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**DETERMINATION OF LEAD TRACES IN HONEY USING  
A FLUORIMETRIC METHOD**

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**Abstract**

Heavy traces metals may be present in honey being their detection very important for the quality control and it also serves as an indicator of environmental pollution. A new methodology for lead traces determination has been developed based on the quenching effect of the metal on fluorescent emission of 8-hydroxyquinoline and o-phenanthroline at  $\lambda_{em} = 360$  nm ( $\lambda_{exc} = 250$  nm). Experimental variables that influence on fluorimetric sensitivity were optimized by uni-variation assays. The calibration graph using zero<sup>th</sup> order regression was linear from  $0.105 \mu\text{g L}^{-1}$  to  $51.8 \mu\text{g L}^{-1}$ , with correlation coefficient better than 0.998. Under the optimal conditions, the limits of detection and quantification were of  $0.035 \mu\text{g L}^{-1}$  and  $0.105 \mu\text{g L}^{-1}$ , respectively. The trueness of the methodology was assessed through parallel samples analysis by ICP-MS. The proposed method showed good sensitivity, adequate selectivity with good tolerance to foreign

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ions, and was applied to the determination of lead trace amounts in honey from San Luis city (Argentina) with satisfactory results.

**Keywords:** Lead traces; o-Phenanthroline; 8-Hydroxyquinoline; Molecular fluorescence; Honey.

## 1. Introduction

The Argentine Food Code (Article 782, Res 2256, 16.12.85) defines the honey as “the natural sweet substance produced by honey bees from the nectar of plants or from secretions of living parts of plants or excretions of plant sucking insects on the living parts of plants, which the bees collect, transform by combining with specific substances of their own, deposit, dehydrate, store and leave in the honey comb to ripen and mature” ([http://www.anmat.gov.ar/alimentos/codigoa/Capitulo\\_X.pdf](http://www.anmat.gov.ar/alimentos/codigoa/Capitulo_X.pdf), 2018).

Nowadays, Argentina is the third honey producing country in the world. However, its domestic consumption is not so high and 95% of production is exported almost entirely in bulk (<http://supercampo.perfil.com/2018/02/argentina-en-el-podio-de-los-productores-de-miel/>, 2018). For this reason, it is vitally important to expand the controls in terms of a metal contents to ensure the product quality.

In nutrition, honey consumption is worldwide considered one of the most natural and effective remedies for the health development of all children (except in the first years of life), and a natural growth enhancer, helping to prevent diabetes mellitus, cancer, asthma, and cardiovascular, neurological, and gastrointestinal diseases (Miani & Quinto, 2017; Samarghandian, Farkhondeh, & Samini, 2017; Oduwole, Meremikwu, Oyo-Ita, Udoh, 2012). Only recently, the presence of potential unhealthy substances in honey such as heavy metals and pesticides, has been subject of studies in this food,

always considered as a cure-all substance (Ioannidou, Zachariadis, Anthemidis & Stratis 2004; Silici, Uluozlu, Tuzen & Soylak, 2016; Yarsan, et al., 2007).

During its work, honeybee may in that sense cover a wide area coming in contact not only with the air but also with soil and water of unknown purity. Hence, honey could serve as a good biomonitor of environmental pollution by the determination of certain heavy metals such as lead, cadmium, copper, nickel, among others (Madejczyk & Baralkiewicz, 2008).

Metal contamination of foodstuffs is a current issue that arouses concern in the scientific community and health professionals (Commission Regulation EC No. 1881/2006). Until 15 years ago for example, there were no specific maximum residue limit (MRL) values for lead in honey, but values of 1 mg/kg have been suggested by some European Authorities (Bogdanov, Haldimann, Luginbühl & Gallmann, 2007). In Argentine legislation (Argentine Food Code) there are no established maximum limits for this metal.

Lead has been classified for IARC as carcinogenic to humans (Group 1) (International Agency for Research on Cancer (IARC), 2009; Jaishankar, et al., 2014; Järup, 2003). This metal ion circulates in the bloodstream and accumulates in tissues and bones, or is eliminated from the body, mainly in urine (Martinello, et al., 2017). Children are particularly vulnerable and an elevated blood lead level has been associated with a reduced Intelligence Quotient score (IQ) and reduced cognitive functions (White, et. al., 2007).

Given the great evolution in instrumental methods of analysis, such as elemental inductively coupled plasma sector field mass spectrometry (Bogdanov, Haldimann, Luginbühl & Gallmann, 2007), flame atomic absorption spectrometry (Lachman, et. al., 2007), inductively coupled plasma optical emission spectroscopy ((Ioannidou, et al.,

2005; Louppis, et. al., 2017), inductively coupled plasma mass spectrometry (Chudzinska & Baralkiewicz, 2011), atomic fluorescence spectrometry (Zhou, Zhao & Xie 2011) heavy metals may be determined with a high degree of accuracy and repeatability, in a short time of analysis. However, in many cases the low levels of toxic metal are incompatible with instrumental sensitivity. Added to this, high prices and expensive instrumental inputs constitute a major constraint for many control laboratories.

The application of luminescent methods to lead traces determination has shown several analytical advantages such as high sensitivity, proper selectivity and dynamic wide range when they have been associated to separation/preconcentration/sensitization steps (Talio, et al., 2015a; Talio, 2014; Talio, et.al., 2015b).

Like traditional surfactant micelles, bile salt (BS) micelles present unique properties that can be used in chemical analysis and separation science applications; between others, BS can to solubilize selectively solutes, alter the rate and equilibrium of chemical processes, modify the effective microenvironment such as polarity, viscosity and surface tension of bound solutes (Acosta, et al., 2014). In contrast to synthetic surfactants (e.g. Triton X-110, Triton X-114, SDS), BS micellar solutions are much easier to work with since they are former and scatter light to lesser extent, which is beneficial in spectroscopic measurements (Hinze, 2000). The BS aggregation behavior is also different from that observed for synthetic surfactants. Namely, they exhibit a sequential aggregation pattern and their micelle aggregation number is typically much less than that of other micelles.

Taking into account the need to develop a simple, fast, sensitive and precise methodology for lead traces determination, the aim of present work is to propose an alternative methodology to traditional atomic spectroscopies for Pb(II) monitoring in

honey, just using one sensitization stage with the bile salt sodium cholate. Fluorimetric determination of lead is based on the formation of a ternary association complex with 8-hydroxyquinoline (8-HQ) and o-phenanthroline (o-phen) evidencing a quenching phenomenon.

## 2. Experimental

### 2.1 Reagents

Stock solutions of Pb(II)  $1 \times 10^{-4}$  mol L<sup>-1</sup> were prepared by dilution of 100 µg mL<sup>-1</sup> standard solution plasma-pure (Leeman Labs, Inc.).

Tris-(hydroxymethyl)-aminomethane (Mallinckrodt Chemical Works, St Louis, USA – 0.01 mol L<sup>-1</sup>), potassium dihydrophosphate (0.01 mol L<sup>-1</sup> - Biopack, Buenos Aires, Argentina) sodium tetraborate (Merck & Co., Inc. – 0.01 mol L<sup>-1</sup>), potassium phthalate acid (Merck & Co., Inc. - 0.01 mol L<sup>-1</sup>) and acetic acid/acetate (0.01 mol L<sup>-1</sup>- Mallinckrodt Chemical Works) buffer solutions were prepared. These solutions were adjusted to the desired pH, with aqueous HCl (Merck, Darmstadt, Germany) or NaOH (Mallinckrodt Chemical Works) using a pHmeter (Orion Expandable Ion Analyzer, Orion Research, Cambridge, MA, USA) Model EA 94.

Stock of 8-hydroxyquinoleine solution  $1 \times 10^{-6}$  mol L<sup>-1</sup> (8-HQ, H.E – Daniel Ltd., England ) and o-phenanthroline  $1 \times 10^{-6}$  mol L<sup>-1</sup> (o-phen, Merck, Darmstadt, Germany) were weekly prepared by dissolution of the appropriate amount in Millie-Q ultrapure water. The stability of solutions were checked by spectrophotometric measurements.

Surfactant solution of sodium cholate (NaC) (C<sub>24</sub>H<sub>39</sub>NaO<sub>5</sub>, Sigma Chemical Co.)  $1 \times 10^{-2}$  mol L<sup>-1</sup>, sodium dodecyl sulfate (SDS, Tokyo Kasei Industries, Tokyo Kasei Industries, Chuo-Ku, Tokyo, Japan)  $2 \times 10^{-2}$  mol L<sup>-1</sup>

and hexadecyltrimethylammonium bromide (HTAB) (Tokyo Kasei Industries)  $1 \times 10^{-2}$  mol L<sup>-1</sup> were prepared using an adequate weight of reagent and dissolving them in ultrapure water.

All solutions were prepared with ultrapure water (18M $\Omega$  cm<sup>-1</sup>) obtained from a Milli-Q EASY pure RF (Barnsted, IA, USA). All used reagent were analytical grade.

## 2.2 Apparatus

Shimadzu RF-5301 PC spectrofluorometer (Shimadzu Corporation Analytical Instrument Division, Kyoto, Japan) equipped with a 150W Xenon lamp and solid sample holder with a GF-UV35 filter were used.

Adjustments of pH were carried out using Orion Expandable Ion Analyzer pH-meter (Orion Research, MA, USA) Model EA 940 with a combined glass electrode.

An Inductively Coupled Plasma Mass Spectrometer, Perkin Elmer SCIEX, ELAN DRC-e (Thornhill, Canada) was used for validation measurements. The argon gas with minimum purity of 99.996% was supplied by Air Liquide (Córdoba, Argentina). An HF-resistant and high performance Teflon Nebulizer model PFA-ST, was coupled to a quartz cyclonic spray chamber with internal baffle and drain line cooled with the PC3 system from ESI (Omaha, NE, USA) (**Table 4**). Tygon black/black 0.76 mm i.d. and 40 cm length peristaltic pump tubing was used. Auto lens mode on, peak hopping measure, dwell time of 50 ms, 15 sweeps/reading, 1 reading/replicate, and 6 replicates were used as instrument conditions (Moyano, et al., 2014).

## 2.3 Sampling procedure and samples treatments

This cross sectional study was performed in San Luis, a province in the central region of Argentina, on 2018 honey production. A total of five samples of multi floral

and one monofloral honey were collected from individual beekeepers of regions. All samples were collected in clean and closed recipients and immediately transferred to the laboratory and kept at 4–8 °C in dark place until analysis.

Approximately 0.1 g of each sample were weighed and they were diluted taking a final volume of 100 mL.

#### 2.4 General procedure

An aliquot 0.050  $\mu\text{L}$  of sample, Pb(II) aliquot (1.85 and 3.91), 100  $\mu\text{L}$  buffer phosphate  $1 \times 10^{-3} \text{ mol L}^{-1}$  (pH = 6.0), 500  $\mu\text{L}$  o-phen ( $1 \times 10^{-7} \text{ mol L}^{-1}$ ), 750  $\mu\text{L}$  8-HQ ( $1 \times 10^{-7} \text{ mol L}^{-1}$ ) and 0.050  $\mu\text{L}$  sodium cholate (NaC) ( $1 \times 10^{-4} \text{ mol L}^{-1}$ ), were placed in a 10 mL glass volumetric flask. The whole mixture was diluted to 10 mL with Millie-Q ultrapure water. Concentration Pb(II) was determined by fluorescent emission at 360 nm using  $\lambda_{\text{exc}} = 250 \text{ nm}$ , employed a conventional quartz cell.

#### 2.5 Accuracy study

Adequate volume of honey solutions were spiked with increasing amounts of Pb(II) (1.85 and 3.91  $\mu\text{g L}^{-1}$ ). Pb(II) concentrations were determined by the proposed methodology.

#### 2.6 Precision study

The repeatability (within-day precision) of the method was tested for replicate of samples ( $n = 4$ ) spiked with 3.91  $\mu\text{g L}^{-1}$  of Pb(II) and the contents were determined by proposed methodology.

#### 2.7 Trueness



Pb(II) contents in the matrix of tobacco samples was determined by ICP-MS, using operational conditions previously consigned in apparatus item.

### 3. Results and discussion

#### 3.1. Fluorescent Spectra

Previous investigations have evidenced the feasibility of the metal quantification by spectrofluorimetry based on the formation of ternary complexes between metal ions and different chelating reagents (Talio, Luconi, Masi, Fernández, 2009; Kelani, Bebawy & Abdel-Fattah, 1999). For this reason, the studies were initiated looking for possible chelating agents for Pb(II) that fulfilled the requirements of presenting fluorescence and evidencing a response in the presence of Pb(II) traces.

The fluorophore 1,10-phenanthroline (o-phen) is the parent of an important group of chelating reagents, presenting attracted special interest from researchers due to their various structural and chemical properties as rigidity, planarity, aromaticity, basicity, and chelating capability. In this context, systems containing o-phen ligands as key components are emerging as luminescent sensors for the efficient detection of metal ions (Bonacorso, et. al.2016; Villar-Garcia, Abebe & Chebude, 2012).

Likewise, 8-hydroxyquinoline (8-HQ) is one of the most popular and versatile organic compound. 8-HQ and its derivatives have found a great variety of applications in organic light-emitting diodes and fluorescent chemosensors for metal ions (Sugawara, Weetall & Schucker, 1974). 8-HQ ligand has been widely applied for analytical purposes and separation techniques (Al-Busafi, Suliman & Al-Alawi, 2014).

At the beginning of the experimental part, the fluorescence of 8-HQ/Pb (II) and o-phen/Pb (II) systems in aqueous medium were separately explored with promising results. However, with the aim of evaluating the feasibility of a ternary complex

formation, assays were carried out using 8-HQ, o-phen and Pb(II). In this condition, an important improvement in the sensitivity was observed. Additionally, the ternary association formed in aqueous solution showed stability over time evidencing a quenching effect as the concentration of Pb(II) increased (**Figure 1**).

### 3.2 Optimization of Experimental Conditions

#### 3.2.1 pH optimization

In order to find the optimum conditions for the formation of Pb(II)/8-HQ/o-phen association, experimental parameters such as pH, nature and concentration of buffer solution, concentrations of chelating reagents, solvent nature that affecting the complexation reaction and its spectral behavior were consecutively investigated.

The first parameter examined was the pH; this parameter plays a unique role on metal-chelate formation and the subsequent metal determination. The results illustrated in **Figure 2**, showed that at pH near 6.0 a maximum quenching effect was obtained. Due to this behavior, the pH value of 6.0 was selected as working value for the following experiences.

The effect of different buffers on the quenching effect of Pb(II)/8-HQ/o-phen ternary system was tested. Among the assayed buffer solutions, the best sensitivity was obtained when phosphate buffer was used. Coming up next, the fluorescence response was studied for ternary systems prepared with potassium dihydrophosphate from  $1 \times 10^{-5}$  to  $1 \times 10^{-2}$  mol L<sup>-1</sup>. The best performance and optimal stability in the time was achieved for a buffer concentration  $1.0 \times 10^{-4}$  mol L<sup>-1</sup> and it was chosen as optimal.

#### 3.2.2 Nature and concentration of surfactant agent

It is well known that the use of surfactants in molecular fluorescence provides some advantages that improve the determination of the analyte under study. In this way, micellar media have shown to be effective to minimize intermolecular interactions between the analyte and sample matrix constituents. Additionally, the photophysical properties of the fluorescent solutes can be altered in the micellar medium improving the fluorescent sensitivity.

After the optimal conditions for the chemical interaction between Pb(II) and 8-HQ/o-phen were established, the influence of different surfactants on the quenching effect was tested. The results showed a relationship between the nature of the surfactant and the fluorescent emission of Pb(II)/8-HQ/o-phen system, as follows: for systems with anionic surfactants (SDS and NaC) in a concentration of  $5 \times 10^{-7}$  mol L<sup>-1</sup>, a quenching effect was observed. In the case of NaC, an improvement in the intensity of fluorescent quenching was observed respect to SDS. On the other hand, for systems with cationic surfactant (HTAB) in a concentration of  $5 \times 10^{-7}$  mol L<sup>-1</sup>, no variation in the quenching intensity was observed.

According to the results obtained, bile salt NaC was selected as the optimal condition due to maximum quenching effect showed for Pb(II)/8-HQ/o-phen system.

### *3.2.3 Optimization of fluorophors concentration*

The effect of 8-HQ and o-phen concentrations on the fluorescent quenching of Pb(II) was subsequently studied and the results are presented in Figure 3. With these results, concentrations of 8-HQ of  $7.5 \times 10^{-9}$  mol L<sup>-1</sup> and o-phen of  $5 \times 10^{-9}$  mol L<sup>-1</sup> were chosen as optimal for following assays.

### *3.3 Analytical parameters*

### 3.3.1 Calibration curve and analytical figures of merit

Calibration plots for concentration levels of Pb(II) from 0.105  $\mu\text{g L}^{-1}$  to 51.8  $\mu\text{g L}^{-1}$  was assayed applying the developed methodology. The limits of detection (LOD) and quantification (LOQ) were calculated in accordance to the formulas given by the official compendia methods (Gupta & Shrivastava, 2011), using the relation  $k(\text{SD})/m$  where  $k = 3.3$  for LOD and 10 for LOQ. SD represents the standard deviation from 15 replicate blank responses and  $m$  is the slope of the calibration curve (**Table 3**).

The range of linearity was evaluated by checking the linear regression coefficient ( $R^2 > 0.998$ ) of the calibration curve.

The reproducibility of the method was evaluated repeating 4 times the proposed approach for each addition in a total of two level of spiked Pb(II) for each sample. The recoveries of Pb(II) based on the average of four replicate measurements are illustrated in **Table 2**. Obtained results showed satisfactory agreement with adequate precision (%CV).

### 3.3.2 Quenching mechanism

Under optimized experimental conditions, the luminescence quenching caused by Pb(II) was adequately described by the Stern-Volmer equation:

$$I_0 / I = K_{sv} (Q) + C$$

where  $I_0$  and  $I$  are the luminescence intensity of 8-HQ/o-phen fluorophores in the absence and presence of the quencher (Pb(II)), respectively;  $K_{sv}$  is the Stern–Volmer constant representing the affinity between 8-HQ/o-phen system and Pb(II);  $Q$  is the Pb(II) concentration; and  $C$  is a constant near to 1. Under optimal experimental conditions, the obtained  $K_{sv}$  for Pb(II)-8-HQ/o-phen system is  $1.53 \times 10^8 \text{ M}^{-1}$ .

To determine the nature of the quenching in the system under study, fluorescent emissions were determined at different temperatures (25, 30 and 40 °C) for 8-HQ/o-phen in presence of Pb(II) increasing concentrations. As  $K_{SV}$  increased with the temperature, it could be concluded that a collisional quenching is occurring for Pb(II)/8-HQ/o-phen system (**Figure 4**).

In the following experiments, the working temperature was kept to 25 °C.

### 3.4 Applications

To establish the applicability of proposed method, it was applied to Pb(II) quantification in samples of honey commercialized in San Luis city (Argentina). The results obtained showed an adequate precision (see **Table 2**).

Robustness of the assay was evaluated by repeating the analysis four times for each sample. **Table 2** shows the recovery results achieved. The obtained results indicate that the proposed method is suitable for Pb(II) determination in all studied samples.

## 4. Conclusions

Although atomic spectroscopies are the most important techniques for metal traces analysis, they require specialized and very expensive instruments. The continued importance and employment in work areas of lead and its toxicity in environmental health has demanded the development of simple and rapid analytical methods for its determination. The proposed methodology represent a promising approach for Pb(II) monitoring with low operation cost, simplicity of instrumentation and non-polluting solvents. Samples of honey produced and commercialized in San Luis (Argentina) were successfully analyzed with an average recovery close to 100%. The method was validated by ICP-MS and was applied at real samples with good tolerance to regular

foreign constituents. The presence of heavy metals in honey is an indicator of environmental pollution. Sufficient care must be taken to regulate the amount of heavy materials entering the environment as exposure to these elements can adversely affect human health.

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**Figure captions**

**Figure 1:** Fluorescence spectra of Pb(II)/8-HQ/o-phen systems.

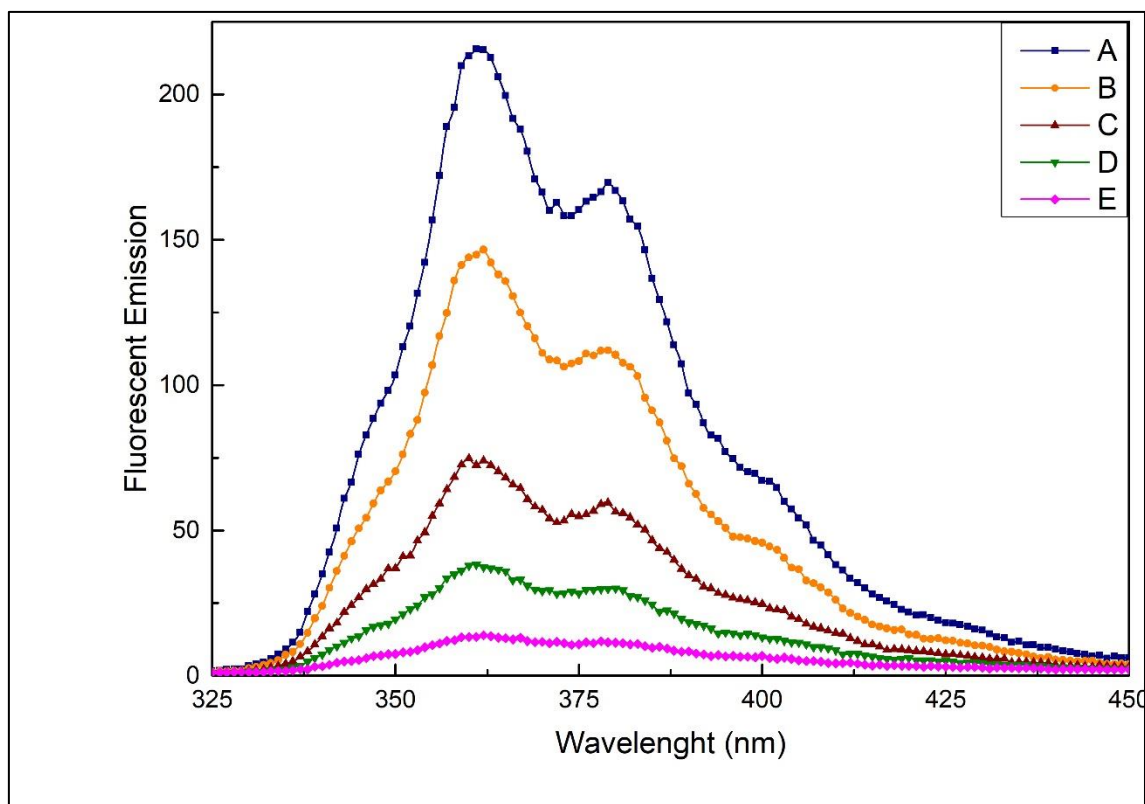
**Figure 2:** Influence of pH on Pb(II)/8-HQ/o-phen.

**Figure 3:** Effect of 8-HQ and o-phen concentrations on the Pb(II)/8-HQ/o-phen fluorescent signal.

**Figure 4:** Stern–Volmer plot for Pb(II)/8-HQ/o-phen systems.

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Figure 1



A: 8-HQ/o-phen system.

B: Idem A with Pb(II)  $1.85 \mu\text{g L}^{-1}$

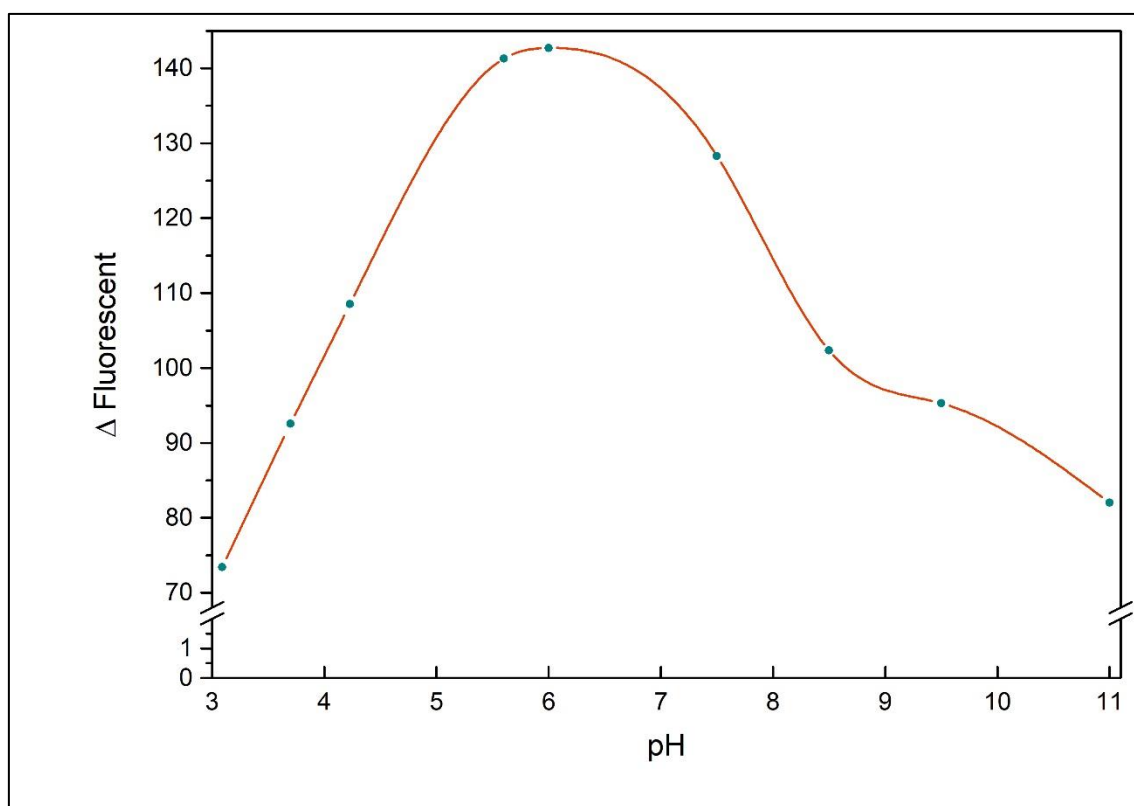
C: Idem A with Pb(II)  $3.91 \mu\text{g L}^{-1}$

D: Idem A with Pb(II)  $5.97 \mu\text{g L}^{-1}$

E: Idem A with Pb(II)  $6.83 \mu\text{g L}^{-1}$

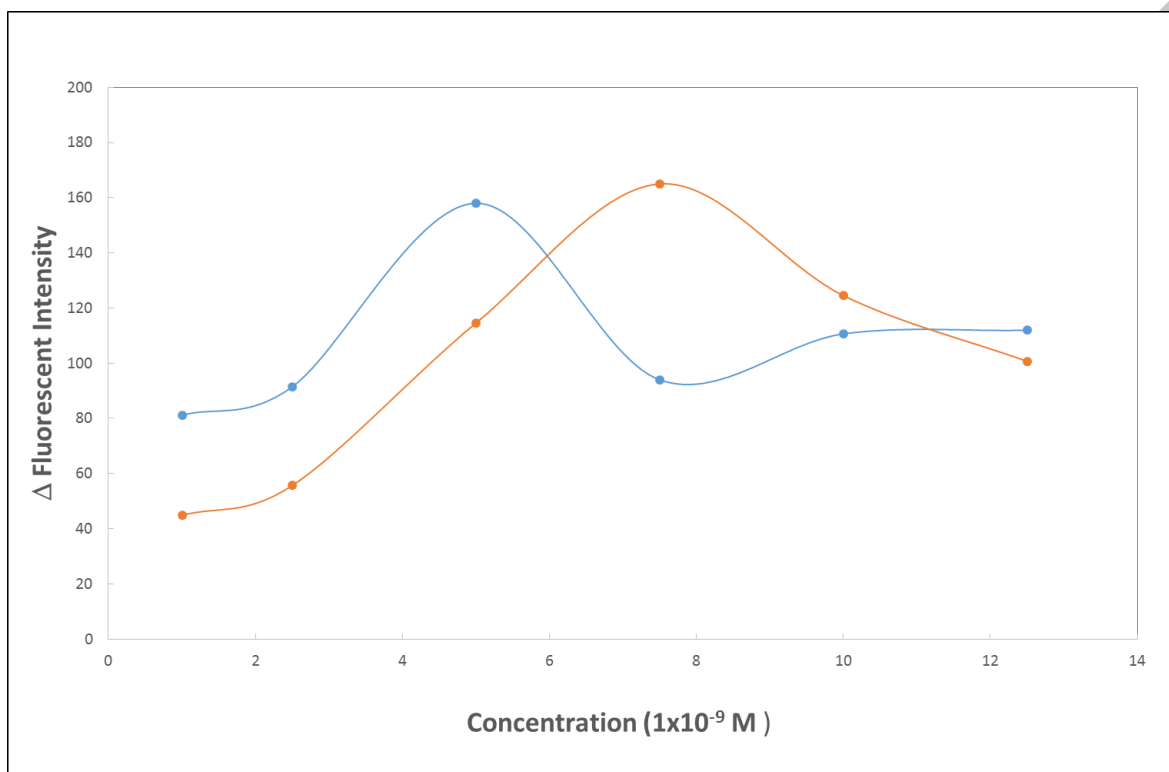
Conditions:  $\lambda_{\text{em}} = 360 \text{ nm}$ ;  $\lambda_{\text{exc}} = 250 \text{ nm}$ ;  $C_{8\text{-HQ}} = 7.5 \times 10^{-8} \text{ mol L}^{-1}$ ,  $C_{\text{o-phen}} = 5 \times 10^{-5} \text{ mol L}^{-1}$ ;  $C_{\text{NaC}} = 5 \times 10^{-7} \text{ mol L}^{-1}$ ;  $C_{\text{buffer phosphate}} = 1 \times 10^{-4} \text{ mol L}^{-1}$ , pH 6.0. Other experimental conditions are described under procedure.

Figure 2



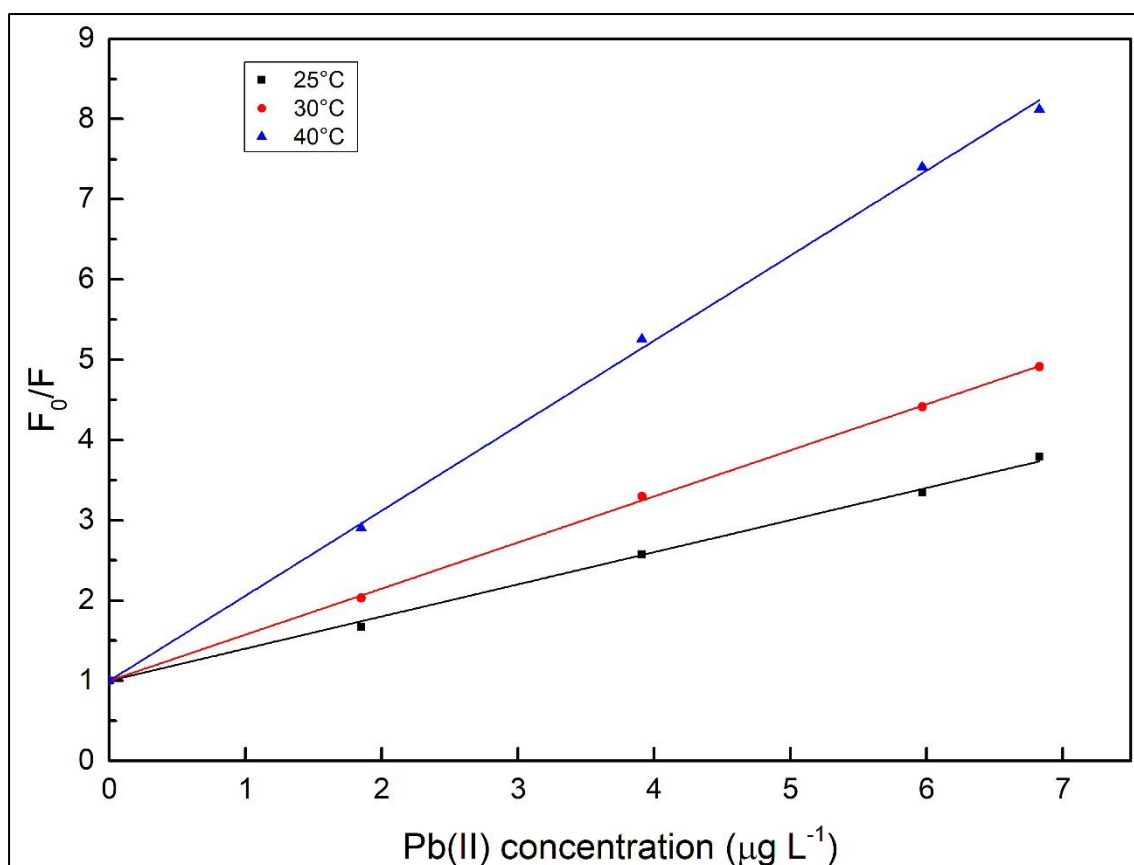
Conditions:  $\lambda_{em} = 360$  nm;  $\lambda_{exc} = 250$  nm;  $C_{8-HQ} = 7.5 \times 10^{-8}$  mol L $^{-1}$ ,  $C_{o-phen} = 5 \times 10^{-5}$  mol L $^{-1}$ ;  $C_{NaC} = 5 \times 10^{-7}$  mol L $^{-1}$ ;  $C_{(Pb(II))} = 3.91$   $\mu$ g L $^{-1}$ ;  $C_{buffer\ phosphate} = 1 \times 10^{-4}$  mol L $^{-1}$ , pH 6.0. Other experimental conditions are described under procedure

Figure 3:



Conditions:  $\lambda_{em} = 360$  nm;  $\lambda_{exc} = 250$  nm;  $C_{(Pb(II))} = 3.91 \mu\text{g L}^{-1}$ ;  $C_{NaC} = 5 \times 10^{-7}$  mol  $\text{L}^{-1}$ ;  $C_{\text{buffer phosphate}} = 1 \times 10^{-4}$  mol  $\text{L}^{-1}$ , pH 6.0. Other experimental conditions are described under procedure.

Figure 4:



Conditions:  $\lambda_{\text{em}} = 360 \text{ nm}$ ;  $\lambda_{\text{exc}} = 250 \text{ nm}$ ;  $C_{8\text{-HQ}} = 7.5 \times 10^{-8} \text{ mol L}^{-1}$ ;  $C_{\text{o-phen}} = 5 \times 10^{-5} \text{ mol L}^{-1}$ ;  $C_{\text{NaAc}} = 5 \times 10^{-7} \text{ mol L}^{-1}$ ;  $C_{\text{buffer phosphate}} = 1 \times 10^{-4} \text{ mol L}^{-1}$ , pH 6.0. Other experimental conditions are described under procedure.

**Table 1:** Analytical parameters of methodologies for Pb(II) determination in honey samples.

Method	Comments	Reference
ICP-AES	RSD = 6.2 %. LOD = 6.7 $\mu\text{g/L}$ Slop $\pm$ SD = 6.1 $\pm$ 1.4 (0.9770)	Ioannidou, et al., 2005
ICP-MS	Range lead determinate=0.05 and 200 $\mu\text{g L}^{-1}$ Recovery (%) = 99.0 RSD =0.9–4.3%	Chudzinska & Baralkiewicz, 2011
AFS	LOD = 0.95 $\text{ng L}^{-1}$ Linear range of 0.01–100 $\text{ng mL}^{-1}$ $r^2 = 0.9990$ Recovery (%) = 92.9–97.4	Zhou, Zhao & Xie, 2011
Spectrophotometric associate with silver nanoparticle	Linear range= 0.0500 - 0.167 $\text{mg L}^{-1}$ $R^2=0.994$ LOD = 0.0135 $\text{mg L}^{-1}$ LOQ = 0.0451 $\text{mg L}^{-1}$	Borges Bittar, et al., 2018
GFAAS	Recovery (%) = 85 and 115 LOD = 0.6 $\mu\text{g L}^{-1}$	Ajtony et al., 2007
This method	LOD = 0.035 $\mu\text{g L}^{-1}$ LOQ = 0.105 $\mu\text{g L}^{-1}$ Linear range= 0.105 – 51.8 $\mu\text{g L}^{-1}$ $R^2 = 0.998$ Recovery (%) = 96.33–104.09	-



**Table 2:** Recovery studies by lead determination in honey commercialized in city San Luis.

Sample	Pb(II) added ( $\mu\text{g L}^{-1}$ )	Proposed methodology			ICP-MS	%RE*
		Pb(II) found $\pm$ CV ( $\mu\text{g L}^{-1}$ )	Recovery (% , n=4)	Pb(II) found ( $\mu\text{g g}^{-1}$ )	Pb(II) found $\pm$ SD ( $\mu\text{g L}^{-1}$ )	
1	-	0.122 $\pm$ 0.01	-	1.238	0.132	7.75
	1.85	1.977 $\pm$ 0.09	104.09			
	3.91	4.030 $\pm$ 0.06	98.36			
2	-	0.171 $\pm$ 0.03	-	1.779	0.166	2.92
	1.85	2.018 $\pm$ 0.04	98.24			
	3.91	4.085 $\pm$ 0.02	102.33			
3	-	0.213 $\pm$ 0.08	-	1.790	0.201	5.63
	1.85	2.060 $\pm$ 0.05	98.59			
	3.91	4.128 $\pm$ 0.08	102.35			
4	-	0.249 $\pm$ 0.04	-	2.530	0.270	7.77
	1.85	2.104 $\pm$ 0.02	102.00			
	3.91	4.153 $\pm$ 0.01	97.69			
5	-	0.109 $\pm$ 0.04	-	1.121	ND	-
	1.85	1.955 $\pm$ 0.02	96.33			
	3.91	4.996 $\pm$ 0.01	99.63			

\*%RE=  $100 \times (\text{measured value} - \text{actual value})/\text{actual value}$ .

**1:** Organic honey A (Multiflora: chañar, piquilin, white carob, and yellow stick).

**2:** Organic honey B (Multiflora: melilotus, chilca, usillo and jarilla).

**3:** Multiflora I: not specified.

**4:** Multiflora II: not specified.

**5:** Monoflora: carob tree.

**Highlights**

- New spectrofluorimetric methodology for lead traces determination is proposed.
- The possible mechanism of quenching is proposed.
- The reached sensitivity was comparable at those of atomic spectroscopies.
- This methodology showing good reproducibility with low operation cost.
- It was applied to the determination of trace amounts of lead in honey from San Luis city (Argentina).

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**Declaration of Interest Statement**

The authors declare no conflicts of interest.

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