metazosulfuron standard solution at three levels (MLOQ, 10 MLOQ and 100 MLOQ). And then those were extracted with acetonitrile, concentrated, and partitioned with ethyl acetate. Then the extracts were concentrated again and cleaned-up through NH2 (aminopropyl) SPE cartridge with acetone : dichloromethane (1% acetic acid) (20 : 80, v/v) before concentration and analysis with HPLC.

CONCLUSION(S): ILOQ of metazosulfuron was 2 ng (S/N ≥ 10) and good linearity was achieved between 0.05 and 12.5 mg/Kg of metazosulfuron standard solutions, with coefficients of determination of 0.9999. MLOQ was 0.02 mg/Kg. Good recoveries from 74.1 to 116.9% with coefficients of variation (C.V.) of less than 10% were obtained, regardless of sample type, which satisfies the criteria of Korea Food and Drug Administration (KFDA). Those results were reconfirmed with LC-MS (SIM). The method established in this study is simple, economic and efficient to be applied to most of crops as an official and general method for residue analysis of metazosulfuron.

Key Words: High performance liquid chromatography, Limit of quantitation, Metazosulfuron, Method limit of quantitation, Recovery

Introduction

Pesticides have been used to protect food crops from pests (diseases, insects, weeds, nematodes and etc.) for many years. Approximately one-third of the world’s food crops is destroyed by pests during growth, harvesting and storage (Ware and Whitacre, 2004).

However, with their use, the risk of residues remaining on the food is a major concern of food safety issues. Legislations were enacted through the world to regulate pesticides in food products (Ahmed, 2001). The pesticide residue levels in foodstuffs are
High Performance Liquid Chromatographic Method for Determination of Metazosulfuron Residue in Representative Crops

controlled by MRLs (maximum residue limits) (Torres et al., 1996), which have been set by government agencies to guarantee consumer safety and to regulate international trade. Korean Food and Drug Administration (KFDA) established 11,376 MRLs for 427 pesticides and 368 crops/food in 2012 (KFDA, 2012). For this reason, a variety of analytical methods has been developed and applied routinely for the quantitative detection of pesticide residue in food. Due to the low detection levels required by regulatory bodies and the complex nature of the matrices in which the target compounds are present, efficient sample preparation through many distinctive steps and trace-level detection and identification are important aspects of analytical methods (Stoytcheva, 2011). Therefore, analytical methodologies employed must be capable of residue measurement at very low levels and must also provide unambiguous evidence to confirm both the identity and the magnitude of any residues detected (KFDA, 2012). In the Food Code (KFDA, 2012), many analytical methods have been developed and applied routinely for the monitoring of pesticide residues in food.

Metazosulfuron [1-[(3-chloro-1-methyl-4-[(5RS)-5,6-dihydro-5-methyl-1,4-dioxazin-3-yl]-pyrazol-5-ylsulfonyl]-3-(4,6-dimethoxypyrimidin-2-yl)urea] (Fig. 1), the subject pesticide, is a pyrimidinyl sulfonylurea herbicide developed by Nissan Chemical Industries, Ltd. (Lee et al., 2011). It was registered in 2012 in Korea for control of Echinochloa crus-galli, Monochoria vaginalis, Bindens tripartita, Cyperus difformis, Ludwigia prostrata, Eleocharis kuroguwai, Sagittaria trifolia, and Scirpus juncoides in rice (Lee et al., 2011; KCPA, 2012).

Sulfonylurea herbicides interfere with acetolactate synthase (ALS), the enzyme responsible for synthesis of branched-chain amino acids such as valine, leucine, and isoleucine. The inhibition of this enzyme disrupts the plant’s ability to manufacture proteins, with such disruption leading subsequently to the cessation of all cell division and eventual death of the plant (Krämer et al., 2012).

Analysis of sulfonylurea herbicide residues were reported for grains (Ishimitsu et al., 2002; Saha et al., 2003; Kang et al., 2011), seeds (Ishimitsu et al., 2002), vegetables (Ishimitsu et al., 2002), soil (Holloway et al., 1999; Menne et al., 1999; Saha et al., 2003) and drinking water (Gallitzendörfer et al., 2011) using HPLC/UVD or MS, capillary electrophoresis, bioassays, and enzyme-linked immunosorbent assays. However, no report was available for the analysis of metazosulfuron residues in crop or food, since it is a relatively new sulfonylurea herbicide.

The purpose of this study is to develop a novel analytical method of metazosulfuron residues in crop/food using HPLC, which can be used generally and officially for many different crop/food samples through full method validation. As crop samples for study, representative crops were selected among five crop groups such as cereal, fruits, vegetables, beans/oily crops and potatoes.

Materials and Methods

The subject pesticides and crops

Metazosulfuron (91.3%, purity) (Fig. 1) was obtained from Kyung Nong Corporation (Seoul, Korea). Brown rice, apple, mandarin, Kimchi cabbage and soybean of “residue-free grade” were purchased from local market. They were chopped, macerated and kept in a freezer at a temperature below -20 °C in polyethylene bags.

Chemicals, reagents and standard solutions

Acetonitrile, acetone, n-hexane, dichloromethane, methanol and ethyl acetate were HPLC grade (Burdick and Jackson®, Ulsan, Korea). Sodium sulfate, anhydrous (GR grade) and sodium chloride (GR grade) were from Samchun Pure Chemical Co. Ltd. (Pyeongtaek City, Korea). Florisil® (60-100 mesh, Fluka™, Sigma-Aldrich Co., Switzerland) and silica gel (60-230 mesh, Merck, Frankfurter, Germany) were used for glass column clean-up while Florisil®, silica gel, aminopropyl (NH₃) SPE cartridges (1 g, SepPak, Waters corp.), and alumina N SPE cartridge (1 g, Supelco, Sigma-aldrich Co.) were used for solid phase extraction clean-up system. Acetic acid and formic

Fig. 1. Structure of metazosulfuron.