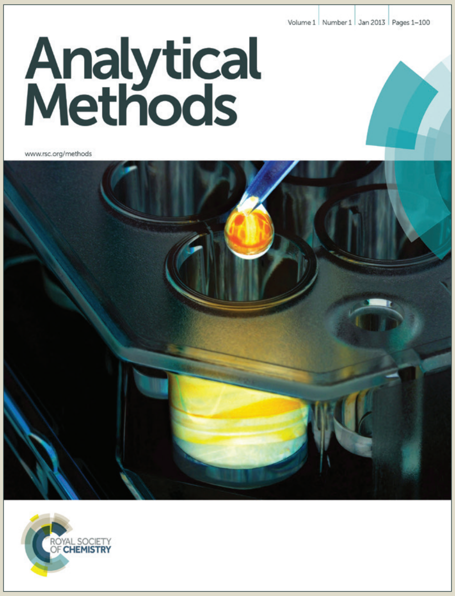
Analytical
Methods



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Speciation of copper in agricultural soils contaminated by lead using screen-printed electrodes and Square-Wave Anodic Stripping Voltammetry (SPE-SWASV)

Stéphane Faucher, Cyril Cugnet, Laurent Authier, Gaëtane Lespes

Université de Pau et des Pays de l'Adour
Laboratoire de Chimie Analytique Bio-Inorganique et Environnement (LCABIE),
UMR 5254 IPREM
2 Avenue Pierre Angot, 64053 Pau Cedex 9, France

Corresponding author. Tel.: +33 540 17 50 26; fax: +33 559 40 76 74
E-mail address: cyril.cugnet@univ-pau.fr (C. CUGNET)

Abstract

An analytical strategy, involving carbon screen printed working electrodes (SPE) modified with mercury combined with Square Wave Anodic Stripping Voltammetry (SWASV) was optimized for total and electrolabile copper concentration determination in presence of lead. The accuracy of the method was checked by analyzing a solution of mineralized certified reference soil, the mineralization step allowing the quantity of metals to be assumed totally under electrolabile form in the solution of mineralized soil. The influence of the calibration modes and potential matrix effects were investigated. In optimized conditions, the accuracy (i.e. trueness and repeatability) and robustness were verified. For that, the electrolabile copper concentration and the Relative Standard Deviation (RSD) were evaluated according to the change frequency of the SPE and the solution analyzed after each SPE-SWASV measurement in the solution of the mineralized soil. Additionally, analyses were performed by Inductively Coupled Plasma Mass Spectrometry (ICP-MS) as a confirmation mean. From statistical tests in a 95 % confidence interval, a copper recovery of 97 % (regarding certified value) and a RSD of 9 % were found for the electrochemical method. Agricultural soils often containing lead in significant concentration, analyses were also carried out on lead. This allowed verifying there was no interaction between copper and lead on the measurement. Then, the developed method was applied for the first time in an original strategy consisting in the quantification of the total and the electrolabile amounts of copper and lead in agricultural soils in order to estimate the potential hazard brought by their mobilization from the soil. For that, soils were (i) mineralized to obtain the total metal amounts and (ii) leached to reach the electrolabile ones.

Keywords · Copper · Screen-printed electrodes (SPE) · Square Wave Anodic Stripping Voltammetry (SWASV) · Electrolabile · Agricultural soil · Leachate

Introduction

The metal contamination of agricultural soils may lead to hazard to fauna and flora [1–3]. The first source of this contamination is the soil due to the weathering of the parent rock. Moreover, the use of fertilizers and pesticides presents a major source of the anthropogenic contamination as well as industrial and municipal activities, sewage sludge and gasoline [4–8]. Viticulture represents an important agricultural practice in many countries as France [8]. The extensive application of specific pesticides, such as the Bordeaux mixture ($CuSO_4$ + $Ca(OH)_2$) or the arsenate lead ($PbHAsO_4$) resulted in increasing copper and lead concentrations especially in vineyard soils [7–13]. Copper and lead concentrations in soils vary from 5 to 500 mg kg^{-1} and from 1 to 200 mg kg^{-1} respectively, according to their level of contamination directly linked to anthropogenic activities [14–16]. It is now well known that the hazard of metals to fauna and flora is not directly linked to the total metal concentrations in soil [1,17–19], even if this last one is a necessary prerequisite in a speciation strategy [9,20–25]. Concerning total analyses, electrochemical techniques, Inductively Coupled Plasma based spectrometry (Inductively Coupled Plasma Mass Spectrometry (ICP-MS) and Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES)) have been used after solid sample mineralization. Nowadays, atomic spectrometry is the technique of choice due to its multi-elemental capabilities and ICP-MS is preferred for its high sensitivity. Regarding electrochemical techniques, the stripping methods based on metal preconcentration onto a modified solid working electrode at a fixed deposition potential are preferred for metal determination [26,27]. The advantages of this electrochemical method are its sensitivity, its capacity to make multi-elemental speciation analyses, the low cost and the suitability to online measurements [28]. Square Wave Anodic Stripping Voltammetry (SWASV) is mainly used during the stripping step because it enhances the sensitivity of the analytical signal [29]. However, electrochemical methods employ generally a hanging mercury electrode with a glass capillary. Thus, the solution of mineralized sample damages the glass capillary, which leads to inaccuracy because of the etching of the capillary caused by the hydrofluorhydric acid (HF) contained in the solution [30]. Nevertheless, stripping analyses were lately performed by using modified carbon screen-printed working electrodes (SPE) [31]. These SPE have the advantages to be easily and rapidly produced at the laboratory, to be cheap, to favor repeatability of measurements and their geometrical shape is adapted as the analytical constraints [32–34]. The active carbon surface of the working SPE is usually coated by a metal in order to reach low limit of detection [35,36]. SPE are generally modified by mercury or bismuth, which allows preconcentrating analytes on the SPE [37]. The bismuth is more “environmental friendly” but less sensitive than mercury. SPE modified by bismuth allows generally determining electrolabile metals in acetate buffer solutions [35,38]. However, the passivation of bismuth at neutral pH disturbs the analysis quality in natural samples. The use of SPE modified by mercury offers better analytical performances, authorizes a wide pH range and low detection limits for relatively short analyses times and mercury deposition [39]. Moreover, the SPE being devoid of glass, they are more robust than classical hanging mercury electrodes regarding the present application employing a solution of HF-based mineralized soil.

Concerning speciation analyses, SWASV electrochemical method can be used to determine electrolabile forms [40–42]. This method needs no treatment of the soil solution (leachate), which is not the case with other analytical techniques [43].

Considering all this, the strategy involving only SPE-SWASV for total and electrolabile metal determination appears interesting especially in the perspective of evaluating the hazard of metal transfer from the soil to living organisms after mobilization in aqueous phase and diffusion [23]. There are few papers relating SWASV analyses applied to total and electrolabile metal concentrations in complex samples [30,31,44] : Truzzi et al. [30] have reported Cd, Cu and Pb in a hydrofluoridric acid solution of siliceous spicules of marine sponges and Meucci et al. [31] Cd, Cu, Pb and Hg determination in fish by SWASV. However, to our knowledge, no study related to total and electrolabile concentrations determined using the same technique has been reported for soil. Moreover, few studies have been made on SWASV calibration modes and matrix influence on measurements. However, the calibration strategy and matrix effects are essential parameters to understand and control in order to ensure the analytical accuracy and reproducibility. The aim of the study was to investigate an electrochemical-based strategy in order to evaluate its potentialities in determining metal amount and assessing the potential hazard from the metal contamination of soils. The strategy used was to quantify preliminary the quantity of copper and lead in different types of agricultural soils. For that, the total copper and lead were preliminary determined in agricultural soils. Then, copper and lead which can be mobilized from these soils were obtained by leaching and the corresponding electrolabile amounts were determined. SPE-SWASV was the technique of the choice for total and electrolabile copper determination. The method was firstly optimized using a solution of mineralized certified reference soil. The mineralization step allows assuming that all the metal contained in the soil was under electrolabile form in the solution of mineralized soil. Then, copper analyses were performed by SPE-SWASV using different calibration modes to determine the conditions that allow assessing the reference copper value in presence of lead. Additionally, the influence of pH and conductivity was studied, and interferences due to high concentrations of lead considered on the determination of electrolabile copper in soil.

Experimental

Chemicals

Standard solutions of copper at 1000 mg L^{-1} and lead at 1000 mg L^{-1} (purity 99 %, Sigma-Aldrich) were used to carry out the calibrations. A standard solution of indium at 1000 mg L^{-1} was used as internal standard for ICP-MS analyses. A mixture of nitric acid HNO_3 (70 %, Atlantic laboratory, Bruges, Belgium), hydrofluoric acid HF (60%, Atlantic laboratory, Bruges, Belgium) and hydrogen peroxide H_2O_2 (30 %, Atlantic laboratory, Bruges, Belgium) was used for the mineralization step. Boric acid HBO_2 (99.99 %, Prolabo, Paris, France) was used to neutralize HF. Sodium hydroxide monohydrate $NaOH,H_2O$ (99.9995 %, Sigma-Aldrich) and acetic acid $C_2H_4O_2$ (99 %, Fluka, USA) were used to prepare 0.2 mol L^{-1} buffer acetate. Leachings were carried out with ultra pure water exit of Milli-Q System (18MΩ.cm). The conductivity of the leachates was increased by potassium nitrate KNO_3 addition (99.995 %, Merck, France). The electrodes were screen-printed by a commercial carbon solution (Electrodag PF 407A) purchased from Acheson Colloïds and High-Impact PolyStyrene (HIPS) from Sericol. The working electrodes were modified using mercury standard acetate solution purchased from Sigma-Aldrich.

Soil samples

The certified reference soil used for optimization step was the *loam soil ERM-CC141* (Institute for Reference Materials and Measurements, Retieseweg, B-2440, Geel, Belgium). Concerning the environmental applications, five agricultural soils (named A, B, C, D and E) were sampled in 2012 at 0-30 cm depth interval in two farms, on plots where wheat and sunflower (for soils A and B respectively, in Auzeville-Tolosane, France) and vineyards (for soils C, D and E, in Bordeaux, France) were cultivated. Then, they were air-dried and they were ground to pass throw a 2 mm sieve. The main physico-chemical characteristics of these soils are summarized in Table 1.

The physico-chemical characterisation of soils has been performed prior to any analysis of ETM. It has been carried out by the Laboratory of soil analyzes of Arras (LAS of Arras) in accordance with the French standardized methods (AFNOR, 1999; ISO, 1999).

Sensor

Carbon screen-printed electrodes (SPE) were developed at the laboratory, their preparation being described elsewhere [34]. Mercury electrodeposition on sensor was obtained from 0.2 mol L^{-1} acetate buffer (pH 4.5) solution containing 170 mg L^{-1} of mercury. After 5 successive cyclic voltammetries from -0.1 V to 0.8 V, 5 µg of mercury, corresponding to a plating procedure charge (*Qplating*) of -5 mC, were electrodeposited at a potential of -0.1 V in stirred and non deaerated solution [34].

Apparatus

SWASV analyses were achieved by a potentiostat Autolab PGSTAT12 (Eco Chemie, The Netherlands) controlled by a personal computer equipped with software GPES 4.9. The measuring cell consists in a mercury modified screen-printed electrode as working electrode, a carbon screen-printed electrode as auxiliary electrode, and an Ag/AgCl/KCl 3 mol L^{-1} reference electrode (Model DRIREF-5SH, World Precision Instruments Ltd). A rotary agitator (STR4 STUART) was used to carry out the leaching of soils. Soil mineralizations were carried out with concentrated nitric acid, hydrofluoric acid and hydrogen peroxide in closed system microwave (Ethos touch control, Milestone GMBH, Shelton, the USA).

Preparation of the solution of mineralized soil for total copper and lead determination in soil

A mass of 0.250 g of soil was mineralized by microwave with 6 mL of nitric acid HNO_3, 2 mL of hydrofluoric acid HF and 2 mL of hydrogen peroxide H_2O_2. A blank solution was mineralized by microwave with the same mixture in the same proportion but without soil addition. The mineralization procedure was in two steps: the first one consisted in increasing the temperature linearly from 20 °C to 200 °C for 20 minutes with a power of 1000 W and the second in stabilizing the temperature to 200 °C for 20 minutes [45].

For SPE-SWASV analyses, the solution of mineralized soil and the solution of mineralized blank were recovered and diluted 15 times in acetate buffer to decrease metal and acid concentrations. Boric acid was added to neutralize the hydrofluoric acid [2]. Then, the acidic solutions were diluted and filtered at 0.45 µm before analysis. Later on in this paper, these solutions are named MS for the diluted solution of the mineralized soil and MB for the diluted solution of mineralized blank respectively.

The mineralization step allowed assuming that all the copper and lead presents in the MS were in electrolabile form. Thus, the values obtained by SPE-SWASV analysis must correspond to the reference values.
For ICP-MS analyses, after the step of soil mineralization just described, the solution of mineralized soil was diluted 50 times and filtered at 0.45µm.

Leaching procedure

The leaching procedure follows the Standard NF ISO 18772. For each soil sample, 8 grams of dry soil were introduced into a 100 mL centrifugation tube in polypropylene beforehand cleaned with nitric acid (10 %), rinsed with MilliQ water and dried. Then, 80 grams of MilliQ water (for a liquid to solid ratio (L/S) equal to 10) were added as leaching solution. The tubes containing samples were fixed on the rotary agitator and shaken for 24 hours with a speed of 10 turns per minute in a thermostated room at 20°C. Once agitation ended, the tubes were left elutriated for 30 minutes and the samples were centrifuged for 30 minutes at 3500 rpm. Finally, the supernatant was recovered with a pipette, filtered at 0.45 µm and stored at 4 °C until analysis.

Voltammetric analysis procedure

Concerning SPE-SWASV analyses, copper and lead were reduced at a potential of - 0.8 V during 60 seconds under stirring. After 30 seconds of equilibration time, an anodic square wave potential scan from - 0.8 V to + 0.1 V was applied and the stripping voltammogram was recorded. The surface of peak was retained as the analytical answer. A volume of 10 ml of sample were used for each measurement. Each analysis was replicated 3 times in order to evaluate the repeatability and the accuracy of the measurement.
Concerning ICP-MS analyses, copper and lead were determined by standard addition calibration after indium addition as internal standard. The ICP-MS used was an Agilent 7500ce model equipped with a Meinhard nebulizer (concentric), a Scott spray chamber cooled to 2°C and an octopole collision/reaction cell. Because of the presence of hydrofluoric acid in analyzed solutions, the torch and spray chamber were in Teflon-perfluoroalkoxy (PFA). The parameters employed were as follow: Plasma gas flow rate (argon), 1.15 L min^{-1}; collision/reaction cell gas, H_2 3 mL min^{-1}; radio frequency power, 1500W; nickel sampler and skimmer cones; dwell time, 0.1 s. These parameters were optimized using a solution of 1 µg L^{-1} Li, Y, Tl, Ce in 2 wt% HNO_3. The following isotopes were monitored: ^{63}Cu, ^{65}Cu, ^{115}In, ^{204}Pb, ^{206}Pb, ^{208}Pb.

Results and discussion

Optimization of the SPE-SWASV method

SPE measurement conditions

Modified SPE were designed especially as disposable sensors, with all the benefits that come from the concept of an easily renewable sensor. In particular, the possibility to frequently change the sensor allows reducing the risk of contamination from one analysis to another. The frequency of SPE replacement depends on the metal studied and on the metal deposed onto the SPE to modify its surface (mercury in the present case). Indeed, if the oxidation potentials of these two metals are close, there are two possibilities. If the oxidation potential set during the oxidation procedure is (i) lower than the metal ones, the metal studied concentration would be underestimated because copper won’t be totally released from the SPE to the solution. Moreover, next analyses would be falsified because a part of metal studied would be still amalgamated with mercury onto the SPE or (ii) higher than the metal ones, a part of mercury, previously electrodeposited onto the surface of the SPE, is equally released (with the entirety of metal studied) from the SPE to the solution during the oxidation procedure. In this case, the SPE would contain less mercury for the next analyses. Thus, SPE would be modified after the analysis. Additionally, the analytical medium, i.e. the medium where the measurement is performed, has also to be considered since the SPE performances depend on it.

According to these different considerations, the first point checked was the frequency of the SPE replacement, additionally to how to use the SPE, i.e. if the solution (analytical medium and sample) had to be also changed. The most relevant response for this is the evaluation of the measurement repeatability for the copper concentration determination. Thus, the Relative Standard Deviation (RSD) was estimated from three copper measurements performed in different conditions summarized in the left part of Table 2. The analytical medium first used was acetate buffer, considered as a medium of reference since it is usually used to ensure satisfactory electrochemical conditions [46].

The use of the same solution and the same SPE for three analyses leads to a RSD of 20 % (Table 2, entry 1). This not satisfactory repeatability comes from the increase of the copper peak surfaces with measurement replication. This phenomenon is due to the proximity of the oxidation potentials of copper and mercury, which generates a partial release of copper in solution and its accumulation on the electrode. This copper is then released during the next replicated analysis. An alternative to try to avoid this accumulation is to increase the higher limit of sweeping (oxidation potential). However, if the totality of the copper amalgamated with mercury is well released in these conditions, a part of mercury is also released and contaminates the solution. Measurements were then performed changing the SPE (Table 2, entry 2) or renewing the solution (Table 2, entry 3) for each analysis. However RSD still remains high, which seems to show that the accumulation of copper onto the electrode is not the only phenomenon occurring. Indeed, when the solution is renewed and the SPE is not changed (Table 2, entry 3), the copper peak decreases because of the loss of mercury from the SPE into the previous solution analyzed. When the solution is not renewed between analyses and the SPE is changed (Table 2, entry 2), the mercury lost from the electrode into the solution during the previous analysis is partly deposited onto the new SPE during the step of copper deposition. Thus, the mercury deposited onto the SPE increases leading to an enhancement of the RSD value as reported elsewhere [34]. Another alternative is to change the SPE and the solution between each analysis (Table 2, entry 4). These conditions, allowing the RSD obtained being the lowest, less than 10 %, were chosen and used later on. To complete this investigation, the analytical media in relation with total and electrolabile copper determination were then considered, i.e. diluted solution of the mineralized soil (MS) and leachate from a potassium nitrate aqueous solution, respectively. RSD was then calculated in the same conditions as those previously determined, i.e. by changing the SPE and the solution between each analysis. RSD, previously found to be 9 % in acetate buffer, was then 13 % in MS and 6 % in leachate with KNO_3 respectively. Additionally, RSD was also evaluated for Pb, with the perspective to study vineyard soils contaminated by both copper and lead. The values found were similar to those obtained for Cu. These results show that the repeatability does not only depend on the renewal of the SPE/analytical medium but also depends on the nature of the analytical medium. This information is important to keep in mind for the future applications necessarily made in the solution of mineralized soil and in leachate, for the determination of total and electrolabile metal concentrations respectively.

Effect of the pH and conductivity of the analytical medium

According to the conclusion of the previous part, the investigation was then focused on the analytical medium, and more especially on its pH and conductivity. A particular attention was given in case of electrolabile copper determination, because the pH and conductivity conditions in the medium can modified the original speciation of copper in the sample. Indeed, the more acidic is the solution and the more the copper is present under free form, while the conductivity has to be balanced in order to favor ionic transport without promote possible complexation with anionic ligands brought by the analytical medium. Additionally, in case of total analysis performed in solutions of soil mineralized, copper has to be totally under electrolabile form, which means that pH and conductivity have also to promote these forms. To check these effects, a solution containing 20 µg L^{-1} of copper was prepared in acetate buffer and the solution was analyzed by SWASV. In a first step, the solution was successively acidified by concentrated nitric acid until to obtain a pH corresponding to MB and MS.

Figure 1 shows the evolution of the copper peak surface according to the pH. For a pH ranging between 2 and 4.5 (4.5 = pH of the acetate buffer), the surfaces of copper peak obtained do not vary. Below a pH of 2 (0 = pH of the MB and MS), the surface of the copper peak decreases significantly: on average, a reduction from 30 % to 52 % of the copper peak surface is observed compared to the surface obtained at pH 4.5. However, theoretically, if the pH is reduced from 4.5 to 0, the copper would mostly become under free form. Consequently, the surface peak would be higher with the decrease of the pH but it is not the case occurring here. The gap between theory and results obtained can be due to the addition of nitric acid, which, at elevated concentration, may lead to the degradation of the mercury previously deposited onto the SPE.

In a second step, successive additions of KNO_3 were carried out in the solution to increase the conductivity (Figure 2). The highest sensitivity (i.e. the highest surface of peak) was expected, with no significant change in speciation regarding electrolabile forms. This is why speciation was calculated in the experimental field studied using Visual MinteQ software. From Figure 2, the highest sensitivity is reach from a concentration of KNO_3 of 10^{-2} mol L^{-1}. The best compromise between sensitivity and preservation of copper free form is obtained for a concentration of KNO_3 of 10^{-2} mol L^{-1}. In these conditions 97 % of copper proves to be under free form. The fact that the surface of peak remains the same over a concentration of KNO_3 of 10^{-2} mol L^{-1} while the free form decreases indicates that electrolabile forms (i.e. the addition of the concentration of Cu^{2+} and $CuNO_3^+$) remain also constant in the same range of concentration. This observation indicates that $CuNO_3^+$ is an electrolabile inorganic complex. Thus, if the electrolabile forms are the target analytes, a concentration of $KNO_3 \geq 10^{-2}$ mol L^{-1}, corresponding to a conductivity $\sigma = 1.4$ mS cm^{-1}, allows a satisfactory determination.

According to these results, the analytical medium of the standards was adjusted such that the pH and the conductivity were the same as in the sample solutions.

Quantification procedure of total copper and lead in the reference soil

The choice of the quantification procedure depends on both the analytical method and the sample nature. It is why a particular attention was given to this point, especially regarding the sample complexity. The accuracy of the copper determination was then evaluated. Ideally, a certified reference solution sample for its copper amount obtained from soil and should be used. However, such material does not exist. So, a certified reference soil was mineralized. Regarding the future applications of the method with environmental copper monitoring and the previous results obtained, the quantification was carried out by external standard calibration in (i) acetate buffer as a reference (pH = 4.5, σ = 13.2 mS cm^{-1}) and (ii) MB (pH = 0, σ = 129.0 mS cm^{-1}). The calibration was made in these two media and the quantification performed by standard additions in order to evaluate the possible matrix effects occurring from the mineralization process or other effects coming from the SPE measurement. For that, the limits of detection and slopes associated to the calibration curves were calculated for both media. The resulting limits of detection (LD) are 5.5 µg(Cu) L^{-1} and 6.8 µg(Cu) L^{-1} for the calibration in acetate buffer and MB respectively with the same deposition conditions. The corresponding linear regressions are presented in Figure 3.

The slope is significantly higher in acetate buffer than in MB that gives a supplementary illustration of the effect of the pH and conductivity on the electrochemical response. More quantitatively, for the calibration performed in the MB, the copper peak surfaces are reduced on average of 30 % compared to the calibration in the acetate buffer. This is in agreement with the previous results (Figure 1) showing that the copper signal decrease at least of 30 % when pH is below 2. The direct consequence is the difference of the copper concentrations obtained and reported in Table 3, first line. These results highlight that an accurate determination can be made by SPE-SWASV if the external calibration is performed in MB. Additionally to the copper quantification, lead, which is naturally present in this soil, was also determined. This joint determination (i.e. Cu and Pb) was made in order to confirm that the presence of lead in amounts around three times more important than the copper concentration does not disturb the copper quantification. In case of standard additions, where lead was added such that its concentrations reach 100 µg(Pb) L^{-1}, the resulting presence of lead in significant concentrations in MB does not conduct to any bias in the copper quantification. The corresponding voltammograms obtained are presented in Figure 4. They illustrate that no disturbance occurs even when the concentrations of both metals become high.

Application: Determination of copper speciation information in agricultural soil contaminated by lead

The aim of this part was to determine total and electrolabile copper by SPE-SWASV in order to (i) check its applicability by evaluating the accuracy when possible and the repeatability and (ii) apply to various agricultural soil samples to evaluate its potentialities in assessing hazard of metal contamination and check the robustness of the method. For that, the five soils (A - E, Table 1) previously described were investigated. First, the total copper and lead amounts were determined in soils. Then, copper and lead, which can be mobilized from these soils, were obtained by leaching and the corresponding electrolabile amounts were determined. The different concentrations obtained are presented in Table 4.

Concerning the total copper and lead concentrations in soils, analyses were additionally performed by ICP-MS in order to validate the values obtained by SPE-SWASV. All the values obtained are accurate for both techniques in a range from 10 to 100 mg kg^{-1} for copper and from 20 to 40 mg kg^{-1} for lead, corresponding to the concentration ranges of both metals in these soils. More specifically regarding the copper, these results confirm that copper can be reliably determined by SPE-SWASV in soils with different physico-chemical characteristics and also in the presence of lead.

Then, the electrolabile copper and lead concentrations were determined in the leachates obtained. The determination process was performed in 3 independent leachate analysis replicates, the leaching process being triplicated (i.e. n = 3x3). The concentrations obtained are presented in Table 4. For all the soils, the electrolabile lead concentration was below the limit of detection (0.02 mg kg^{-1}), so no result is presented in the Table 4. Moreover, electrolabile copper concentrations were quantifiable only for the vineyard soils and not for the soils A and B, which present electrolabile copper concentrations below the limit of detection (0.07 mg kg^{-1}).

The RSD evaluated from the measurement of electrolabile copper in the leachates was found to be 6 %. Additionally, no disturbance of the electrochemical signal was observed, especially no interaction between copper and lead signals. Thus, the repeatability of the electrolabile copper determination by SPE-SWASV is also robust regarding the different origins and chemical compositions of soils.

This electrochemical-based strategy is also proved to be able to give information on the consequences of the use of copper-based pesticides on soils with different farming. Thus, total copper concentrations are about ten times superior in vineyard soils than in wheat or sunflower soils whereas total lead concentrations are in the same order of magnitude. However, the part of electrolabile copper remains low in all agricultural soils. Indeed, less than 1 % of the total amount of copper is electrolabile in all the cases. This result shows that the potential hazard related to the copper mobilization from these soils is relatively low.

Conclusion

From analytical point of view, all these results confirm that SPE-SWASV can be used in a speciation strategy assessment of total metal content in soils and the determination of total and electrolabile metals in leachates. For that, some precautions have to be taken in order to ensure accurate and repeatable results. Specifically, SPE have to be changed and the solution renewed after each analysis. Because of the influence of the analytical medium conditions on the electrochemical response, a particular attention has also to be given to the pH and the conductivity of the sample solution and of the standard solutions used during the quantification procedure. By respecting these specifications, SPE-SWASV proves to be an interesting analytical tool in order to perform a mass balance of a metal of interest in environmental media such as agricultural soils. From environmental point of view, in the agricultural soils considered and containing various quantities of copper, a very low part of this element proves to be under electrolabile forms, confirming the relatively low impact of the copper regarding the potential diffusion via the water after mobilization from these soils.

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References

1. S. Yang, D. Zhou, H. Yu, R. Wei, and B. Pan, *Environ. Pollut. Barking Essex 1987*, 2013, **177**, 64–70.
2. S. Melaku, R. Dams, and L. Moens, *Anal. Chim. Acta*, 2005, **543**, 117–123.
3. C. Jie-liang, S. Zhou, and Z. You-Wei, *J. Environ. Sci. China*, 2007, **19**, 50–54.
4. M. Mihaljevič, V. Ettler, O. Šebek, L. Strnad, and V. Chrastný, *J. Geochem. Explor.*, 2006, **88**, 130–133.
5. A. Hildebrandt, M. Guillamón, S. Lacorte, R. Tauler, and D. Barceló, *Water Res.*, 2008, **42**, 3315–3326.
6. P. S. Kidd, M. J. Domínguez-Rodríguez, J. Díez, and C. Monterroso, *Chemosphere*, 2007, **66**, 1458–1467.
7. X.-Y. Tang, Y.-S. Cui, J. Duan, and L. Tang, *J. Hazard. Mater.*, 2008, **160**, 29–36.
8. M. Komárek, E. Čadková, V. Chrastný, F. Bordas, and J.-C. Bollinger, *Environ. Int.*, 2010, **36**, 138–151.
9. J. O. Agbenin, *Sci. Total Environ.*, 2002, **286**, 1–14.
10. S. Wolz, R. A. Fenske, N. J. Simcox, G. Palcisko, and J. C. Kissel, *Environ. Res.*, 2003, **93**, 293–300.
11. S. Ruyters, P. Salaets, K. Oorts, and E. Smolders, *Sci. Total Environ.*, 2013, **443**, 470–477.
12. J. C. Nóvoa-Muñoz, J. M. G. Queijeiro, D. Blanco-Ward, C. Álvarez-Olleros, A. Martínez-Cortizas, and E. García-Rodeja, *Sci. Total Environ.*, 2007, **378**, 23–27.
13. H. El Hadri, P. Chéry, S. Jalabert, A. Lee, M. Potin-Gautier, and G. Lespes, *Sci. Total Environ.*, 2012, **441**, 239–247.
14. L. . Brun, J. Maillet, J. Richarte, P. Herrmann, and J. . Remy, *Environ. Pollut.*, 1998, **102**, 151–161.
15. L. A. Brun, J. Le Corff, and J. Maillet, *Environ. Pollut. Barking Essex 1987*, 2003, **122**, 361–368.
16. T. Chirenje, L. Q. Ma, M. Reeves, and M. Szulczewski, *Geoderma*, 2004, **119**, 113–120.
17. R. Buzier, M.-H. Tusseau-Vuillemin, M. Keirsbulck, and J.-M. Mouchel, *Phys. Chem. Earth Parts ABC*, 2011, **36**, 500–505.
18. J. S. Meyer, *Mar. Environ. Res.*, 2002, **53**, 417–423.
19. M.-H. Tusseau-Vuillemin, C. Gourlay, C. Lorgeoux, J.-M. Mouchel, R. Buzier, R. Gilbin, J.-L. Seidel, and F. Elbaz-Poulichet, *Sci. Total Environ.*, 2007, **375**, 244–256.
20. H. Ming, W. He, D. T. Lamb, M. Megharaj, and R. Naidu, *Ecotoxicol. Environ. Saf.*, 2012, **78**, 344–350.
21. S. Sauvé, M. B. McBride, and W. H. Hendershot, *Environ. Pollut.*, 1997, **98**, 149–155.
22. L. A. Brun, J. Maillet, P. Hinsinger, and M. Pépin, *Environ. Pollut.*, 2001, **111**, 293–302.
23. J. L. Stauber, S. Andrade, M. Ramirez, M. Adams, and J. A. Correa, *Mar. Pollut. Bull.*, 2005, **50**, 1363–1372.
24. S. Tao, Y. J. Chen, F. L. Xu, J. Cao, and B. G. Li, *Environ. Pollut.*, 2003, **122**, 447–454.
25. M. Komárek, J. Száková, M. Rohošková, H. Javorská, V. Chrastný, and J. Balík, *Geoderma*, 2008, **147**, 16–22.
26. C. M. A. Brett, A. M. O. Brett, and L. Tugulea, *Anal. Chim. Acta*, 1996, **322**, 151–157.
27. P. R. M. Silva, M. A. El Khakani, M. Chaker, A. Dufresne, and F. Courchesne, *Sens. Actuators B Chem.*, 2001, **76**, 250–257.
28. O. Zaouak, L. Authier, C. Cugnet, E. Normandin, D. Champier, M. Rivaletto, and M. Potin-Gautier, *Electroanalysis*, 2010, **22**, 1159–1165.
29. S. P. Kounaves, J. J. O’Dea, P. Chandresekhar, and J. Osteryoung, *Anal. Chem.*, 1986, **58**, 3199–3202.
30. C. Truzzi, A. Annibaldi, S. Illuminati, E. Bassotti, and G. Scarponi, *Anal. Bioanal. Chem.*, 2008, **392**, 247–262.
31. V. Meucci, S. Laschi, M. Minunni, C. Pretti, L. Intorre, G. Soldani, and M. Mascini, *Talanta*, 2009, **77**, 1143–1148.

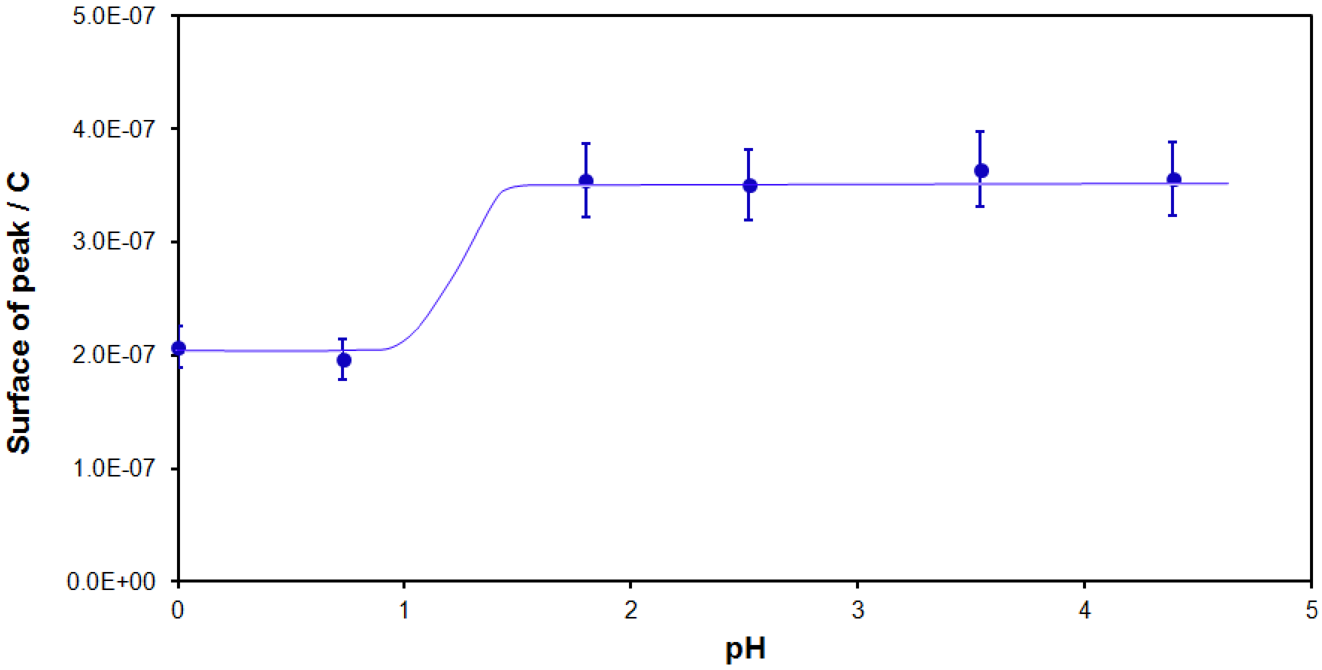
32. M. Li, Y.-T. Li, D.-W. Li, and Y.-T. Long, *Anal. Chim. Acta*, 2012, **734**, 31–44.
33. K. C. Honeychurch and J. P. Hart, *TrAC Trends Anal. Chem.*, 2003, **22**, 456–469.
34. O. Zaouak, L. Authier, C. Cugnet, A. Castetbon, and M. Potin-Gautier, *Electroanalysis*, 2010, **22**, 1151–1158.
35. O. Zaouak, L. Authier, C. Cugnet, A. Castetbon, and M. Potin-Gautier, *Electroanalysis*, 2009, **21**, 689–695.
36. S. B. Hocevar, I. Švancara, B. Ogorevc, and K. Vytřas, *Anal. Chem.*, 2007, **79**, 8639–8643.
37. Wang, Lu, Hocevar, Farias, and Ogorevc, *Anal. Chem.*, 2000, **72**, 3218–3222.
38. R. Pauliukaitė, S. B. Hočevar, B. Ogorevc, and J. Wang, *Electroanalysis*, 2004, **16**, 719–723.
39. W. Lund and D. Onshus, *Anal. Chim. Acta*, 1976, **86**, 109–122.
40. C. L. da Silva and J. C. Masini, *Fresenius J. Anal. Chem.*, 2000, **367**, 284–290.
41. J. Buffle and M.-L. Tercier-Waeber, *TrAC Trends Anal. Chem.*, 2005, **24**, 172–191.
42. A. Annibaldi, S. Illuminati, C. Truzzi, and G. Scarponi, *Mar. Pollut. Bull.*, 2011, **62**, 2813–2821.
43. M. Pesavento, G. Alberti, and R. Biesuz, *Anal. Chim. Acta*, 2009, **631**, 129–141.
44. S. Faucher, C. Cugnet, L. Authier, and G. Lespes, *Anal. Bioanal. Chem.*, 2013, 1–4.
45. J. Tolu, I. Le Hécho, M. Bueno, Y. Thiry, and M. Potin-Gautier, *Anal. Chim. Acta*, 2011, **684**, 126–133.
46. J. Wang and B. Tian, *Anal. Chem.*, 1992, **64**, 1706–1709.

Figure caption 1 Evolution of copper signal by SPE-SWASV with pH variations between 0 and 4.5. Square wave parameters: Electrodeposition -0.8 V for 60 s, equilibration time 30 s, frequency 25 Hz, potential step 8 mV, potential amplitude 25 mV.

Figure caption 2 Evolution of copper signal obtained by SPE-SWASV (◊) with conductivity fixed by potassium nitrate and comparison with free copper (Cu^{2+}, Δ) and labile copper ((Cu^{2+} + $CuNO_3^+$), +) predicted by Visual MinteQ software (same square wave parameters as in Figure 1).

Figure caption 3 Copper calibration plots from 0 to 63 µg L^{-1} concentrations in acetate buffer (pH = 4.5, σ = 13.2 mS cm^{-1}) (Δ) and in the MB solution (pH = 0, σ = 129.0 mS cm^{-1}) (◊) by SPE-SWASV (same square wave parameters as in Figure 1).

Figure caption 4 Typical voltammograms obtained for copper and lead standard addition quantification. (a) Solution of mineralized blank diluted 15 times (MB); (b) Solution of mineralized certified reference soil diluted 15 times (MS); (c) Same as (b) + Addition of 10 µg(Cu) L^{-1} and 30 µg(Pb) L^{-1}; (d) Same as (b) + Addition of 20 µg(Cu) L^{-1} and 60 µg(Pb) L^{-1} (same square wave parameters as in Figure 1).



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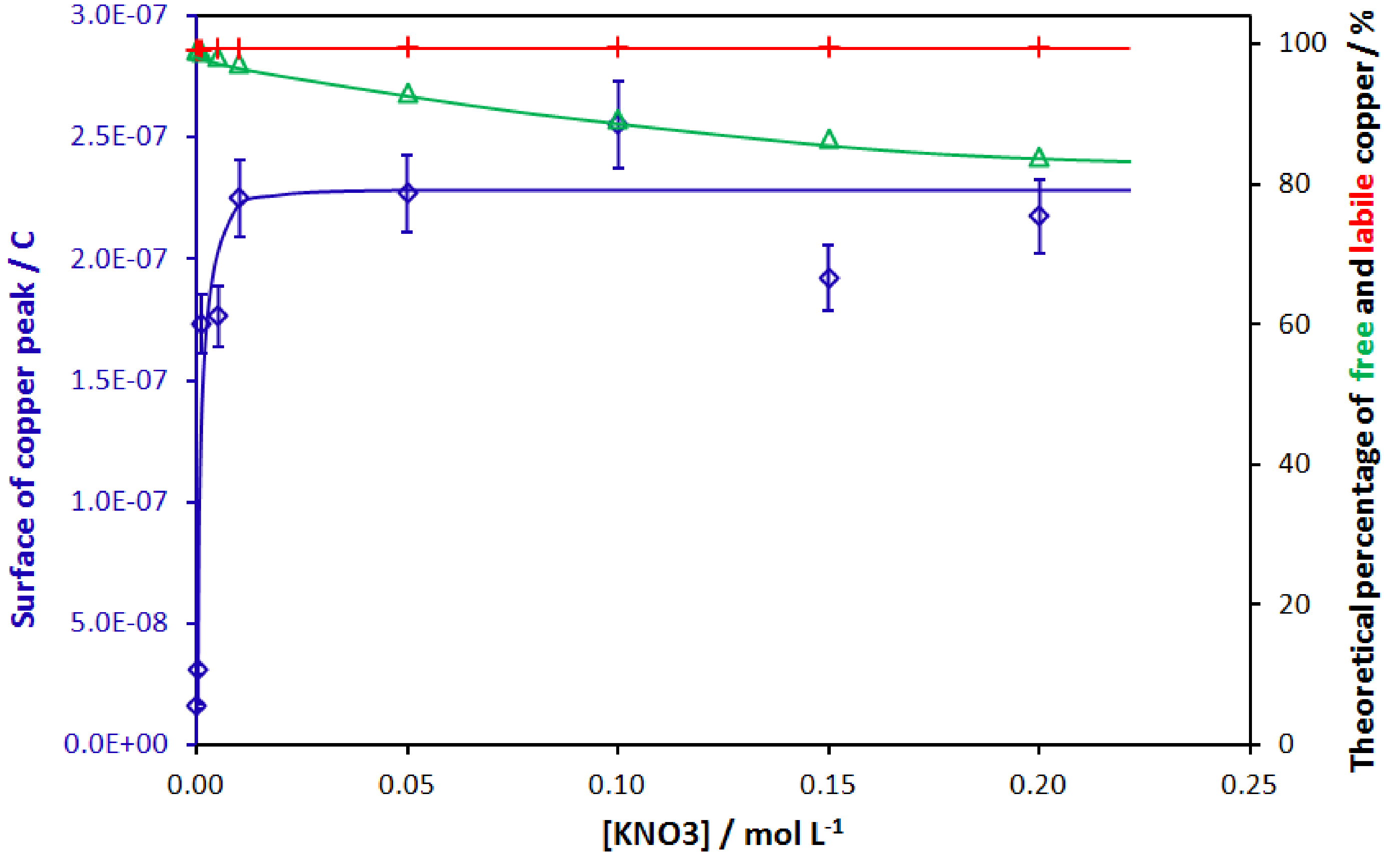

Figure 2 Evolution of copper signal obtained by SPE-SWASV (◊) with conductivity fixed by potassium nitrate and comparison with free copper (Cu^{2+}, Δ) and labile copper ((Cu^{2+} + $CuNO_3^+$), +) predicted by Visual MinteQ software (same square wave parameters as in Figure 1).

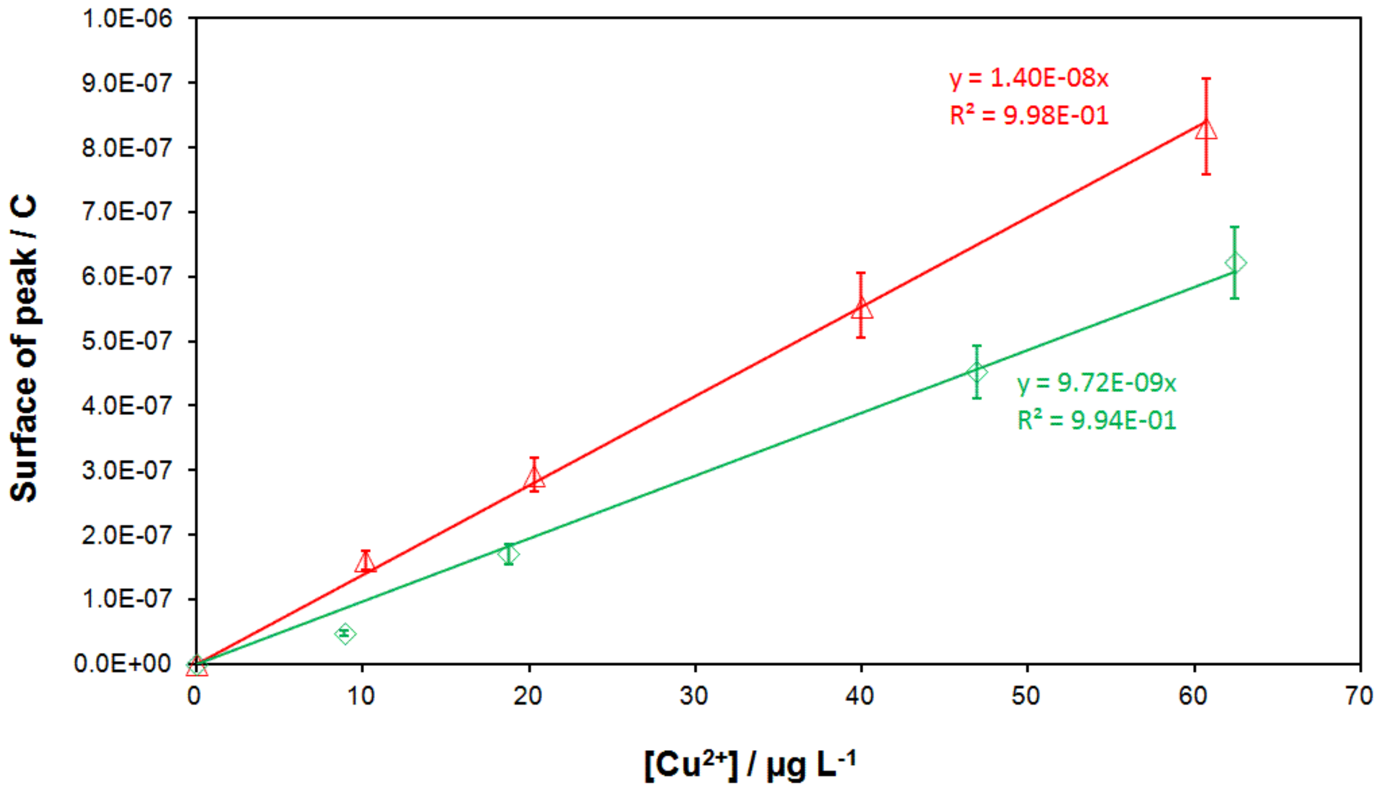

Figure 3 Copper calibration plots from 0 to 63 µg L^{-1} concentrations in acetate buffer (pH = 4.5, σ = 13.2 mS cm^{-1}) (Δ) and in the MB solution (pH = 0, σ = 129.0 mS cm^{-1}) (◊) by SPE-SWASV (same square wave parameters as in Figure 1).

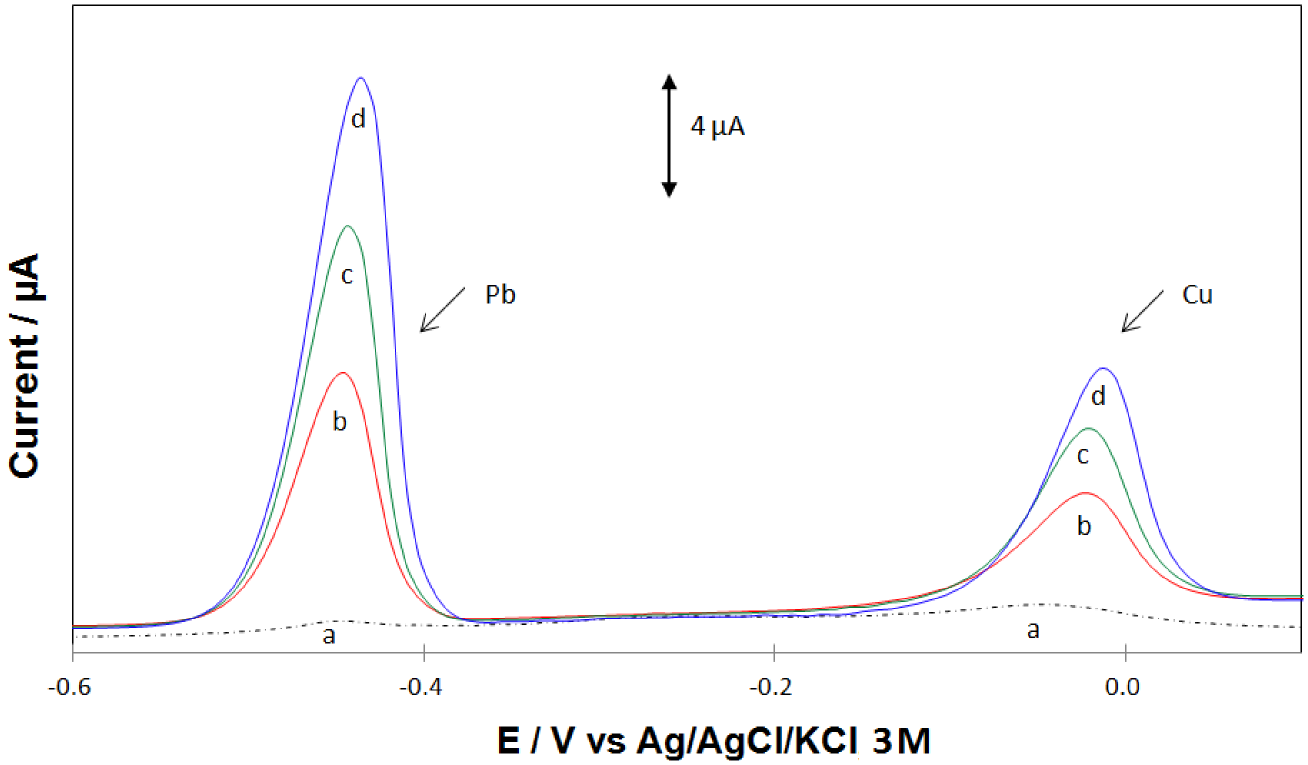

Figure 4 Typical voltammograms obtained for copper and lead standard addition quantification. (a) Solution of mineralized blank diluted 15 times (MB); (b) Solution of mineralized certified reference soil diluted 15 times (MS); (c) Same as (b) + Addition of 10 µg(Cu) L^{-1} and 30 µg(Pb) L^{-1}; (d) Same as (b) + Addition of 20 µg(Cu) L^{-1} and 60 µg(Pb) L^{-1} (same square wave parameters as in Figure 1).

Table 1 Selected physical and chemical properties of agricultural soils

Soil	**A**	**B**	**C**	**D**	**E**
Type	Loam	Loam	Sandy Loam	Sandy Loam	Loamy Sand
Farming	Wheat	Sunflower	Vineyard	Vineyard	Vineyard
pH (H_2O)	8.46	8.47	8.01	7.94	7.38
Organic carbon (g.kg^{-1})	6.84	7.27	5.94	8.88	7.21
Total azote (g.kg^{-1})	0.73	0.73	0.49	0.67	0.46
Phosphore P_2O_5 (g.kg^{-1})	0.02	0.02	0.08	0.07	0.08
CEC (cmol.kg^{-1})	11.3	11.4	8.13	9.14	6.19
Particle size composition (%)					
Sand (> 0.05 mm)	42.1	39.4	77.0	74.5	82.7
Silt (0.002 - 0.05 mm)	38.4	42.4	9.6	9.7	6.8
Clay (< 0.002 mm)	19.5	18.2	13.4	15.8	10.5

Table 2 Conditions of copper determination

Entry	**Conditions**	**RSD in % (from triplicates)**
1	Same solution[1] – Same sensor	20 %
2	Same solution – Different sensor	18 %
3	Different solution[2] – Same sensor	13 %
4	Different solution – Different sensor	9 %

[1] Same solution means that the three analyses were carried out in the same solution containing 20 µg L^{-1} Cu

[2] Different solution means that each analysis from the triplicate were carried out in a renewed solution containing 20 µg L^{-1} Cu

Table 3 Total metal in mg(metal) kg^{-1}(soil) determined by different quantification procedures from the mineralization solution of the certified reference soil (loam soil ERM-CC141)

	External calibration		by standard additions	**Reference value**
	in acetate buffer	in MB		
Copper	8.6 ± 0.8	12.1 ± 1.6	14.0 ± 1.9	**14.4** ± **1.4**
Lead	52 ± 5	36 ± 5	44 ± 6	**41** ± **4**

Table 4 Total and electrolabile metal concentrations from agricultural soils

Soil	**Lead**	**Copper**		
	Total concentration in soil (mg kg^{-1})	Total concentration in soil (mg kg^{-1})	Electrolabile concentration from soil (mg kg^{-1})	Percentage of electrolabile from soil (%)
A	26 ± 4	10 ± 2	≤ LD*	-
B	25 ± 4	11 ± 2	≤ LD*	-
C	29 ± 4	93 ± 12	0.38 ± 0.03	0.4 ± 0.1
D	33 ± 5	98 ± 13	0.47 ± 0.03	0.5 ± 0.1
E	35 ± 5	99 ± 13	0.50 ± 0.03	0.5 ± 0.1

*LD = 0.07 mg(Cu) kg^{-1}soil