Method Development for Determination of EDTA in Water by Using Traditional Split/Splitless Injector – Comparing External and Internal Standard Methods of Quantification

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Abstract. Ethylenediaminetetraacetic acid (EDTA) has increasing potential as an environmentally hazardous material. Although EDTA exhibits relatively low acute toxicity, it has been found to be cytotoxic and weakly genotoxic in laboratory animals. In addition, oral exposures can cause reproductive and developmental effects. EDTA is commonly used in wood industry, pulp and paper industry, textile industry, cement industry and food industry. It is also widely used in laundry applications in order to reduce the water hardness and in many cleaning solutions.
Due to chemical properties of EDTA (polarity, relatively good solubility in water and chelating ability towards metal ions) it cannot be efficiently removed on common water treatment plants. As a result, the EDTA can be observed in the aquifer downstream near the outputs from water treatment plants of larger industrial entities. Therefore, the reliable monitoring of EDTA in water samples is of great importance. Commonly, the chromatographic methods are used for EDTA analysis with dominance of liquid chromatography coupled with UV-VIS or MS detectors. However, these methods suffer often from the lack of sensitivity towards EDTA at ppt levels. The combination of gas chromatography with high resolution MS can offer significantly lower detection limits (units of ug/l) as well as powerful identification tool. However, the derivatization of EDTA is required when GC-MS is being used. In addition, according to the Czech standard for EDTA determination the Programmed Temperature Vaporising (PTV) injector or cool on-column injection are recommended. In our paper we report on the GC-MS method development for determination of EDTA in water by using traditional split/splitless injector. We compare the external and internal standard methods of EDTA quantification for several different internal standards. The developed method was applied to analysis EDTA in real aqueous samples.

1. Introduction

The monitoring of EDTA in environmental samples is of growing interest due to increasing applications of EDTA in many industrial branches. It is widely used in wood industry, pulp and paper industry, textile industry, cement industry and food industry [1]. Significant amount of EDTA is also used in laundry applications in order to reduce the water hardness and in many cleaning solutions. [1, 2, 3]. However, the chemical properties of ETDA especially polarity, relatively good solubility in water and chelating properties cause the removal of EDTA on common water treatment plants to be difficult [4, 5]. For effective removal of EDTA the pH of treated water must be higher than8 and longer retention times of

treated water in the plant are required (more than 12 days) [6,7]. As a result, EDTA is only poorly degraded in traditional wastewater treatment plants. Synergy of these effects results in occurrence of EDTA in surface water as anthropogenic compound at significant concentration levels (hundreds of µg/l). The main environmental hazards of EDTA consist in possible mobilization of heavy and radioactive metals from sediments under specific conditions [2]. In addition, the molecule of EDTA might be a source of bioavailable nitrogen or can carry chelated phosphate [1, 2]. This way, the EDTA can indirectly cause eutrophication. The free molecule of EDTA is able to disrupt cellular division or cause necrosis in photosynthetic plants due to withdrawal of essential metals. It is also hard to be removed from the drinking water due to its hydrophilic character. Moreover, the products of EDTA degradation may support microbial growth [1, 2].

The determination of EDTA in water is commonly carried out by chromatographic methods with dominance of liquid chromatography coupled with UV-VIS or MS detectors. However, these methods are suitable for analysis of EDTA mostly at mg/l levels. The combination of gas chromatography with high resolution MS equipped with Programmed Temperature Vaporising (PTV) injector or cool on-column injection enables to achieve significantly lower detection limits (units of ug/l) for analysis of derivatized EDTA samples. [8, 9, 10, 11]

In this paper, we report on the GC-MS method development for determination of EDTA in water by using traditional split/splitless injector. We tested the external and internal standard methods of EDTA quantification using several internal standards – polyaromatic hydrocarbons, 1-chlorotetradecane, 1,2-diaminopropane-N, N, N',N'-tetraacetic acid (1,2-PDTA) and beta-alanindiacetic acid. The developed method was applied to analysis of EDTA in real aqueous samples.

The main aim of this work was to examine the applicability of traditional split/splitless injector in GC-MS system for EDTA determination as tetrabutylester derivate and validate the overall method for determination of EDTA in water by GC-MS with split/splitless injector.

2. Experimental work

Procedure for sample treatment was based on EN ISO 16588:2003[11] with small modifications. All water samples were preserved by addition of formaldehyde (1:100) in situ and immediately filtrated. After transport to the laboratory, the samples were diluted and internal standard of 1,2-PDTA or beta-alanindiacetic acid was added. This aqueous solution was evaporated to dryness under N_2 atmosphere at 90°C, acidified by 1 ml of 1M HCl and dried again. 2 ml of esterification agent (acetyl chloride, butanol 1:9) were pipetted to the dry residue and heated for 1 hour in closed vial at 90 °C. After the vials cooled down to the laboratory temperature, hexane with internal standard (16 PAHs or 1-chlorotetradecane) was added, the solution was transferred to 50 ml volumetric flask with 1 ml of 1M NaOH and filled with deionized water. After 2 minutes of vigorous shaking, the organic phase was pipetted into 4 ml vial with 0.5 g of anhydrous sodium sulphate to get the rid of the residual water. After 3 minutes of shaking, the solution was transferred to 2 ml vials and got ready to GC-MS analysis.

Calibration series of EDTA concentrations ranging between 5 - 100 ug/l was prepared by diluting appropriate amount of stock solution in 5 ml of deionized water. These calibration solutions were handled the same way as samples.

The all GC-MS analyses were done on GC-MS system consisting of gas chromatograph Bruker 456-GC, ion source Apollo II with atmospheric pressure chemical ionization and mass spectrometer Bruker compact. GC configuration was following: Autosampler Combi PAL, split/splitless injector heated to 300 °C set to splitless at the first 0.3 min and following split 1:10. Column Rxi-5ms (5% diphenyl, 95% dimethylpolysiloxane) 30 m long with inner diameter 0.25 mm and film thickness 0.25 µm was heated to 105 °C for 1.5 min followed by rate 30 °C/min up to 180 °C, then with rate 10°C

up to 300 °C held for 16 minutes. The helium mobile phase flow was set to constant flow of 1 ml/min. To minimize contact between sample and the liner in injector, pressure pulse 1.2 bar for 0.3 min has been set.

3. Results and discussions

3.1. Elimination of EDTA losses in injector

The CSN EN ISO 16588:2003 recommends cool on-column injection or PTV injector. However, our GC-MS system is equipped only with traditional split/splitless injector. After a few injections of EDTA standard solution, we observed a significant loss of EDTA when using this traditional split/splitless injector with gooseneck standardly deactivated liner with silanized glass wool. Surprisingly, the analysis of PAHs in the same samples was running without any problems or peak shape distortions proving that at least MS system was running well. In our opinion, the loss of EDTA was result of some specific interactions of EDTA in liner with active sites. In order to verify our hypothesis and eliminate this issue, we examined several injection methods (with and without pressure pulse) and types of liners - gooseneck standardly deactivated (Agilent CrossLab 8004-0101) and gooseneck highly deactivated TOPAZ (Restek) with and without silanized glass wool. After optimization process, we found that the best results with no observable loss of EDTA after 100 injections were obtained for pressure pulse injection with single goose neck highly deactivated TOPAZ liner (Restek) without any silanized glass wool in the liner. PAHs were observed in all tested variants. On the basis of obtained results we can state that the application of traditional split/splitless injector for determination of EDTA by GC-MS is possible, but the injector must be free of any active sites with highly deactivated liner TOPAZ (Restek). As it was mentioned above, even silanized glass wool can make the determination of derivatized EDTA impossible.

3.2. Comparison of external and internal standard methods of EDTA quantification

After solving the problems with losses of EDTA in injector the external standard method (ESTD) for determination of EDTA was tested as the first due to its simplicity and economy (no internal standard needed). As it is demonstrated on figure 1, the application of ESTD method for quantification of EDTA was practically impossible due to poor correlation coefficient for ESTD calibration curve (R^2=0.762).

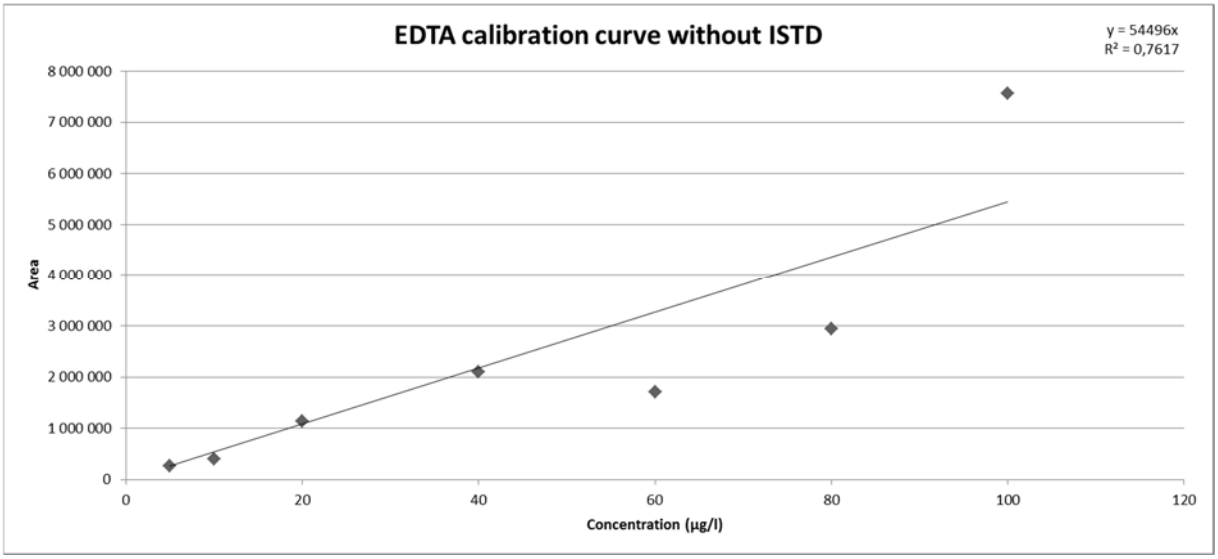

Figure 1. Calibration curve for determination of EDTA – ESTD method

That is why we focused on the development of reliable internal standard method (ISTD) for quantification of EDTA. We selected four suitable and available internal standards - 1,2-PDTA, PAHs and beta-alanindiacetic acid and 1-chlorotetradecane.

After preliminary experiments with real samples we had to exclude beta-alanindiacetic acid as internal standard due to its presence in the studied samples. The mixture of PAHs as internal standard showed insufficient correction ability of injection errors due to different behaviour in the injection system compared to EDTA. Therefore,PAHs as internal standard were used just for verification of the proper function of analytical MS system. The application of 1-chlorotetradecane as an internal standard was found to be problematic due to its thermal lability at 300°C in the injector. Thus, only the usage of 1,2-PDTA as an internal standard eliminates these random errors effectively and sufficiently due to very similar chemical composition to EDTA and the same behaviour in the injection system as EDTA. The calibration curve for determination of EDTA in water using 1,2-PDTA as an internal standard is shown on figure 2.

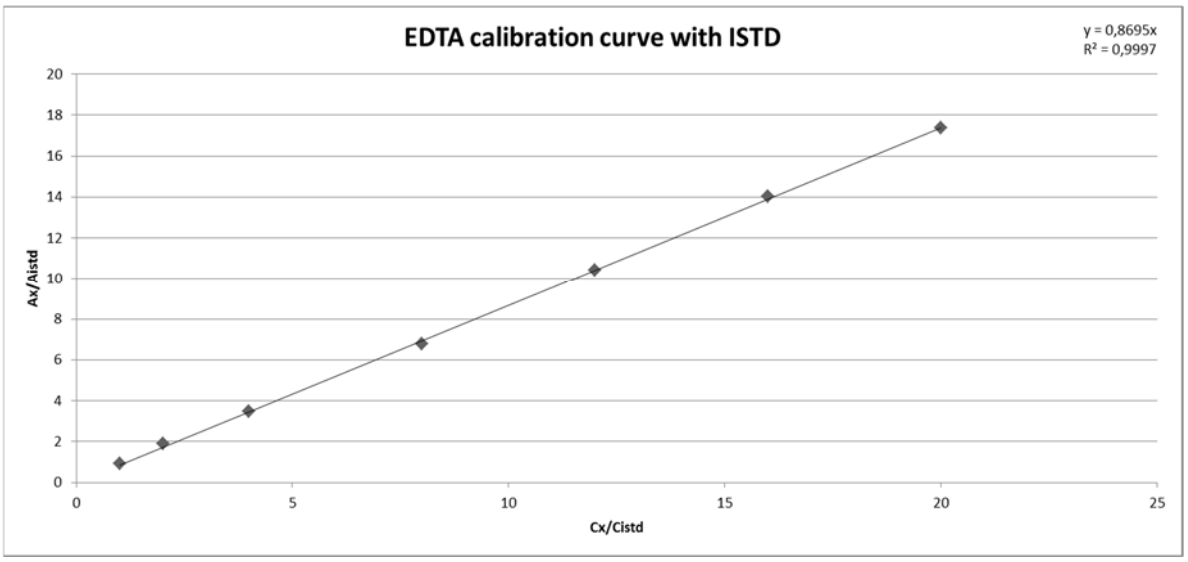

Figure 2. Calibration curve for determination of EDTA – ISTD method with 1,2-PDTA as internal standard

The distinct improvement of calibration curve for EDTA determination by ISTD method in comparison to ESTD method especially regarding the correlation coefficient (R^2=0.999) can be seen from the figure 2. Linear response of ISTD calibration curve ranges between 5–100 ug/l.

3.3. Performance characteristics of ESTD and ISTD methods for EDTA quantification

The basic performance characteristics e.g. standard deviation (STD), limit of detection (LOD), limit of quantification (LOQ), relative standard deviation (RSD) of ESTD and ISTD methods for EDTA quantification were tested at 20 µg/l level of EDTA in water. LOD with ESTD method was estimated to be 15.9 µg/l and LOQ 53.1µg/l. Correction with ISTD improved the values to 4.0 µg/l for LOD and 13.3µg/l for LOQ respectively. The RSD value for EDTA determination by ESTD method was calculated to be ± 31.0 %. By using optimized ISTD method, the RSD dropped to 3.4 %. On the basis of above mentioned data it is possible to state that the ISTD method using 1,2-PDTA as an internal standard is in all determined characteristics (LOD, LOQ, STD, RSD) better than ESTD method.

3.4. Analysis of real samples from Labe and Bílina rivers

The developed ISTD method was used for determination of EDTA in Labe and Bílina rivers . The samples were taken during July 2018in Ústí nad Labem and processed without dilution in order to achieve low concentration levels of EDTA. Concentration of EDTA in Labe was found to be 202,9 µg/l, Bílina contained 67,3µg/l.

4. Conclusions

On the basis of obtained results we proved that the application of traditional split/splitless injector for determination of EDTA by GC-MS is possible, but the injector must be equipped with highly deactivated liner TOPAZ without silanized glass wool and free of any active sites. The best results with no observable loss of EDTA after 100 injections were obtained for pressure pulse injection technique.

It was developed a reliable ISTD method for quantification of EDTA in water. We showed that only the usage of 1,2-PDTA as an internal standard eliminates random errors during injection effectively and sufficiently due to very similar chemical composition to EDTA and the same behaviour in the injection system a. When comparing ESTD versus ISTD methods we distinctly improved the calibration curve for EDTA determination by ISTD method in comparison to ESTD method especially regarding the correlation coefficient (R^2=0,999 for ISTD versus 0.762 for ESTD). Linear response of ISTD calibration curve ranges between 5–100 ug/l.

The basic performance characteristics e.g. standard deviation (STD), limit of detection (LOD), limit of quantification (LOQ), relative standard deviation (RSD) of ESTD and ISTD methods for EDTA quantification were determined at 20 µg/l level of EDTA in water. LOD with ESTD method was estimated to be 15.9 µg/l and LOQ 53.1µg/l. Correction with ISTD improved the values to 4.0 µg/l for LOD and 13.3 µg/l for LOQ respectively. The RSD values for EDTA determination were 31.0 %. for ESTD method and 3.4 % for optimized ISTD method.

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