Fully automated spectrophotometric procedure for simultaneous determination of calcium and magnesium in biodiesel

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ABSTRACT

An easily performed stepwise injection (SWIA) procedure based on on-line dilution of biodiesel samples and the formation of color-forming calcium (II) and magnesium (II) complexes with Eriochrome Black T (EBT) in an organic medium followed by spectrophotometric determination is presented. A sample of biodiesel was placed at the bottom of a mixing chamber connected to an automatic SWIA manifold. Isopropyl alcohol was used as the diluent under bubbling. The solution was submitted for on-line spectrophotometric simultaneous determination of calcium and magnesium based on the classic least-square method. The linear ranges were from 2 to 20 $\mu g\,g^{-1}$ and from 1.2 to 12 $\mu g\,g^{-1}$, and the detection limits, calculated as 3 s for a blank test ($n=5$), were found to be 0.6 $\mu g\,g^{-1}$ for calcium and 0.4 $\mu g\,g^{-1}$ for magnesium. The sample throughput was 30 h^{-1}. The method was successfully applied to the analysis of biodiesel samples.

1. Introduction

Biodiesel is a relatively new source of renewable energy, which has recently gained an increased attention worldwide [1]. Biodiesel is composed of a mixture of mono-alkyl esters of long chain fatty acids obtained through the transesterification of vegetable oils or animal fats with short-chain alcohols in the presence of a catalyst, such as compounds of calcium and magnesium [2–11]. Besides, magnesium sulfate and calcium oxide are also used as drying agents for biodiesel [12]. The presence of these metals in biodiesel may lead to the formation of solid deposits in engines [13,14]. Currently, European (EN 14214) legislation regulates Ca and Mg content in biodiesel, which cannot exceed higher than 5 $\mu g\,g^{-1}$.

Highly sensitive analytical methods, such as atomic absorption (AAS) [12–15], atomic emission [16] and inductively coupled plasma optical emission spectrometry [17–21], are widely used to determine Ca and Mg content in biodiesel. To reduce the viscosity of biodiesel samples and matrix interferences, most of these methods require sample preparation by dilution in organic solvents such as n-propanol [13,14,17], o-xylene [18], aviation kerosene [20] and ethanol [21] or microemulsion formation [22]. These sample preparation procedures allow us to use aqueous standards for calibration. Other procedures for extraction of Ca and Mg from biodiesel in

an aqueous medium, such as capillary zone electrophoresis [23,24], ion chromatography [25] and electrochemical methods [26–29], are also employed for analytical determination. Comparisons of existing methods for determination of calcium and magnesium in biodiesel are summarized in Table 1. These methods are not automatic and thus require manual sample preparations.

An important and rapidly growing trend in modern analytical chemistry is the automation of analytical procedures. Nowadays, automation of analytical procedures based on flow analysis is continuously being developed. In this way, labor costs for the analyses decrease and the volumes of both samples and reagents as well as the waste generated substantially reduce. A large number of methods based on flow analysis have been proposed for the automated analysis of biodiesel [30–36]. To the best of our knowledge, there is no procedure for the determination of calcium and magnesium in biodiesel in a flow system. SWIA is a universal solution for the automation of analytical reactions, wherein dispersion of the reaction products is prevented [37–43]. In this case the analytical signal is at a maximum and similar to the signal obtained using a manual technique. The aim of this work was to develop a fully automated procedure for the simultaneous determination of calcium and magnesium in biodiesel in stepwise injection analysis. In our work, the selective color-forming reagent EBT was chosen for the spectrophotometric determination of calcium and magnesium in an organic medium. EBT is mainly used for spectrophotometric determination of calcium and magnesium in aqueous medium [44]. To eliminate the procedure of Ca and Mg extraction from biodiesel

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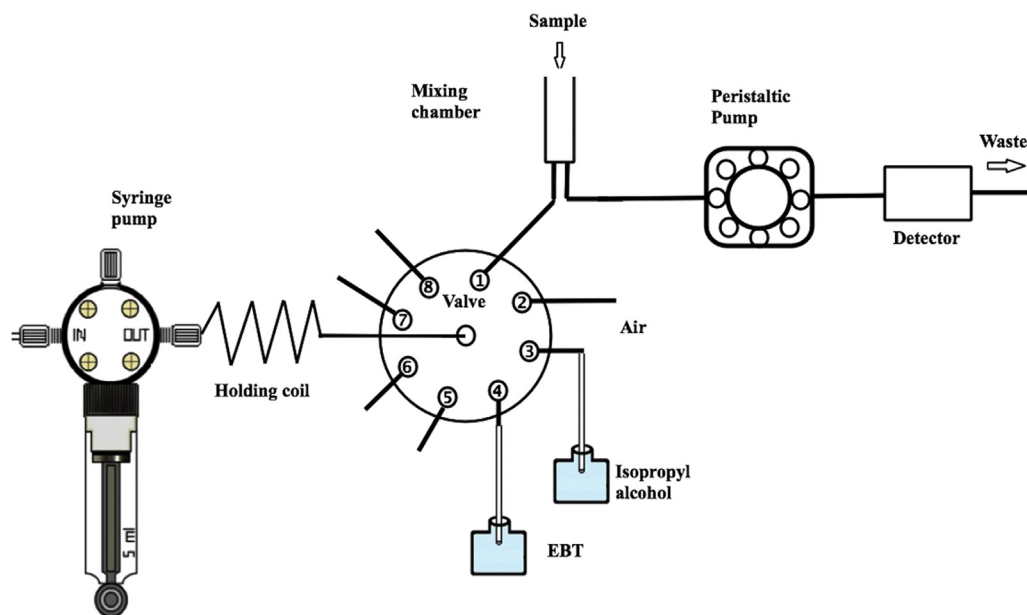*E-mail address:* andrey.shishov.rus@gmail.com (A.Y. Shishov).

Fig. 1. SWIA manifold for determination of calcium and magnesium in biodiesel.

in aqueous medium, we carried out the reactions in isopropyl alcohol based medium followed by simultaneous spectrophotometric determination of analytes based on the classical least squares method.

2. Experiment

2.1. Reagents and solutions

Analytical grade chemicals were used throughout the work. The stock solutions ($0.5\,\mu M\,g^{-1}$) of calcium and magnesium were prepared by dissolving their 2-ethylhexanoates (ABCR, Germany) in biodiesel. The EBT solution ($100\,\mu M$) (Merck, Germany) was prepared by dissolving the reagent in 0.5 M solution of triethanolamine (TEA) in isopropyl alcohol.

2.2. Samples

Biofuel samples were obtained from the following feedstocks: sunflower, canola, corn and palm. These samples were previously prepared using sodium hydroxide as an alkaline catalyzer [45] to avoid the ingress of calcium and magnesium at this stage. From GC–MS analysis [46], it was observed that the biofuel samples contained methyl esters of n-nonanoic, n-hexadecanoic, n-octadecanoic, 9-octadecenoic, and 9,12-octadecadienoic acids. The working samples of biodiesel (B20) were obtained by mixing biofuel and diesel in 1:4 (m/m) ratio.

2.3. Manifold and apparatus

The SWIA manifold (Fig. 1) included an eight-way valve (Sciware Systems, Spain), syringe pump (Sciware Systems, Spain), peristaltic pump MasterFlex L/S (Cole-Parmer, USA) (the flow rate was from 0.5 to 5 mL min^{-1}), mixing chamber (a cylindrical-shaped PTFE tube with a funnel-shaped inlet at the bottom, 20 mm height and 10 mm i.d.), holding coil and communication tubes (PTFE, 0.5 mm i.d.). The manifold was equipped with a USB 4000 spectrophotometer (Ocean Optics Inc., USA) with a 50 mm path-length flow cell (FIAlab® Instrument Systems Inc., Bellevue, USA), optical fibers QP400-2-UV–VIS (Ocean Optics Inc., USA) and

a Model D 1000 CE UV source (Analytical Instrument System Inc., USA). The analyzer was operated automatically using a computer.

An AA-7000 atomic absorption spectrometer (Shimadzu, Japan) was used for calcium and magnesium determination in the biodiesel samples. A GC–MS-QP2010 Ultra gas chromatograph mass spectrometer (Shimadzu, Japan) was applied to identify fatty acid methyl esters in the biofuel samples.

2.4. SWIA procedure

In the first step of the measurement, 0.01 g of the biodiesel sample was placed at the bottom part of the mixing chamber (Fig. 1). The syringe pump was then set to the "Out" position and 0.2 ml of isopropyl alcohol (3) was aspirated through the corresponding ports of the multiport valve into the holding coil by backward movement of the syringe pump plunger. The isopropyl alcohol was then transferred by forward movement of the syringe pump from the holding coil through the channel (1) into the mixing chamber. After the completion of this operation, 1 ml of air (2) was injected first into the holding coil and then passed through the mixing chamber at a rate of 6 ml min^{-1}.

In the second step, 0.2 ml of 100 μM EBT (4) was delivered into the mixing chamber through the holding coil. To stir the reaction mixture and complete the color-forming reactions, 1 ml of air (2) was delivered into the mixing chamber through the holding coil at a rate of 6 ml min^{-1}. This resulted in the formation of complexes of Ca (II) and Mg (II) with EBT. The formed complexes were then transferred from the mixing chamber into the flow cell of the spectrophotometric detector using the peristaltic pump. The absorbance measurement was performed under stopped-flow conditions for 5 s and the solution was discharged as waste. In the next step, all the components of the manifold were washed with isopropyl alcohol. The measurement of the analytical signal for a blank solution was carried out using the above-mentioned algorithm, but in this case isopropyl alcohol was aspirated instead of the sample solution.

2.5. Multivariate calibration

For multivariate calibration [47], we prepared a set of 10 standards containing calcium and magnesium of known concentrations in the