Quantitative determination of urinary trichloroacetic acid as an index of trichloroethylene exposure by high performance liquid chromatography.

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Abstract

A high performance liquid chromatographic method for the determination of urinary trichloroacetic acid, a metabolite of trichloroethylene, is described. A stainless steel column packed with Hitachi gel 2618 (H form) was used and the mobile phase was one per cent aqueous phosphoric acid. Urine can be analyzed directly without any solvent extraction or pretreatment. The minimal detection limit was 0.5 micrograms per analysis. The present method is simple and specific, and can be performed within 10 min.

KEYWORDS: trichloroacetic acid, dichloroacetic acid, monochloroacetic acid, high performance liquid chromatography

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BRIEF NOTE

QUANTITATIVE DETERMINATION OF URINARY TRICHLOROACETIC ACID AS AN INDEX OF TRICHLOROETHYLENE EXPOSURE BY HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

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Abstract. A high performance liquid chromatographic method for the determination of urinary trichloroacetic acid, a metabolite of trichloroethylene, is described. A stainless steel column packed with Hitachi gel 2618 (H form) was used and the mobile phase was one per cent aqueous phosphoric acid. Urine can be analyzed directly without any solvent extraction or pretreatment. The minimal detection limit was 0.5 μg per analysis. The present method is simple and specific, and can be performed within 10 min.

Key words: trichloroacetic acid, dichloroacetic acid, monochloroacetic acid, high performance liquid chromatography.

As trichloroethylene is widely used as an anesthetic and a solvent, many people are exposed to trichloroethylene vapor during their working day. The urinary concentration of trichloroacetic acid (TCA), one of the trichloroethylene metabolites, has been suggested as an index of trichloroethylene exposure (1). Forssman and Ahlkam (2) gave 75 mg/liter of urinary TCA as the threshold limit value (T.L.V.).

Recently, a simple liquid chromatographic procedure for TCA was described by Turkelson et al. (3). However, the procedure was complicated and separation of TCA was not sufficient. The purpose of this paper is to describe a high performance liquid chromatographic procedure which is specific and sensitive for quantifying urinary trichloroacetic acid, dichloroacetic acid (DCA), and monochloroacetic acid (MCA).

The high performance liquid chromatograph (HLC Hitachi type 633), had a stainless steel column 500 mm × 8 mm internal diameter packed with Hitachi gel 2618 (H form). It also had a UV detector. A solution of one per cent aqueous phosphoric acid was a favorable mobile phase for the separation of urinary

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TCA, DCA and MCA. The flow rate was 1.0 ml/min, pressure was 15 kg/cm² and column temperature was about 25°C. The acids were detected spectrophotometrically at 220 nm and determinations were made from their peak heights.

Fig. 1. High performance liquid chromatographic separation of A: authentic samples (4 μg of TCA, DCA and MCA), and B: 6 μg of TCA added to normal urine.

Fig. 2. Calibration curves of TCA in water (— • —) and in urine (— ○ —) and MCA (— △ —) in water.
The chromatograms show that TCA, DCA and MCA were well separated from each other and eluted within about 20 min after injection (Fig. 1-A). The chromatogram of a mixed solution of TCA and normal urine is shown in Fig. 1-B, indicating that TCA was well separated from the components contained in normal human urine.

Standard solutions of TCA and MCA, ranging from 2.5 to 20.0 μg/ml, were mixed with an equal volume of water or normal urine, and one μl of this analyzed directly.

The calibration curves of TCA in water and urine were identical (Fig. 2), indicating that normal human urine did not contain TCA or any interfering substances.

Similar results were obtained for MCA solution, which appeared in small amounts in the urine of workers exposed to trichloroethylene in early stages. As up to 100 μl of urine could be applied to the column, the minimal detectable concentration of TCA was 5 mg/l, which is sufficient for determination of the TLV of urinary TCA.

REFERENCES