Supplementary data

Theory

Square Wave Voltammetry

Square wave voltammetry (SWV) is an electrochemical technique that is currently being used in a wide variety of analytical chemistry fields[1-3]. It is well known that SWV offers a number of advantages over linear sweep, normal, and differential pulse excitation signals[3-4]. Many articles can be found in the literature citing SWV applications in biochemistry[5], pharmaceutical products[6-7], veterinary formulations[8], natural waters[9], trace quantity of metals[10-11], organic phase biosensors[12], food chemistry[13], and so also in cosmetics[14].

The theoretical aspects of SWV are well documented in literature[15], here it is only recalled that the excitation signal in SWV consists of a symmetrical square-wave pulse of amplitude E_{sw} superimposed on a staircase waveform of step height ΔE, where the forward pulse of the square wave coincides with the staircase step. The net current, i_{net}, is obtained by taking the difference between the forward and reverse currents ($i_{for} - i_{rev}$) and is centered on the redox potential. The peak height is directly proportional to the concentration of the electroactive species. The equation which relates the voltammetric peak high to the analyte concentration is:

$$i_{net} = k'\frac{n^2F^2}{RT}\Delta EC \qquad (1)$$

where k' is a constant, C is the electrolyte concentration, n the number of electrons, F the Faraday's constant, R the thermodynamic gas constant, T the absolute temperature and ΔE is the amplitude of the potential or of square wave.

In this study Ti^{4+} species are reduced to the Ti^{3+} state in acid media and well formed voltammograms and accurate current readings are obtainable with the HMDE[16-17]. Quantitative information are obtained with good sensitivity and selectivity by a single potential scan and employing an appropriate supporting electrolyte and applying the standard addition method[15].

Field Flow Fractionation techniques

The theory of these separation techniques is also well known and documented in literature[18-20] so that in this paragraph only the mathematical relations which relate the retention time, which is experimentally measured, with the particle sizes are reported.

The observed retention ratio R is defined as the ratio between the residence time of an un-retained species t^0 and the retention time of a retained species t_R , i.e.

$$R = \frac{t^0}{t_R} \qquad (1)$$

In FlFFF, where the applied field is a cross flow, when R is lower than 0.15[18], the retention time t_R is directly proportional to the particle's hydrodynamic diameter d, i.e. the diameter of a sphere defined by the particle rotating in all directions plus the hydration layer, which diffuses with the same speed as the particle under examination, according the simplified expression[18]:

$$t_R = t^0\,\frac{\pi\eta w^2 Q_c d}{2kTV^0} \qquad (2)$$

where η is the carrier viscosity, w the channel thickness, Q_c the applied cross flow (expressed as volumetric flow rate), k the Boltzmann constant, T the absolute temperature during elution, V^0 the channel void volume.

In SdFFF the external applied field consists in a centrifugal force, then the retention time t_R is proportional to the cubic of diameter d:

$$t_R = t^0 \frac{\omega^2 r w \pi \Delta\rho d^3}{36kT} \tag{3}$$

where ω is the angular velocity expressed as $\omega = 2\pi(\text{rpm}/60)$ (s^{-1}), r the radius of the centrifuge from rotation axis to the channel, $\Delta\rho$ is the difference between the particle ρ_p and the carrier medium ρ_c densities.

When a sample contains particles covering a very broad range of diameters, it become advantageous to change the field strength as the run proceeds so that eluting species at different times are subject to different average field strengths. There are several forms of field programming, but the only one which yields uniform fractionation power[21] throughout a broad diameter range is

$$G(t) = G_0 \left[\frac{(t_1 - t_a)}{(t - t_a)} \right]^p \tag{4}$$

where G is the centrifugal field strength, which is kept constant at G_0 for a time t_1; the optimum condition (yielding uniform resolution) is obtained when $t_a = -pt_1$, and $p = 8$. When power programming is used, between t_R and d the following relation exists

$$t_R - t_a \propto d^{1/3} \tag{5}$$

Methods

P-25 microwave assisted digestion: Method 1

Six different aliquots of Aeroxide TiO_2 Degussa P-25 were accurately weighed (see Table 1) into clean, dry TFM vessels, to which 2 mL of concentrated HNO_3 , 3 mL of HF and 4 mL of H_2O were added. The TFM vessels were sealed, placed in a high-pressure rotor, and then in the microwave oven for which an optimized digestion program was employed (see Table S4). At the end of the digestion program, the samples were left to cool, transferred to volumetric flask (100 mL) without filtering and diluted with deionised water. The acidic solutions were kept in PE-HD containers.

Foundation sampling method

According to the “Cosmetic legislation, volume 2 – Methods of Analysis”[22] the foundations were sampled as “Basic sample”, i.e. a unit taken from a batch offered for sale. The cosmetic products were sampled in their original containers and forwarded to the analytical laboratory unopened. Basic samples were all stored in the dark at 25 °C, the extracted suspensions in the refrigerator at 4°C. Since the basic samples were all creams (semi-solid), the containers were shaken, the test portions extruded, and the containers resealed immediately.

Foundation cream microwave-assisted digestion: Method 2

In order to mineralize the foundation cream, “Method 1” has been slightly modified as following: to at about 0.1g of sample were added 2 ml of concentrated HNO_3 and 4 mL HF, following the procedure described in the previous paragraph; the microwave program is reported in Table S4. At the end of the digestion program, the samples were left to cool, transferred to volumetric flask (50 mL) without filtering, and diluted with deionised water. For the exact foundation weights see Table 2 and 3 in the main text.

Foundation cream: TiO_2 particle solvent extraction

TiO_2 particles were extracted with a ternary mixture of solvents (water/methanol/hexane) as done in a previous work[16], however since the sample were different, also in this study the most suitable ratio among the solvents was determined through a set of preliminary experiments in which different volumes of water, methanol and hexane were mixed. Table 4 reports the data while the procedure can be summarized as follows: a weighed amount of foundation was dispersed in water and mixed for 60 s with the ultrasound probe set at 50% of maximum nominal power; after the addition of methanol, the suspension was mixed again for 60 s with the ultrasonic probe, transferred in a separation funnel where the hexane was added; the suspension was vigorous shaken to favor the mixing, and left to equilibrate for 1 hour then the two phases were separated. The aqueous phase was kept for the mineralization, the ICP-AES determinations, the SdFFF, FlFFF separations and TEM observations. The whole procedure was performed at room temperature (23-24 °C).

Since the methanol might switch off the torque during the ICP-AES analysis performed on the slurries or it could be an annoying interfering agent during the mineralization process, it was always removed from the samples by evaporation under N_2 atmosphere, and the lost volume substituted with deionised water. When concentrated sample slurries were necessary, the methanol was eliminated by evaporation at 70°C.

Aqueous phase microwave-assisted digestion: Method 3

To mineralize the TiO_2 particles contained in the aqueous phases achieved by the solvent extraction procedure, the following optimized method was applied. All mineralization were done inside of clean, dry TFM vessels, and the applied microwave oven program is reported in Table S4.

To a 2.5 mL of extracted sample were added 2 mL of concentrated HNO_3 , 3 mL of concentrated HF; the vessels were allowed to cool, then 1.5 g of H_3BO_3 was added to each of them, and they were returned to the microwave oven for 5 min at a power of 300 W.

Also in this case, the cooled samples were transferred to volumetric flask (50 mL) without filtering and diluted with deionised water.

SWV analysis procedure

Quantitative determinations were performed with standard additions method on the sample, digested as above, appropriately diluted with the supporting electrolyte (See next paragraph) in the polarographic cell; the analyzed volume was always 20 mL.

The P-25 and the foundation mineralized solutions were diluted 1:100 (sample volume 200 µL); the extracted mineralized solutions were analyzed after a dilution 1:10 or “as it”, for very diluted samples. The supporting electrolyte solution was a mixture of oxalic acid $H_2C_2O_4$ 0.25 M and H_3BO_3 0.20 M. The final pH value of the sample solution was always <2.

The instrumental experimental conditions were the following: Start potential = 0 mV and end potential = -700 mV both vs.Ag|AgCl|KCl_{sat}; wave amplitude = 80 mV; wave period = 40 ms; wave increment = 10 mV; sampling time = 8 ms; purge and stir time = 600 s; stirring speed = 400 r.p.m.; drop size = 60 a.u.

SWV method: choice of the supporting electrolyte solution

Examples of accurate and rapid determination of Ti^{4+} at trace levels in acidic media by voltammetry at the DME are reported in literature[23-24], so that the SWV method can be considered a good alternative for quantifying the TiO_2 in commercial foundations to the more established ICP-AES method, which is surely rapid but very expensive.

A preliminary check was done by using only oxalic acid 0.25 M as supporting electrolyte[16, 25-27] and an aqueous solution of Ti^{4+}; the Ti^{4+} reduction to Ti^{3+} was observed at E_p = -280 mV (vs.Ag|AgCl|KCl_{sat}) and the current intensity and the peak position were not sensitive to oxalic acid concentration up to 1 M.

In order to verify all the conditions for the analysis of the mineralized foundations, 2 mL of blank reagents solution (concentrated HNO_3, concentrated HF and deionised H_2O) were diluted with 18 mL of supporting electrolyte solution (oxalic acid 0.25 M – dilution ratio of 10). The dilution is required because of the high TiO_2 content in the foundation and especially since the cell is not HF resistant. Figure S1**a)**, curve A shows the result of this check, in which a shift of 100 mV in the peak position was observed (about E_p = -380 mV vs.Ag|AgCl|KCl_{sat}); since the low pH of the solution was not enough to justify such a difference in the peak potential, the presence of an interfering ion, concentrated enough to give a peak at -380 mV which completely hid the Ti^{4+} signal was supposed. The solution was analyzed by GF-AAS, which confirmed a contamination due to Pb^{2+}, coming from the N_2 used to de-aerate the solution before the measurements. To prove it, a sequence of purging of 5 min each was done on the acidic solution and the result are reported in Figure S1**b**); after 40 min of degassing about 7 mg L^{-1} of lead were detected in the solution. The hypothesis was that F^- could complex the Pb^{2+} as PbF_4^{2-}; to impede this reaction, a weighed amount of H_3BO_3 able to capture the excess of F^- ions, was added to the oxalic acid 0.25 M solution to get a 0.20 M concentration (see Figure S1**a)** and 1**c)**, curve B). When an addition of Ti^{4+} standard was done to this new solution, the Ti^{4+}/Ti^{3+} peak reduction was registered at -260 mV (see Figure S1**c)**, curve C).

The electrolyte solution to dilute all the mineralized foundation samples was a mixture of $C_2O_4H_2{\cdot}H_2O$ 0.25 M and H_3BO_3 0.20 M.

Voltammetric determinations: mineralized TiO_2 Aeroxide Degussa P-25 powder

Figure S2**a)** shows a series of square wave voltammograms for the reduction of Ti^{4+}/Ti^{3+}. The weaker peak is due to the sample, while the following more intense peaks are due to the addition of Ti^{4+} standards (5, 10, 15, 20, 25 mg L^{-1} respectively). The net current profiles are gaussian-shaped and symmetrical, which indicated a reversible process; the peak potentials E_p = -280 mV coincided with the potential registered when the Ti^{4+} standard was tested. All the standard addition curves had a correlation coefficient very close to 1, as that one reported as example on the top of the plot.

Voltammetric determinations: mineralized foundation samples

Figure S2**b**) reports as example a series of voltammograms registered for **Sample #1**, they were also for this “real” sample bell-shaped and symmetrical with the peak potentials close to the experimental value (E_p = -280 mV vs Ag/AgCl) determined with the Ti^{4+} standard solution. Also for this sample the standard addition curves had a correlation coefficient very close to 1, as that one reported as example on the top of the plot.

Figure S4**a**) reports as example a series of voltammograms registered for **Sample #6**, in which are visible the interfering peak of Cu^{2+} (E_p =-100 mV) on the left of the Ti^{4+} peak (E_p = - 280 mV) and a scarcely visible shoulder on the right, likely due to Pb^{2+} (E_p = -360 mV). Analogous voltammograms were recorded during the analysis of **Sample #4**. It is reasonable to suppose that the perturbation in the baseline due to the Cu^{2+} peak introduced a bias, which affected the Ti determinations; in Figure S4**b**) is shown as example the Cu^{2+} determination done on **Sample #6**, which assessed a Cu contamination of the order of 1.7 mg L^{-1}.

Table S1: List of the studied foundation creams

Sample #	**Commercial sample**	**Sold in: Country**
1	MaxFactor - Facefinity with permawear – SPF 15	USA
2	MaxFactor - Lasting Performance	IT
3	MAYBELLINE – Affinitone	IT
4	MAYBELLINE - Instant Liss' Result - SPF 18	IT
5	CLINIQUE - Stay-true makeup oil free formula	IT
6	RIMMEL	IT

Table S2: SdFFF programmed field parameters

Program	**Initial RPM**	**Relaxation time(min)**	**t_1 (min)**	**t_a (min)**	**Final RPM**	**Hold RPM**
A	350	10	8	-64	20	20
B	500	10	8	-64	20	20

Table S3 : Operating conditions and instrumentation of the ICP-AES

Parameter	**Value**
RF generator	40 MHz, free –running
RF power	1350 W
Torch type	Fassel type
Injector, id	Alumina, 2.0 mm
Viewing mode	Axial
Auxiliary argon flow rate	0.50 L min^{-1}
Nebulizer argon flow rate	0.65 L min^{-1}
Plasma gas flow rate	15 L min^{-1}
Spray chamber type	Cyclonic buffled
Sample introduction	Peristaltic pump
Sample uptake flow rate	1.5 mL min^{-1}
Detector	Segmented-array charge-coupled (SCD)

Table S4. Temperature Microwave Program for the Aeroxide Degussa P-25 and foundation mineralization.

Aeroxide Degussa P-25			**Foundation cream**		
Time (min)	**Power (W)**	**Temperature (°C)**	**Time (min)**	**Power (W)**	**Temperature (°C)**
5	250	120	1	250	120
5	400	190	1	0	120
5	600	220	5	250	140
5	250	220	4	400	200
			3	550	200
			5	300	200

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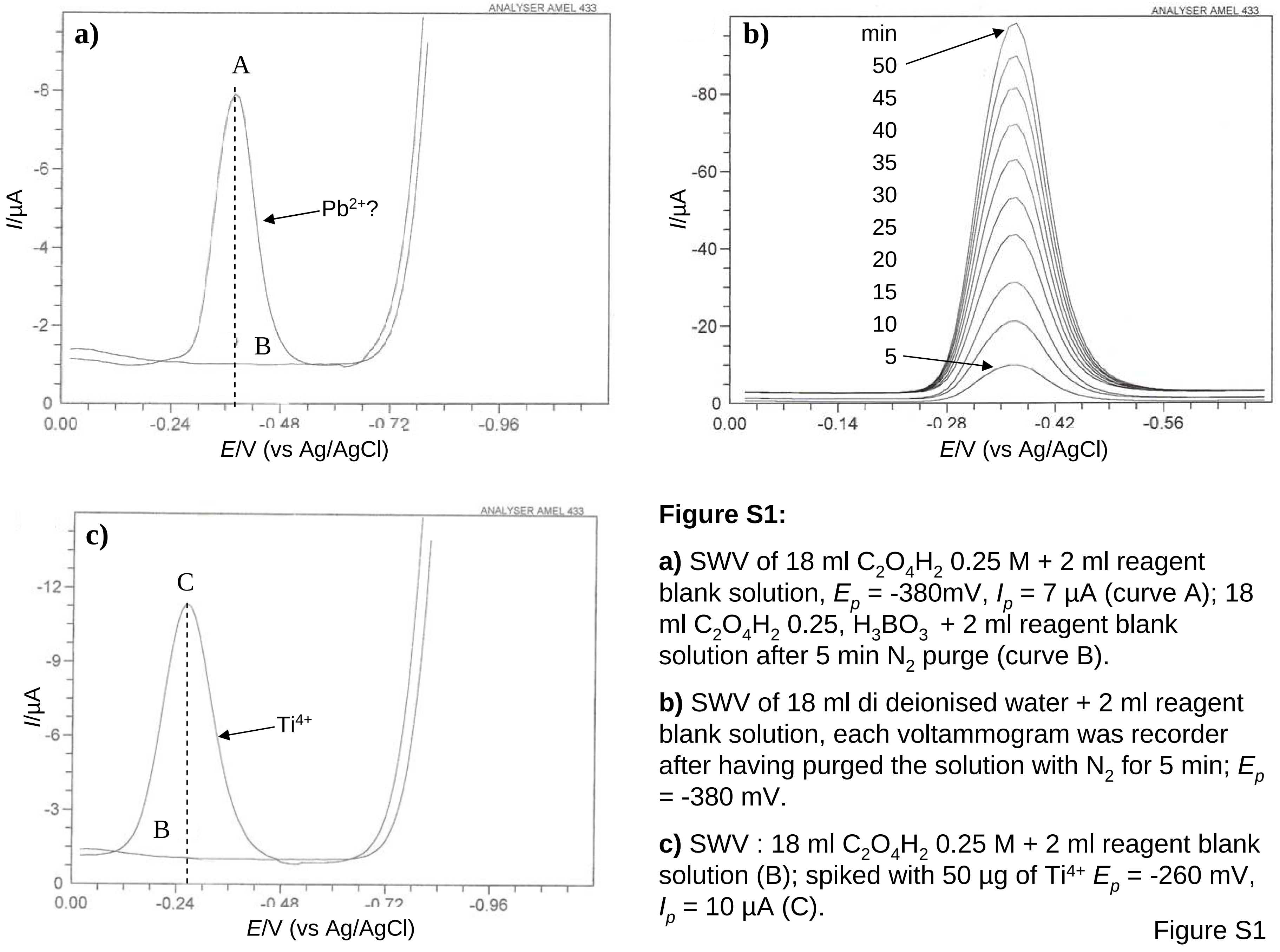

Figure S1:

a) SWV of 18 ml $C_2O_4H_2$ 0.25 M + 2 ml reagent blank solution, E_p = -380mV, I_p = 7 µA (curve A); 18 ml $C_2O_4H_2$ 0.25, H_3BO_3 + 2 ml reagent blank solution after 5 min N_2 purge (curve B).

b) SWV of 18 ml di deionised water + 2 ml reagent blank solution, each voltammogram was recorder after having purged the solution with N_2 for 5 min; E_p = -380 mV.

c) SWV : 18 ml $C_2O_4H_2$ 0.25 M + 2 ml reagent blank solution (B); spiked with 50 µg of Ti^{4+} E_p = -260 mV, I_p = 10 µA (C).

Figure S1

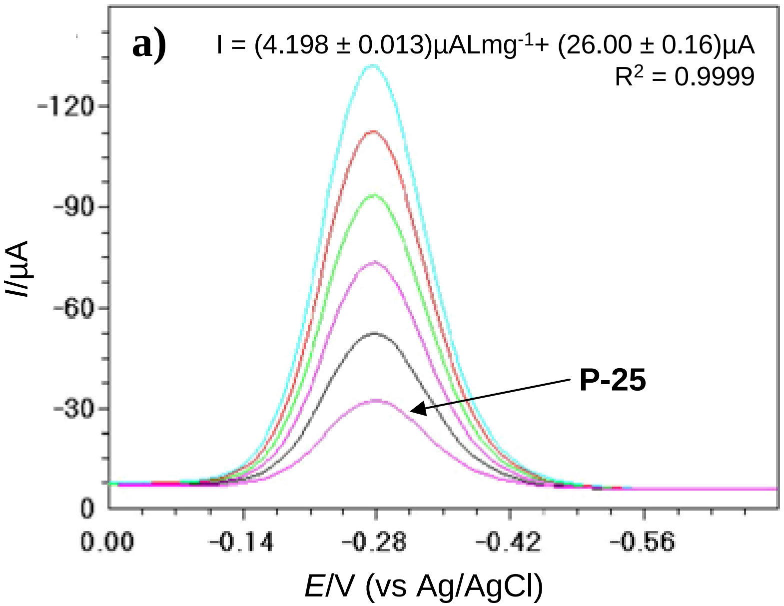

Figure S2: a) SWV of the Degussa P-25 sample, after acid digestion, and dilution with $H_2C_2O_4$ 0.25M + H_3BO_3 0.20M: standard addition of Ti^{+4} (5,10,15,20,25 mg L^{-1}).

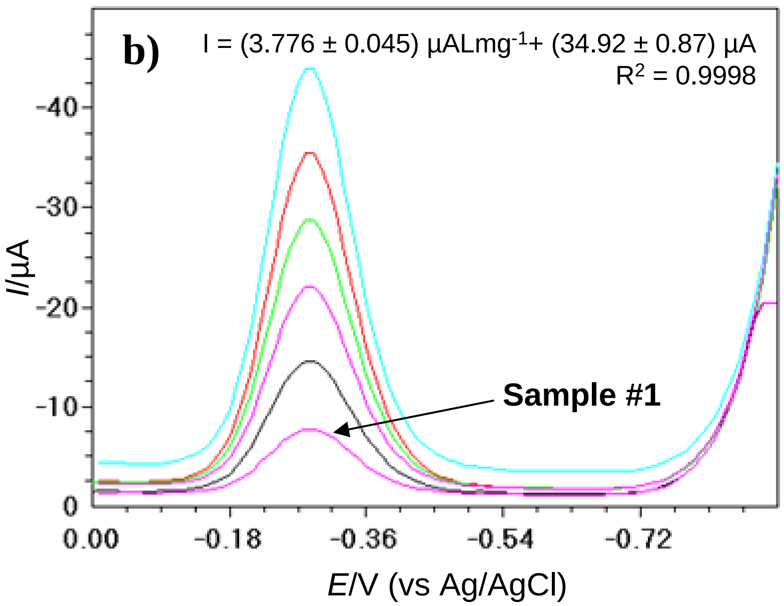

Figure S2: b) SWV of **Sample #1**, after acid digestion and dilution 1:10 with $H_2C_2O_4$ 0.25M + H_3BO_3 0.20M: standard addition of Ti^{+4}.

Figure S2

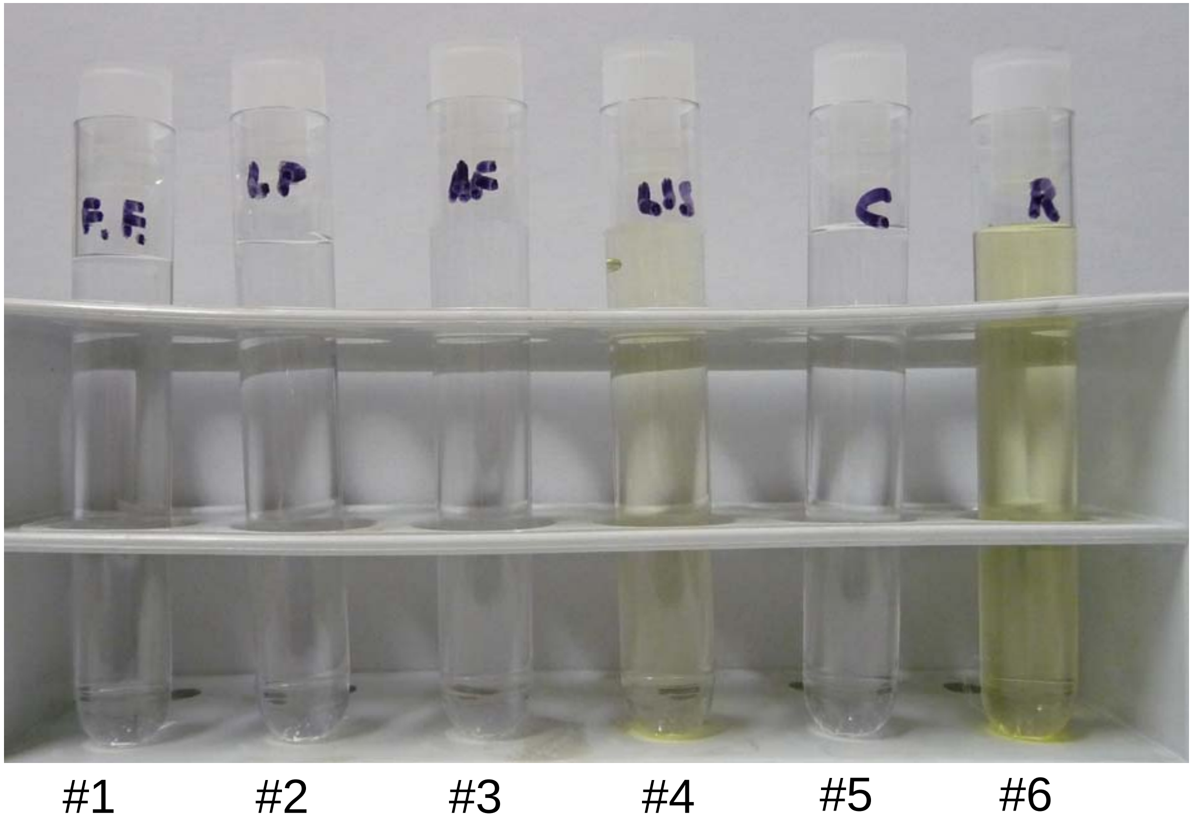

Figure S3: Photograph of the mineralized solutions achieved from **Samples #1, 2, 3, 4, 5, 6**.

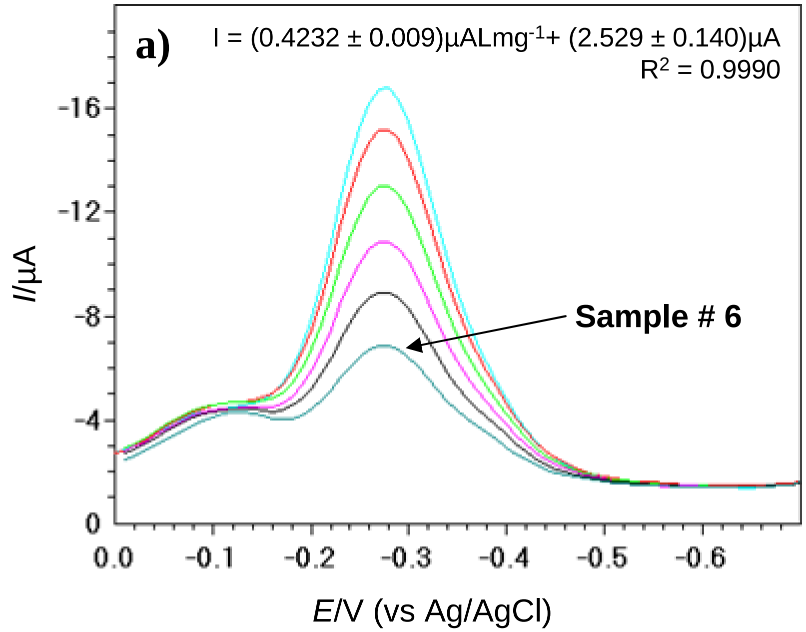

Figure S4: a) SWV of **Sample #6** after acid digestion and dilution 1:100 with $H_2C_2O_4$ 0.25M + H_3BO_3 0.20M: standard addition of Ti^{+4} (5,10,15,20,25 mg L^{-1}) in presence of Pb^{+2} (Ep = -360 mV) and Cu^{+2} (Ep = -100 mV).

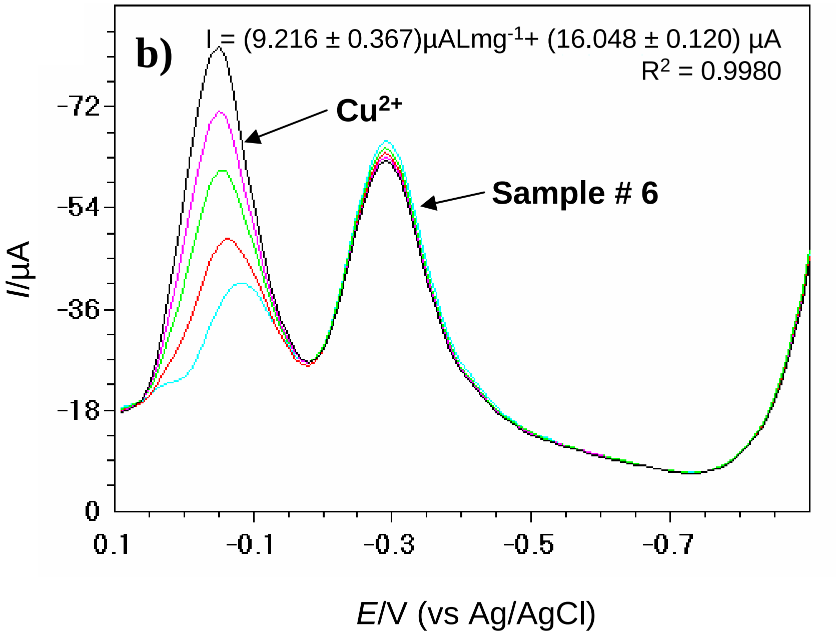

Figure S4: b) Example of Cu^{2+} determination in **Sample #6** (diluted 1:10); standard addition of Cu^{2+} = 1.25, 2.50, 3.75, 5.00 mg L^{-1}.

Figure S4

#3 #4 #6 #1 #2 #5

Figure S5: Photograph of the extracted concentrated suspensions (aqueous phases) of **Samples #1, 2, 3, 4, 5, 6**.