Analysis of metals and phosphorus in biodiesel B100 from different feedstock using a Flow Blurring® multinebulizer in Inductively Coupled Plasma-Optical Emission Spectrometry

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Abstract

A simple and fast method for determining the content of Na, K, Ca, Mg, P, and 20 heavy metals in biodiesel samples with Inductively Coupled Plasma Optical Emission Spectrometry (ICP OES) using a two-nozzle Flow Blurring® multi-nebulizer prototype and on-line internal standard calibration, is proposed. The biodiesel samples were produced from different feedstock such as sunflower, corn, soybean and grape seed oils, via a base catalyst transesterification. The analysis was carried out without any sample pretreatment. The standards and samples were introduced through one of the multi-nebulizer nozzles, while the aqueous solution containing Yttrium as an internal standard was introduced through the second nozzle. Thus, the spectral interferences were compensated and the formation of carbon deposits on the ICP torch was prevented. The determination coefficients (R^2) were greater than 0.99 for the studied analytes, in the range 0.21 - 14.75 mg Kg^{-1}. Short-term and long-term precisions were estimated as relative standard deviation. These were acceptable, their values being lower than 10%. The LOQ for major components such as Ca, K, Mg, Na, and P, were within a range between 4.9 ng g^{-1} for Mg (279.553 nm) and 531.1 ng g^{-1} for Na (588.995 nm), and for the other 20 minor components they were within a range between 1.1 ng g^{-1} for Ba (455.403 nm) and 2913.9 ng g^{-1} for Pb (220.353 nm). Recovery values ranged between 95% and 106 %.

Keywords: Multi-nebulizer, ICP OES, Inorganic Elemental Analysis, Biodiesel**.**

1. Introduction

As a result of the decline in oil world reserves and global warming, biodiesel has taken an increasing importance as an alternative fuel over the last years. One of the most relevant aspects of biodiesel is the fact that it is obtained from biomass feedstock and is therefore considered a renewable energy source. Emissions from combustion of this biofuel are essentially free of aromatic compounds, heavy metals such as Cd, Co, Cu, Pb, V and Ni and other pollutants such as NO_x and SO_x that affect the air quality [1].

According to the ASTM D6751-08 specification, biodiesel is defined as a fuel comprised of mono-alkyl esters of long chain fatty acids derived from vegetable oils or animal fats, and it is designated B100 [2]. Usually, biodiesel is obtained from vegetable oils by means of a base-catalyzed transesterification owing to the high efficiency and the low cost of this process.

During biodiesel production, contamination may occur with metals such as Na, K, Ca, and Mg, as well as sulfur or phosphorus compounds, which could arise from raw materials or from chemical reagents used. These contaminants could affect the stability and some properties of the biofuels [3, 4]. Furthermore, environmental pollution may be caused if contaminants are at high concentrations and also, undesirable compounds could be formed during the combustion, thus causing engine malfunction.

Governments and agencies around the world regulate the quality of biofuels. The European Standard EN 14214 [5], the American Standard ASTM D 6751 [2] and other Standards such as the Brazilian norm RANP 07/08 [6] or the Argentine norm IRAM 6515-1:2006 [7], describe the requirements and control methods for the most common biodiesel (B 100). These Standards establish the maximum limits for the content of Na plus K as 5 mg Kg^{-1} and Ca plus Mg as 5 mg Kg^{-1}, as well as a maximum limit of 10 mg Kg^{-1} for P. Moreover, the maximum limit established for the content of S is 10 mg Kg^{-1} according to EN 14214, and 500 mg Kg^{-1} according to ASTM D 6751. The above-mentioned regulations suggest the use of FAAS and/or ICP OES for determining the content of Na, K, Ca, Mg, S and P. In addition, the European standards EN 14107, 14108 and 14109 [8-10] require the use of xylene to dissolve biodiesel samples and recommend the determination of P content by ICP OES, and Na and K content by atomic absorption spectrometry (AAS). The ICP OES has the advantage of being a multielemental and highly sensitive analytical technique. However, interferences and operational instability may occur when organic samples are directly introduced into the plasma. Spectral interferences such as stray radiation, overlapping lines, and poor resolution of lines may be observed [11], and non-spectral interferences such as differences in nebulization, sample transport and desolvation between calibration standards and samples, carbon deposits formation at the injector tube of the ICP torch may be present. Other drawbacks for biodiesel analysis by ICP OES, are the instability of analyte solutions when prepared in an organic medium, and the need to use organometallic standards and hazardous organic solvents, which require special safety conditions.

In the last years, a number of researchers have focused their efforts on the development of new methods for the elemental analysis of biodiesel. The proposed method by Dos Santos et al. [12] for the determination of Ca, P, Mg, K and Na in biodiesel obtained from different raw materials by ICP OES with axially-viewed plasma, was based in the dilution of sample in ethanol. De Jesus et al. [13] developed a method for determining Na and K by flame atomic absorption spectrometry (FAAS) in biodiesel obtained from different sources and vegetable oils, by using water-in-oil microemulsion. Paiva de Oliveira et al. [14] developed a method for Na determination in biodiesel fuel by flame atomic emission spectroscopy (FAES), using dry decomposition as sample preparation. Henrique Lyra et al. [15] proposed a procedure for determining Na, K, Ca and Mg in biodiesel samples by FAAS and the preliminary treatment of the sample was prepared as a microemulsion without surfactant. Lobo et al. [16] proposed a method for determining Ni and Cd in biodiesel samples by graphite furnace atomic absorption spectrometry (GFASS) and samples were prepared as microemulsions. Amais et al. [17] developed a method for the determination of Ca, Mg and Zn in biodiesel samples by FAAS involving the microemulsification of samples and discrete aspiration for sample introduction. De Souza et al. [18] evaluated two micronebulizers, PFA and Miramist, using a method for elemental determination by ICP OES in lubricant and residual fuel oils diluted in xylene. This investigation allowed the improvement in the signal-to-background for Ag, Al, Ba, Cr, Cu, Fe, Mn, Si, Ti and V using an oxygen flow that reduced spectral interferences from carbon emission bands. PFA micronebulizer enabled the authors to obtain higher sensitivity, while the limits of detection (LOD) obtained for both micronebulizers were comparable. The above-mentioned methods suppose some disadvantages from an analytical and environmental point of view such as the requirement of organic solvents and complicated procedures for the pretreatment of biodiesel sample such as dilution, emulsification and decomposition of the sample.

On the other hand, the use of a multiple nebulizer for simultaneous introduction of different liquid samples/reagents into plasmas, which allows the application of “in chamber” sample preparation procedures, has been proposed recently [19]. The initial four-nozzle prototype, based on the Flow Focusing® nebulization principle has been followed by more an advanced prototype based on the Flow Blurring® nebulization technology (Flow Blurring® multinebulizer (FBMN)). It has been applied to various “in-chamber” sample preparation procedures such as on-line standard addition calibration for organic sample analysis using aqueous calibration standards [20], for the correction of matrix effects caused by high dissolved salts [21], as well as for volatile forms generation by in-chamber derivatization [22].

In this work, an evolution in the FBMN design is presented. The prototype described, incorporates two nebulization nozzles, as opposed to three in previous designs. This allows higher nebulization gas pressure to be used for the same flow rate, aiming to obtain better operational characteristics and to generate finer aerosols. Another advantage of the current prototype is that it can be used in commercial spectrometers without the need of external peristaltic pump or nebulization gas supply.

In view of the above, the aim of this work is to propose an alternative strategy for ICP OES analysis of trace elements in biodiesel obtained from different feedstock, based on the implementation of a new evolution of the Flow Blurring® multinebulizer (FBMN) in the sample introduction system. The FBMN allows the simultaneous nebulization of non-pretreated biodiesel samples and water in order to decrease the Swan band emission background, and to prevent accumulation of carbon deposits on the ICP torch injector. In comparison with previously published applications of earlier FBMN prototypes to the analysis of biodiesel samples by in-chamber standard addition calibration (aiming for high analysis accuracy), the methodology proposed in this work is a simple procedure that achieves significantly higher sample throughput, while maintaining the above mentioned benefits of simultaneous nebulization of organic sample and water.

2. Experimental

2. 1. Reagent and standards

Pro-analysis grade chemicals and ultra-pure water (18.3 Ω cm^{-1}) were used. Nitric Acid (Suprapur® 65%) and Yttrium monoelemental standard (Certipur® 1000 mg L^{-1}) were purchased by Merck (Darmstadt, Germany). Yttrium internal standard solution of 1 mg L^{-1}

was prepared by diluting appropriate amounts of 1000 mg L^{-1} Yttrium internal standard solution in 1% nitric acid.

Conostan S-21 multielement standard solution containing 500 µg g^{-1} of Ag, Al, B, Ba, Ca, Cd, Cr, Cu, Fe, Mg, Mn, Mo, Na, Ni, P, Pb, Si, Sn, Ti, V, Zn blended in 75 cSt blank oil, Conostan K standard solution containing 1000 µg g^{-1} of K blended in 75 cSt blank oil and Conostan Base Oil of 75 cSt, all purchased by SCP Science (Baie d'Urfé, Canada) were used. Certified biodiesel matrix blank (used as blank for calibration) and a 5-element biodiesel certified reference material (NIST SRM: 1085b) containing 9.9 µg g^{-1} (±1%) of Ca, Mg and P, and 10.0 µg g^{-1} (±1%) of K and Na were purchased from Spex CertiPrep (Metuchen, USA). Organometallic calibration solutions were prepared by diluting appropriate amounts of Conostan S-21 and Conostan K in Spex CertiPrep biodiesel matrix blank in the range of 0.2 to 14.7 µg g^{-1}.

2. 2. Samples

Fourteen samples of biodiesel were obtained from sunflower, corn, soybean and grape seed oils, in the FIA Laboratory (INQUISUR-UNS-CONICET), while other samples were purchased from companies in Argentina.

The optimized synthesis process, based on base-catalyzed transesterification to obtain biodiesel from different vegetable oils, and the methods applied to biodiesel quality characteristics assessment such as density, kinematic viscosity, acid index and oxidative stability (according to the Argentine Standard IRAM 6515:1 – 2006) were published in a previous work [23]. These procedures were also used in this work for sample synthesis and assessment.

2. 3. Instrumental

An axial view inductively coupled plasma optical emission spectrometer Agilent 720 ES (Agilent Inc., Melbourne, Australia) was used. The instrumental operation parameters are summarized in Table 1. The following wavelengths were selected to measure the emission intensities of the analytes: 328.068 nm for Ag, 396.152 nm for Al, 249.772 nm for B, 455.403 nm for Ba, 393.366 nm and 396.847 nm for Ca, 226.502 nm for Cd, 267.716 nm for Cr, 324.754 nm for Cu, 238.204 nm for Fe, 766.491 nm and 769.897 nm for K, 279.553 nm and 280.270 nm for Mg, 257.610 nm for Mn, 202.032 nm for Mo, 588.995 nm and 589.592 nm for Na, 216.555 nm for Ni, 213.618 nm for P, 220.353 nm for Pb, 288.158

nm for Si, 283.998 nm for Sn, 336.122 nm for Ti, 292.401 nm for V, and 213.857 nm for Zn. The internal standard (Y) was measured at 360.074 nm. Peak height was used as signal for quantification. A vortex mixer ZX^3 Vortex (Velp Scientifica, Monza, Italy) was used to homogenize all the organometallic standard solutions and samples.

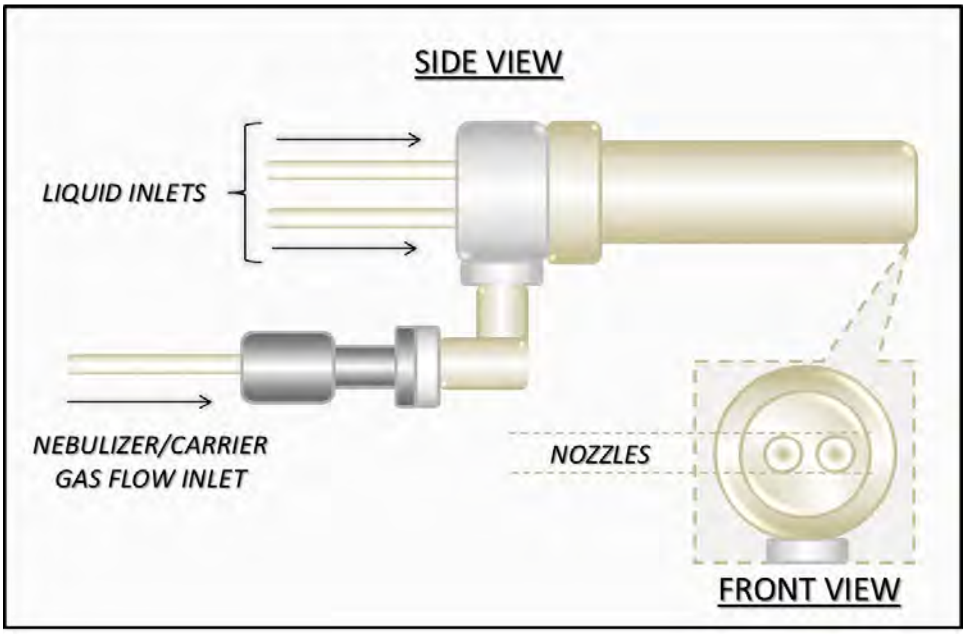
Table 1. Instrumental parameters for ICP OES instrument.

Parameter	***Value***
Radiofrequency power	1.35 KW
Plasma gas flow rate	15 L min^{-1}
Auxiliary gas flow rate	1.5 L min^{-1}
Nebulizing/carrier gas flow rate	0.7 L min^{-1}
Peak processing	peak height
Background correction	2 points
Number of replicates	5
Replicate read time	1 s
Sample flow rate	0.2 mL min^{-1}
Water (with IS) flow rate	0.5 mL min^{-1}
Nebulizer type	two-nozzles Flow Blurring® multinebulizer prototype
Spray chamber	single-pass cyclonic chamber (50 mL)

2. 4. Standards and samples introduction system

The sample introduction system used in this work consists in assembling the new two-nozzle Flow Blurring® prototype (Figure 1) and one commercial single-pass cyclonic spray chamber of 50 mL (Model Tracy, Glass Expansion Ptr. Ltd, Melbourne, Australia). The FBMN prototype consists in two nebulization nozzles (Flow Blurring® technology nebulization units) with separate liquid inlets and a common nebulization gas inlet, all of them set into a cylindrical PTFE body (Figure 1). The hydrodynamic principles, in which the Flow Blurring® nebulization units are based, have been exhaustively discussed in previous works [20-24]. The dimensions and physical characteristics of this multinebulizer prototype permit its direct connection to different commercial spray chambers, commonly used in ICP OES instruments. The advantage of this prototype compared with previously described multinebulizers of three or four nozzles [19-23] consists, on one hand, in the ease of operation, as no additional peristaltic pump is needed (ICP OES instrument with a three channel pump is sufficient). On the other hand, the nebulization gas flow is divided only into two streams which lead to higher nebulization pressure (no external gas supply needed) and, hence to better aerosol characteristics. F-4040-A, id. 0.25 mm (Ismatec, Switzerland) peristaltic pump tubes for organic solutions and Tygon® R-3607, id. 0.51 mm (Ismatec, Switzerland) peristaltic pump tubes for aqueous solutions were employed to pump all liquids throughout the system. Also, a Vulkan® direct injection nebulizer (Glass Expansion, West Melbourne, Australia) and a MicroMist® nebulizer (Glass Expansion, West Melbourne, Australia) have been used for comparison.

Figure 1. Side and front views of the two-nozzle Flow Blurring® multinebulizer.



2. 5. Procedure

External calibration with on-line addition of internal standard was used as calibration method in order to compensate interferences in the analysis of biodiesel samples. Seven organometallic standard solutions at 0.21, 0.54, 1.01, 2.96, 5.76, 12.46, 14.75 mg Kg^{-1} were used. The calibration procedure consisted in the continuous introduction of the aqueous solution (containing 1 mg L^{-1} Yttrium as internal standard) into the spray chamber through one of the two nebulization nozzles, while the organometallic standard solutions were consecutively introduced through the other nebulization nozzle. After calibration, the organic samples were introduced in the same way as the organometallic standard solutions.

3. Results and discussion

3. 1. Biodiesel samples

A number of additive-free biodiesel samples (Table 2) from different feedstock, were obtained by means of a methanolysis reaction using sodium hydroxide as catalyst. The conversion percentages of triglycerides in fatty acid alkyl esters were between 91 and 96 %. The quality of the biodiesel samples was evaluated according to the requirements and methods established by the Argentine standard IRAM 6515:1 – 2006 [7]. Physical-chemical parameters such as density, kinematic viscosity, acid index and oxidative stability were measured for the biodiesel samples obtained at laboratory scale and commercial samples, and the results are shown in Table 2. These results are in accordance to the limits assessed in the Argentine Standard mentioned above [7], except for the kinematic viscosity values in samples S1 and S3, which were higher.

Table 2. Physical-chemical parameters of the different biodiesel samples studied.

Sample	***Density 15 °C ($g\ mL^{-1}$)***	***Kinematic Viscosity 40 °C ($mm^2\ s^{-1}$)***	***Oxidative Stability 110 °C (h)***	***Acid Index ($mg\ KOH\ g^{-1}$)***
IRAM 6515-1 Limits	**0.875 - 0.900**	**3.5 - 5.0**	**Min. 6**	**Max. 0.5**
S1 - Sunflower biodiesel	0.918	13.1	0.4	0.4
S2 - Corn biodiesel	0.872	4.2	0.6	0.3
S3 - Grape seeds biodiesel	0.917	12.9	0.4	0.4
S4 - Soybean biodiesel	0.870	3.6	2.2	0.3
S5 - Biodiesel Company 1	0.876	4.7	0.5	0.3
S6 - Biodiesel Company 2	0.869	3.6	2.0	0.6
S7 - Sunflower biodiesel	0.899	4.9	0.4	0.4
S8 - Corn biodiesel	0.885	3.9	0.6	0.3
S9 - Grape seeds biodiesel	0.891	4.7	0.4	0.4
S10 - Soybean biodiesel	0.889	4.4	2.2	0.3
S11 – Sunflower biodiesel	0.908	4.6	0.6	0.3
S12 – Corn biodiesel	0.878	4.1	0.6	0.3
S13 – Grape seeds biodiesel	0.904	4.3	0.2	0.4
S14 – Soybean biodiesel	0.879	3.9	0.4	0.3

3. 2. Sample introduction system selection

Three different arrangements for the introduction of organic standard solutions and samples were considered in this work, in order to select the most suitable sample introduction system for the determination of sodium, potassium, heavy metals, and phosphorus in samples of biodiesel, obtained from different feedstock, by ICP OES.

The first introduction system consisted in a Vulkan® direct injection nebulizer, the second one was provided with a MicroMist® microconcentrical nebulizer and a cyclonic spray chamber of 50 mL. However, the implementation of these sample introduction systems was not successful owing to the typical issues related to the introduction of organic samples into ICP, such as low plasma stability, formation of carbon deposits on the ICP torch, as well as presence of spectral interferences. Therefore, a third introduction system consisting of a two-nozzle Flow Blurring® multinebulizer prototype and a cyclonic spray chamber of 50 mL, was finally used for the inorganic elemental analysis of the different biodiesel samples. The advantages of the FBMN-based introduction systems have been already described [20-23]. In this work a new version of a multinebulizer prototype was used in order to develop a simple, fast and robust method for determination of metals in organic samples without previous treatment, while maintaining the advantage of simultaneously introducing organic and aqueous solutions (increased plasma stability, elimination of spectral interferences caused by Swan bands and elimination of carbon deposition on the ICP torch).

3. 3. Analytical figures of merit

The performance of the two-nozzles FBMN-based sample introduction system proposed in this work for the elemental analysis of biodiesel by ICP OES, was evaluated studying different analytical figures of merit: (i) linearity, (ii) precision, (iii) limits of detection, quantification and linear range.

(i) Linearity: The 'best fit' line estimation using the least squares method, was applied to a set of test results in order to confirm the correlation between the measured emission signals and the analyte standard concentrations. Table 3 shows the values obtained for the slope, intercept, and coefficient of determination (R^2) for each of the studied analytes, when a set of standard solutions in a range of 0.21 – 14.75 mg Kg^{-1} was used. The coefficient of determination values of the calibration curves were higher than 0.998, and the intercept values were close to zero except for Ca 393.366 nm and Ca 396.897 nm, with values of 1.7 and 2.4 cps $(ng\ g^{-1})^{-1}$, respectively. These results indicate a reasonable linearity, and confirm the fact that the analytical calibration curves are able to correlate test results directly or mathematically/statistically proportional to the concentration of the analytes in the biodiesel samples.

(ii) Precision: In this work, short-term (in-run) and long-term (1 hour) precisions were assessed by means of the RSD of several emission signal measurements consecutively taken in a specific period of time. For this purpose, 1 mg Kg^{-1} multielemental organic standard solution was nebulized into the ICP-based spectrometer using the introduction system proposed in this work at discrete times of 8 min (in-run) and 2 hours during the analysis. The RSD values shown in Table 3 for short-term precision ranged between 0.8% for Cu (324.754 nm) and Fe (238.204 nm), and 5.5% for P (213.618 nm), and the values for long-term precision ranged between 2.6% for Pb (220.353 nm) and 8.8% for P (249.772 nm). Although the proposed method provided a relatively low short- and long-term precisions for the analysis of P at 1 mg Kg^{-1} concentration level, it had better average RSD values for short- and long-term precisions, 1.9% and 4.1% respectively, than the whole set of emission lines at the same concentration level. This means that the average RSD values were close to 3% RSD, which are quite acceptable especially when organic samples/standards with low analyte concentrations are analyzed, and taking into account that these analyses are commonly affected by carbon deposition on the ICP torch.

(iii) Limits of detection and quantification, and the linear range: Table 3 shows the limits of detection (LOD) and the limits of quantification (LOQ) for the different analytes studied in this work. The LOD was based on 3 times the standard deviation of 5 measurements of a blank standard, and the LOQ was based on 10 times the standard deviation of 5 measurements of a blank standard. Biodiesel matrix blank was used for the determination of these parameters. The obtained LOQ values for the major contaminants such as Ca, K, Mg, Na, and P, ranged between 4.9 ng g^{-1} for Mg (279.553 nm) and 326.2 ng g^{-1} for Na (589.592 nm).

It is important to notice that these LOQ values were well below 5000 ng g^{-1}, which is the maximum allowable limit established by the Argentine Standard IRAM 6515:2006 [7], the European Standard EN 14214 [5], and the American Standard ASTM D 6751 [2] for those contaminants in biodiesel. The obtained LOQ values for the other 20 minor components were within a range between 1.1 ng g^{-1} for Ba (455.403 nm) and 2913.9 ng g^{-1} for Pb (220.353 nm).

The linear range of an analytical method evaluated by visual inspection is defined as the interval between the lowest concentration in which the quantitative measurements can be made (LOQ) and the concentration in which the calibration curve loses its linearity, which is called limit of linearity (LOL). As can be seen in Table 3, the linear ranges for the different analytes studied by the proposed analytical method were at least two orders of magnitude. The linear range in the case of Ba was wider, and for Pb it was bounded. Therefore, it can be said that the use of the proposed method is limited for the determination of Pb. This is partially due to the low sensitivities of the emission lines of this element in ICP OES, especially at low sample uptake rates required for the organic sample introduction.

Table 3. Analytical parameters associated with the validation of the proposed method for the inorganic elemental analysis by ICP OES in biodiesel samples.

Element Emission Line	***LOD (ng g^{-1})***	***LOQ (ng g^{-1})***	***Short-term RSD%***	***Long-term RSD%***	***Linear range (ng g^{-1})***	***Slope (cps (ng g^{-1})$^{-1}$)^{a}***	***Intercept (cps)^{a}***	***R^2***
Ag 328.068 nm	3.6	10.9	2.2	3.1	10.9 – 14750.0	0.0812 ± 0.0009	-0.012 ± 0.006	0.9994
Al 396.152 nm	26.9	81.4	3.0	5.0	81.4 – 14750.0	0.068 ± 0.001	0.023 ± 0.004	0.9996
B 249.772 nm	10.3	31.1	1.2	3.1	31.1 – 14750.0	0.0453 ± 0.0004	0.002 ± 0.004	0.9996
Ba 455.403 nm	0.4	1.1	0.9	3.5	1.1 – 14750.0	4.5 ± 0.1	0.7 ± 0.4	0.9994
Ca 393.366 nm	10.6	32.0	0.9	3.7	32.0 – 12460.0	5.2 ± 0.1	1.7 ± 0.5	0.9987
Ca 396.847 nm	19.2	58.2	1.0	3.6	58.2 – 12460.0	8.7 ± 0.1	2.4 ± 0.6	0.9993
Cd 226.502 nm	12.7	38.5	1.3	4.8	38.5 – 14750.0	0.056 ± 0.001	0.004 ± 0.004	0.9995
Cr 267.716 nm	8.6	26.1	1.5	4.2	26.1 – 14750.0	0.058 ± 0.001	0.005 ± 0.004	0.9996
Cu 324.754 nm	2.9	8.9	0.8	3.1	8.9 – 14750.0	0.1127 ± 0.0003	0.001 ± 0.002	0.9999
Fe 238.204 nm	42.0	127.2	0.8	3.0	127.2 – 14750.0	0.0514 ± 0.0004	0.006 ± 0.003	0.9997
K 766.491 nm	16.3	49.5	1.2	3.2	49.5 – 14750.0	0.1092 ± 0.0004	-0.006 ± 0.003	0.9999
K 769.897 nm	25.2	76.3	1.2	4.7	76.3 – 14750.0	0.0711 ± 0.0003	0.014 ± 0.002	0.9999
Mg 279.553 nm	1.6	4.9	3.3	5.1	4.9 – 12460.0	1.18 ± 0.01	0.11 ± 0.05	0.999
Mg 280.270 nm	1.7	5.1	1.4	4.0	5.1 – 12460.0	0.291 ± 0.001	0.015 ± 0.005	0.9999
Mn 257.610 nm	2.4	7.3	1.3	3.8	7.3 – 14750.0	0.297 ± 0.002	0.021 ± 0.017	0.9997
Mo 202.032 nm	58.4	176.9	1.4	3.4	176.9 – 14750.0	0.0129 ± 0.0001	0.0006 ± 0.0004	0.9999
Na 589.592 nm	107.6	326.2	0.9	3.4	326.2 – 14750.0	0.276 ± 0.004	0.167 ± 0.031	0.9992
Ni 216.555 nm	90.4	274.1	2.3	3.4	274.1 – 14750.0	0.0086 ± 0.0001	0.0008 ± 0.0003	0.9999
P 213.618 nm	70.8	214.5	5.5	8.8	214.5 – 14750.0	0.00191 ± 0.00001	0.0004 ± 0.0001	0.9999
Pb 220.353 nm	961.6	2913.9	4.1	2.6	2913.9 – 14750.0	0.00170 ± 0.00001	0.0023 ± 0.0001	0.9998
Si 288.158 nm	81.6	247.3	1.7	5.4	247.3 – 14750.0	0.0116 ± 0.0001	0.033± 0.001	0.9995
Sn 283.998 nm	83.0	251.6	2.5	4.0	251.6 – 14750.0	0.00520 ± 0.00004	0.0026 ± 0.0003	0.9997
Ti 336.122 nm	1.5	4.6	2.4	5.1	4.6 – 14750.0	0.245 ± 0.002	0.024 ± 0.015	0.9997
V 292.401 nm	10.7	32.4	1.7	4.2	32.4 – 14750.0	0.0515 ± 0.0003	0.004 ± 0.002	0.9998
Zn 213.857 nm	12.4	37.6	1.2	3.9	37.6 – 14750.0	0.0352 ± 0.0004	0.002 ± 0.003	0.9994

[a] *± % RSD*

3. 5. Analysis of certified reference material

The trueness of the proposed method was assessed by the analysis of 5-element biodiesel certified reference material containing Ca, Mg, P, K and Na. Table 4 shows the results obtained. The certified reference material was not subjected to any treatment prior to analysis. Recovery values ranged between 95 and 106%, which mean that interferences (matrix effects) are not significant. Students's t-test was used in order to assess the null hypothesis, which establishes that the proposed method is not subject to systematic errors. The results in Table 4 show that the calculated |t| values with $n=5$ for all the analytes in the certified reference material, were always lower than the t-statistical critical value obtained from the literature ($t=2.78$) at 95% confidence level and $n-1=4$ degrees of freedom. Hence the null hypothesis is not rejected and there is no statistical evidence of systematic errors at a significance level of α = 0.05.

Table 4. Analysis of 5-element biodiesel certified reference material by ICP OES applying the proposed method.

Element Emission Line	***Certified value ($\mu g\ g^{-1}$)[a]***	***Found value ($\mu g\ g^{-1}$)[b]***	***Recovery (%)***	***Calculated |t| values***
Ca 393.366 nm	9.9 ± 0.1	9.9 ± 0.2	100	0.39
Ca 396.847 nm	9.9 ± 0.1	9.9 ± 0.2	100	0.08
K 766.491 nm	10.0 ± 0.1	9.5 ± 0.2	95	2.52
K 769.897 nm	10.0 ± 0.1	9.5 ± 0.2	95	2.49
Mg 279.553 nm	9.9 ± 0.1	9.7 ± 0.3	98	0.88
Mg 280.270 nm	9.9 ± 0.1	9.6.± 0.2	97	1.43
Na 589.592 nm	10.0 ± 0.1	9.7 ± 0.2	97	2.68
P 213.618 nm	9.9 ± 0.1	10.1 ± 0.3	102	1.41

[a] *± 1% Uncertainty*

[b] *± % RSD*

[c] *t-statistical critical value t=2.78 (n-1=4, α=0.05)*

3. 6. Analysis of real samples

All lab-made and commercial biodiesel samples analyzed in this work were directly introduced into the plasma without any sample pretreatment.

Table S1a and S1b in the supplementary material (see Appendix A) showed the results obtained from a set of 14 samples of biodiesel from different feedstock. The names of the different analyzed samples showed in Tables S1a and S1b were previously revealed in Table 1. In general, concentration values for most major and minor analytes in almost every biodiesel sample were below the LOQs except for samples S5, S13 and S14. The accuracy of the proposed method for the determination of minor elements (heavy metals) in biodiesel samples was not validated since no CRM was available for such a study. However, in view of the good accuracy obtained for the major CMR analysis, the results for the minor elements in this work could be considered very useful from the point of view of qualitative analysis.

It was expected that major elements such as Na, K, Ca and Mg, minor elements such as Ag, Al, Cu, Cd, Si, Sn, Zn, and others, were to be found in very low concentrations or in concentrations below LOQ in biodiesel samples produced from refined and non-refined vegetable oils. However, it is not unusual that such elements may be present in this kind of organic matrixes because they may come from the reagents and materials used in the transesterification process. The presence of P in biodiesel samples from vegetable oils is entirely understandable because of the phospholipids present in the oils. The Argentine Standard IRAM 6515:1 – 2006 [7], the European Standard EN 14214 [5], and the American Standard ASTM D 6751 [2], establish a maximum limit of 5 µg g^{-1} for Na plus K, and 10 µg g^{-1} for P in biodiesel, but these Standards do not establish maximum limits for Ca, Mg, and trace elements such as heavy metals. Table S1a demonstrates that only the concentration value for Na in sample S5 is not in accordance with the established maximum limits for this element. The particularly high concentration value for Na in sample S5, suggests that the removal of catalyst in the biodiesel refining step was not efficient.

4. Conclusions

The use of the new two-nozzle Flow Blurring® multinebulizer prototype has proven to be a useful tool for the analysis of phosphorus, alkaline and heavy metals in biodiesel samples obtained from different feedstock, by means of ICP OES using the on-line internal standard calibration method. The implementation of this device has allowed the simultaneous introduction of aqueous internal standard solution and organic standard/sample solutions as an effective strategy to avoid the formation of carbon deposits and to reduce spectral interferences. The consequent loss of precision and sensitivity caused by injector clogging has also been prevented. Short-term and long-term precision values ranged between 0.8 and 8.8 % RSD, this being quite acceptable. Student’s t-test permitted assessing that the proposed method does not show evidence of systematic errors for the analysis of major contaminating elements such as Na, K, Ca, Mg and P. This test has also permitted verifying that the difference between the experimental values and the true values for major contaminating elements, are not statistically significant at a 95% confidence level. The new FBMN prototype has enabled the direct introduction of organic standard solutions and biodiesel samples without any pretreatment, which makes the method proposed in this work an easy, fast, and relatively inexpensive alternative for elemental analysis of biodiesel.

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