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A Sensitive SPE-ECD-GLC Procedure for Analyzing Simazine Quantitatively in Food Products

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Abstract

Short and simple solid-phase extraction (SPE) method interfacing and hyphenating gas liquid chromatographic-electron capture detection (GLC-ECD) method has been developed for the qualification and quantification of simazine in some daily used agro products. United States Pharmacopeia G-42 gas chromatography stationary phase with ZB-35a, HTb Inferno-cZ (Zebron, Za) capillary column with an enlarged and extended temperature range of –60° to 400°C and electron capture detector was selected correlating some chemical, physical and structural features of Simazine. Attempted the higher temperature range which was programmed with 20–25 minutes having 0.94 ml/min of gas flow showing theoretical plates (~10^6) and optimum resolution (R_s ~ 2.04 min) with a limit of detection and limit of quantification values of 0.0013 µg/L and 0.13 µg/L, respectively. Accurate LOQ was achieved by using this method and is almost below the maximum residue concentrations recommended by the Indian Pharmacopeia Standards. Recoveries of the simazine obtained from agricultural samples by spiking ranged from 79.77 to 99.60% with relative standard deviations far below 3.17 %. More than five minutes of SPE residual workup makes the proposed method simple and fast having good economic importance for the quantitative analysis of simazine in common environmental institutes, medical colleges, and civil hospitals serving society.

Keywords: Simazine, LOD, LOQ, GLC-ECD, residues

INTRODUCTION

Simazine (Figure 1), a triazine herbicide, is used at higher rates to control broad-leaved weeds in berry, turf grass, vineyards, and vegetable and ornamental crops. It is used for nonselective weed control in industrial areas and in several crops including maize, rice corn, potato, wheat, apple, and common forestry products [1–4]. Simazine (boiling point: 225–227ºC) in common agricultural products has caused public concerns because of its toxicity to humankind. Many organizations like USP and EU concentration of this herbicide in common agroproducts is increasing and exceeds the maximum residual limit of 0.1 µg/L fixed by European Legislation. Most sensitive and selective analytical methods are thus needed to ensure compliance with these criteria and to support environmental, health, and toxicological studies using trace amounts of this chemical simazine [5–7].

UV-vis spectrophotometry [8, 9] electrochemical method [10, 11] thin layer chromatography (TLC) [12, 13] immunoassay [14], Gas-liquid chromatography (GLC)) [14–19] and RP-high performance liquid chromatography (RP-HPLC) [20–22] techniques are more often used for the quality analysis of simazine.

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Gas solid chromatography (GSC), gas-liquid chromatography (GLC), or gas chromatography (GC) methods are more reliable for their quantification due to the large thermal stability of simazine at high temperatures [23] and are advantageous over existing RP-HPLC. SPME and gas chromatography coupled with mass spectrometry (GLC-MS) increase the sensitivity in quantification of simazine but are costlier and require more minutes in comparison to the sensitive flame ionization detector used less in practice and in literature [24]. Moreover, ECD detectors are more sensitive to halogenated compounds and simazine is one of them, which also supports the solid phase extraction (SPE) involving minimum solvent use and represents the easy alternative to old extraction methods and quantification criterion. A very low-cost, cheap, and more effective SPE (60 mesh ~ 250b micron, with internal diameter of 1.2 cm homogeneous column, silica-based) with more polar solvent using methyl and ethyl-acetate elution has been developed and technically fabricated for residual workup [25, 26] and finishes the extraction procedure steps even lesser in 5 minutes efficiently.

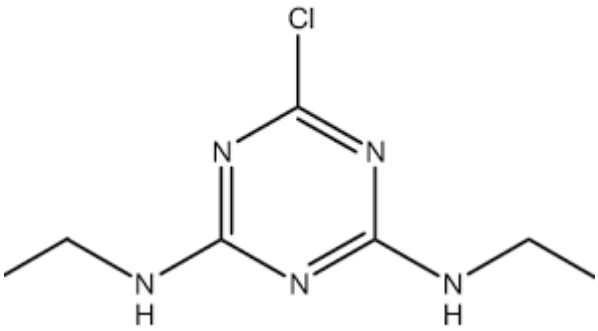

Figure 1. Spectroscopically elucidated structure of simazine.

EXPERIMENTAL

Reagents

The standard simazine (99.00% pure) was procured from sigma-aldrich. High purity methanol and ethyl acetate of the analytical grade of Rankem Company.

Method

A 7890-Agilent GLC system (Germany-made**)** with inbuilt chem station-B.02.01-version software fitted with an ECD detector was used for GC analysis of simazine. The GLC chromatographic capillary column ZB^{c}-35 HT-Inferno-Z (^{c}Zebron) comprising 35.5% phenyl-methyl polysiloxane (dimension: 30 m long, 0.2 mm of internal diameter, with 0.33 μm particle size) was used with care, equivalent to USP G-42 standard long column GLC having splitless injector with negative temperature range of –50° to 410°C with fine injector liner of i.d.0.75 mm to quantify simazine.

Carrier Gas

Absolutely, 99.9% pure Helium gas with Nitrogen having a fixed flow of 0.9 mL/min was optimized and set to the calibrated GLC system.

Programming the Temperature Gradient and Slope

The initial temperature was set to 70°C (hold for 2 min), raised to 210°C at a thermal speed of 10°C/min and finally programmed sharply to 390°C (hold for 2 min) at a rate of 10°C/minute fixing the initial injector temperature at 220°C and 1 μL (splitless mode) simazine volume in milliliters.

Column Preparation

A sensibly simple glass-made column of ~15 cm in length and 1.2 cm in diameter was set, silica 8 g (silica gel 60–120 mesh size) for column chromatography was taken 15 mL of polar methyl-ethyl acetate and filled the S-slurry to spread silica gel fine, uniform-homogeneously to slab and used a glass column of ~15 cm long with a diameter of 1.2 cm in practice. The 8 g (silica gel 60 0150120 mesh size) and 15

mL of polar solvent methyl ethyl acetate were used for column chromatography. It was filled with the slurry to spread a fine layer of silica gel homogeneously to the slab.

Calibration Curve

Standard solutions of simazine were prepared in methanol, and a calibration curve was made for quantitative measurements using the solutions as listed in Tables 1–3. The calibration curve was plotted between area and ppm concentrations as the axis of known simazine solutions to quantify (Figure 2).

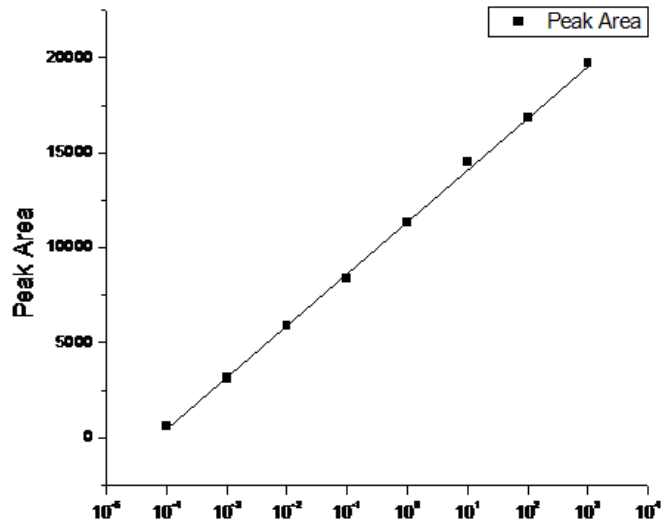

Figure 2. Calibration curve of simazine.

Formulation Analysis of Simazine

Dhanuka Agritech Ltd., Gurgaon procured, a wettable powder (WP) formulation of simazine, containing 50.5% active simazine ingredient was selected. Stock solutions of the pure-formulation equivalent to the standard solution were prepared in methanol and processed for GLC analysis. Retention time in minutes of methanolic extract of the formulation was comparable to those obtained with the purest compound thus establishing the maker's specifications of simazine-formulations and has also been established by an established method [27, 28].

Simazine Recovery Methodology

10 g weight of maize, wheat, potato, and apple were mixed with 100 µg/L solution of simazine in methanol. Simazine samples were well mixed and extracted with 2 installments in 5 ml of ethyl acetate. The simazine known samples were mixed and extracted with 10 ml of methyl-acetate and subsequent extracts were purified in a silica column extractor at a fixed and constant flow rate of 0.7–0.8 mL/minutes. Collected impure elutes were dried in a pure nitrogen gas drier dissolved in 10–15 mL methyl alcohol and quantified using GLC-ECD procedures.

RESULTS

The absolute linearity of the set procedures was investigated sharply with correlation coefficient (r^2) values of 0.998 from the regression equation, y = 2920.62x + 18915.43. Intense temperature slope, programming of 20 minutes, and 0.93 mL/minutes optimum gas flow giving maximum theoretical plates of 10^7 with high resolution of approximately 2.04 minutes precisely and quantifying simazine up to 0.11 µg/L and LOD and LOQ determined by this procedure as 0.0013 µg/L and 0.11µg/L in practice. This new method after quantification of simazine showed recovery >50% of standard ingredient which was 47.9%–55.8% in actual results and showing relative standard deviation (RSD) values in the range

of 3.6% and its GLC chromatogram is listed in Figures 3–8. Also, residual analysis proved and indicated a good-average recovery of simazine of the developed GLC-ECD method as 83.15 to 98.60% of the spiked amount and 3.07% in the row, respectively and efficiently.

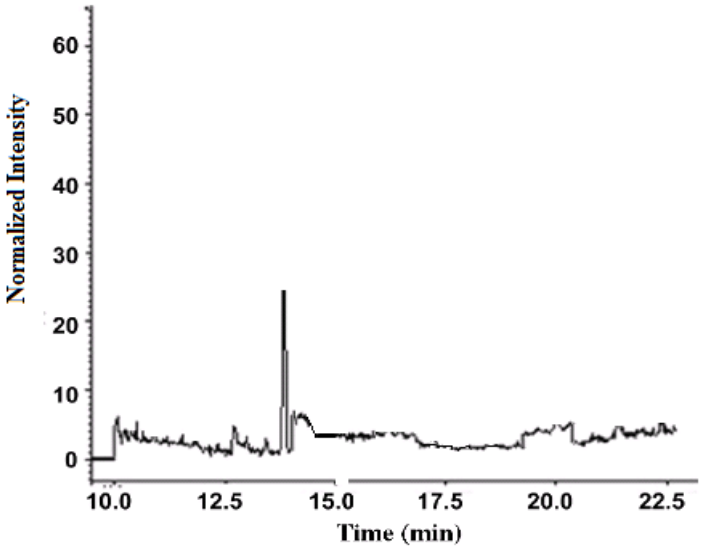

Figure 3. GLC chromatogram of blank.

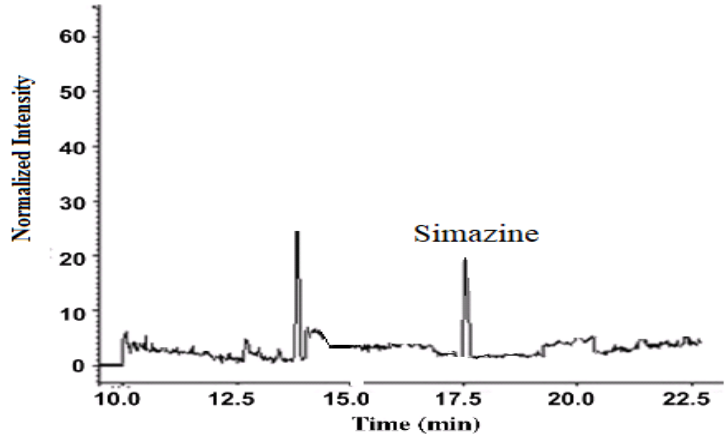

Figure 4. GLC chromatogram of standard simazine; with retention time 17.6.

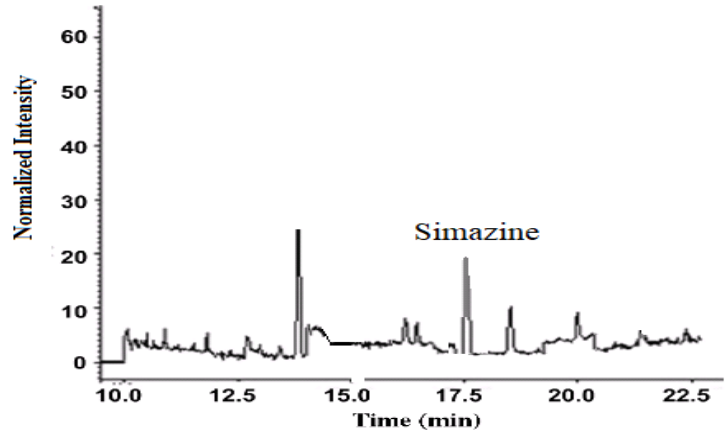

Figure 5. GLC chromatogram of commercial simazine formulation; RT = 17.6 min.

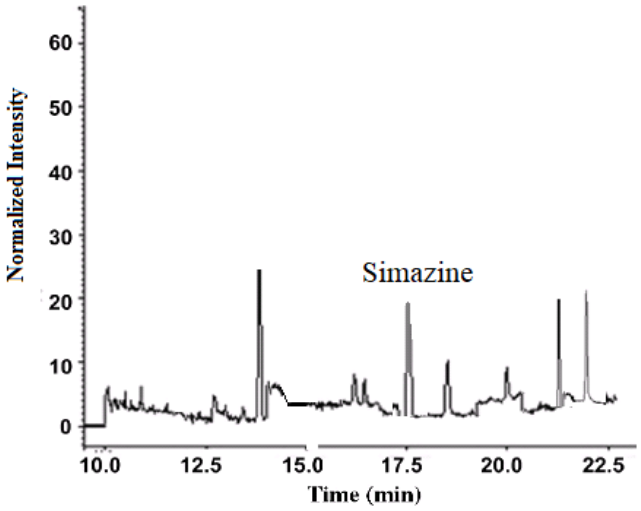

Figure 6. GLC chromatograms for simazine extracted from spiked maize samples.

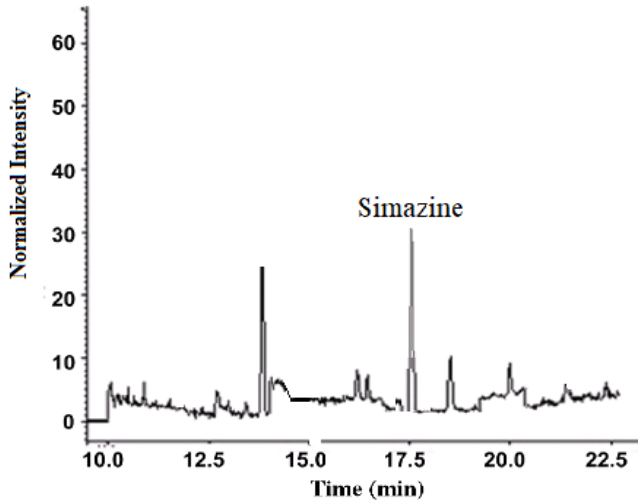

Figure 7. GLC chromatograms of simazine extracted from spiked potato samples; representative of all five recovery results.

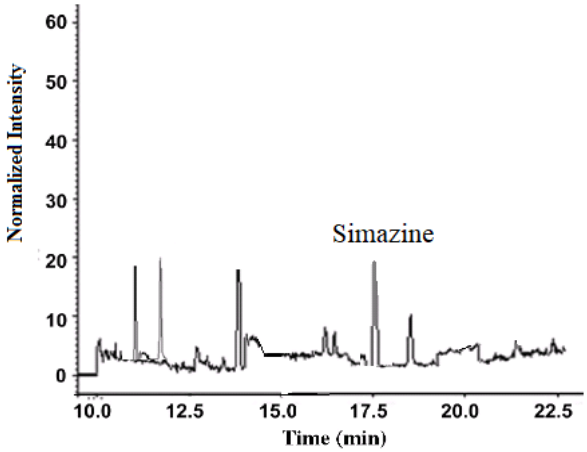

Figure 8. GLC chromatograms of simazine extracted from spiked apple samples; representative of all five recovery results.

Table 1. GLC quantization limits (QL) of standard simazine concentrations.

S.N.	Concentration (µg/L)	Retention Time (Min)	GC Peak Area	Limits	S/N Ratio
1	10^3	17.6	21003		29.9
2	10^2	17.6	19634		28.1
3	10^1	17.6	16589		21.7
4	10^0	17.6	15012		18.9
5	10^{-1}	17.6	10923	LOQ	16.2
6	10^{-2}	17.6	7613		9.0
7	10^{-3}	17.6	5098	LOD	4.6
8	10^{-4}	17.6	3125		2.1

Table 2. GLC quantization limits of simazine formulations.

Dilution Number	Concentration (µg/L)	Retention Time (Min)	GC Peak Area
1	10^3	17.6	27039
2	10^2	17.6	18889
3	10^1	17.6	16588
4	10^0	17.6	15033
5	10^{-1}	17.6	10823
6	10^{-2}	17.6	7599
7	10^{-3}	17.6	6002
8	10^{-4}	17.6	4021

Table 3. Simazine; recovery-data from spiked grains, fruit, and vegetables.

Simazine Concentration (µg)	Absolute Recovery (% Age)[a]			
	[a]Maize	[b]Wheat	[c]Apple	[d]Potato
0.10	96.37 ± 2.21	92.22 ± 2.32	85.81 ± 2.27	87.33 ± 2.08
0.15	96.43 ± 2.80	92.69 ± 2.37	83.15 ± 2.74	89.25 ± 1.27
0.20	96.65 ± 3.07	94.43 ± 2.00	83.64 ± 2.18	90.24 ± 2.83
0.25	98.60 ± 2.36	95.06 ± 2.82	85.38 ± 2.47	91.42 ± 2.49

Note: [a]Values are average ±standard deviation for six points.

DISCUSSION

Taking the advantages of GLC for high boiling point Simazine over RP-HPLC, ZB-35 HT Inferno-Z (Zebron) capillary column with excellent extended temperature range –60 to 400°C has been employed in the method development. The precise and wise selection of stationary phase (SP) and ECD-t system was intense boiling point of simazine, structural matching with long hydrocarbon chain with (N-inductive effect operations) halogen-chloride substituent and maximum residue levels – MRLs, 0.1 µg/L simazine values. Commercial lab-silica when heated to 150°C, it off-course, releases all the Simazine liquids and substances absorbed thereafter procedures can be successfully applied to the quantitative analysis of simazine in its commercial simazine formulation as well as in agricultural produces and samples and residues as well.

CONCLUSIONS

Improved procedure and method are simple and SPE GLC ECD, for the determination of simazine in some daily used agricultural samples has been successfully established, with column ZB-35 HT Inferno-Z (Zebron-C) capillary column-C with intensely-marked temperature range of –55 to 410°C was used. 20 minutes temperature slope and programming with 0.9 mL/minutes, optimized gas flow speed and theoretical plates (10^6) and high resolution > (~2.0 min) allowing the determination of simazine at 0.001 µg/L conforming to the USP, BP, and European-Union, IP standards for MRLs of simazine in agricultural stuffs as well in residues in low amount in vegetables used daily in domestic cooking.

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