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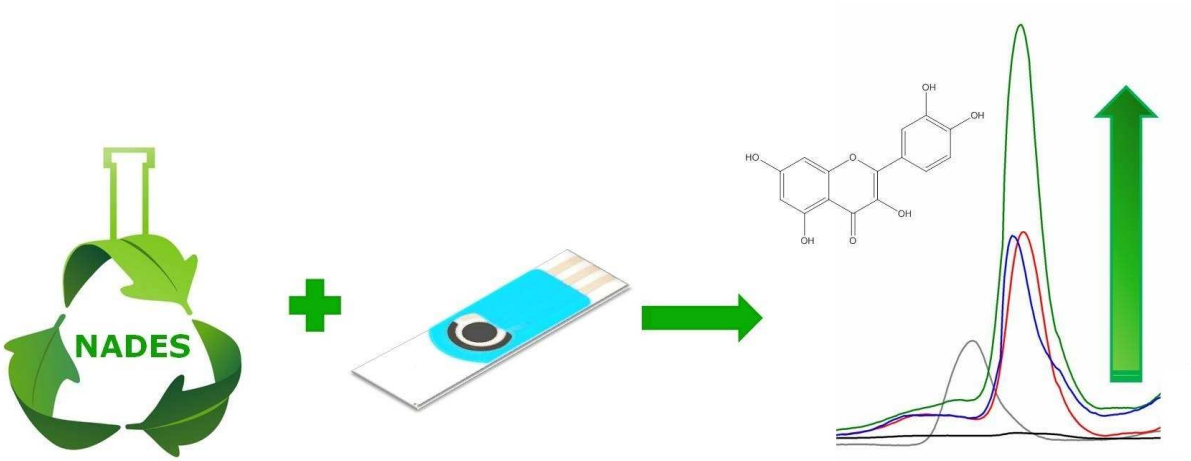

Enhanced electrochemical detection of quercetin by natural deep eutectic solvents

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Abstract

New trends in analytical chemistry encourage the development of smart techniques and methods aligned with Green Chemistry. In this sense, Natural Deep Eutectic Solvents represents an excellent opportunity as a new generation of green solvents. In this work a new application for them has been proposed and demonstrated. These solvents were synthesized by combinations of inexpensive and natural components like, Glucose, Fructose, Citric acid and Lactic acid. The different natural solvents were easily prepared and added to buffer solution in different concentrations, allowing the enhancement of electrochemical detection of an important representative antioxidant like quercetin (QR) with improved signal up to 380%. QR is a ubiquitous flavonoid widespread in plants and food of plant origin. The proposed method using phosphate buffer with a eutectic mixture of Citric acid, Glucose and water in combination with carbon screen printed electrodes exhibited a good analytical performance. Detection and quantification limits were of 7.97 and 26.3 nM respectively; and repeatability with %RSDs of 1.41 and 7.49 for peak potential and intensity respectively. In addition, it has proved to be faster, greener and cheaper than other sensors and chromatographic methods available with the additional advantage of being completely portable. Furthermore, the obtained results demonstrated that the proposed method is able for the determination of QR in complex food samples.

1. Introduction

In the last decades, the concept “green” acquired a new significance in chemistry. The definition of sustainable development and green chemistry changed the way of thinking processes and methods. A critical issue is to look for an alternative to traditional organic solvents due to their low biodegradability, high toxicity and cost. After years of intense research, trying different mixtures with diverse compounds, a spark of light emerged from nature: in 2011 Choi et al. [1] coined the term Natural Deep Eutectic Solvents (NADES) for mixtures that are liquid supermolecules made of natural metabolites bound together by inter-molecular interactions, particularly hydrogen bonding [2]. They have several advantages over synthetic ionic liquids, e.g. their low costs, biodegradability, nontoxicity, sustainability, and simple preparation methods [3]. NADES offer endless opportunities at method development showing very good physicochemical properties as solvents: negligible volatility, liquid state even far below 0℃, adjustable viscosity, sustainability, biodegradability combined with acceptable toxicity profiles, and high solubilization power of both polar and non-polar compounds [3, 4].

The components of NADES are natural metabolites, e.g. sugars, alcohols, organic acids, amino acids and amines, which have several hydroxyl groups, carboxyl groups, or amino groups [1]. Those groups give rise to hydrogen bonding interactions, leading to highly structured liquids [5]. Such liquids can, in turn, form additional hydrogen bonds with solutes, increasing their solubilization ability, e.g. of phenolic compounds [6]. Thus considering the challenge of Analytical Chemistry to develop new techniques and methods aligned with Green Chemistry, NADES represents an excellent opportunity as a new generation of green solvents with many possible applications due to the features can be tailored by changing the nature and molar ratio of their hydrogen-bonding components.

Flavonoids are natural products widely distributed in the plant kingdom and generally present in the common human diet. Quercetin (QR, 3,3´,4´,5,7-penta hydroxyl flavones) is the most common flavonoid widespread in plants and food of plant origin [7]. Onions ranked highest in QR content in a survey of 28 vegetables and nine fruits [8]. The amount of QR in onions varies depending on bulb color and type, being distributed mostly in the outer skin and rings [9]. Most of the studies have revealed various beneficial effects on human health, including anti-viral, anti-cancer, anti-inflammatory and anti-tumor activity [10, 11].

Traditional methods for determination of QR include spectrophotometry [12], gas chromatography combined with mass spectrometry [13], high performance liquid chromatography with UV detection [14], or spectrophotometric and coulometric detection [15]. These techniques often require complicated and time consuming pretreatments and/or expensive experimental equipments. Therefore, simpler electrochemical approaches have been developed for QR determination in a wide range of matrices. These methods usually applied modifications of the electrode with graphene [11, 16-19], carbon nanotubes [18, 20-22], nanoparticles [16, 17, 19, 22, 23], alumina microfibers [24] and several combinations of the foregoing [16-19, 22]. Electrochemical methods involving modified electrodes are simpler than traditional but they are relatively expensive and time consuming; the modification of the electrode itself decreases sample throughput significantly.

In this sense the development of smart methodologies avoiding unnecessary steps that allow polyphenols determination with similar features are necessary. NADES application in electrochemistry allows the possibility of employing unmodified electrodes. Considering the very recent discovery of these natural solvents, no information can be found concerning the study of detection enhancement provided by them. Thus, the main purpose

of this work was to explore the skills of selected NADES as enhancers of electrochemical detection for a representative phenolic compound (QR). NADES were synthesized by different combinations of glucose, fructose, citric acid and lactic acid. Phosphate buffer solutions were prepared with different amounts of NADES to test their effect on the sensitivity in differential pulse voltammetry. Thus, a methodology that fully represents the principles of green analytical chemistry was developed. Indeed, the proposed methodology was applied for the determination of QR in onion samples. The overall profile of the analytical methodology; including extraction and determination was achieved using NADES as solvents for all steps. Afterwards, results were contrasted against HPLC-MWD with satisfactory results.

2. Experimental Section

2.1. Chemicals

QR was purchased from Sigma Chemical (St. Louis, MO, USA). D (-) Fructose, D (-) Glucose, Lactic acid and Citric acid were obtained from Biopack (Buenos Aires, Argentina). Sodium hydrogen phosphate was purchased from Carlos Erba Reagents (Milano, Italy). Phosphate buffer 5 mM was prepared by dissolving appropriate amounts of sodium hydrogen phosphate. Ultrapure water (18 MΩ cm) was obtained from Milli-Q system (Millipore, Bedford, MA, USA).

2.2. Equipment

All electrochemical measurements were performed, at room temperature, on an USB-based portable electrochemical station μ-Stat 200 Bipotentiostat (Dropsens, Oviedo, Spain) controlled by DropView 200 software. The electrodes used in this work were carbon screen-printed electrodes (CSPE), which integrates a three-electrode system based on carbon as counter electrode, carbon working electrode of 4 mm diameter and a silver reference electrode (Dropsens, Oviedo, Spain).

The HPLC instrument was a Dionex Ultimate 3000 (Dionex Softron GmbH, Thermo Fisher Scientific Inc., Germering, Germany) consisting of vacuum degasser unit, autosampler, quaternary pump and chromatographic oven. The detector was a Dionex MWD-3000 (RS) model. The working wavelength was fixed at 370 nm. The Chromeleon 7.1 software was used to control all the acquisition parameters of the HPLC-MWD system and also to process the obtained data. A Zorbax SB-Aq column (4.6 mm × 150 mm, 5 µm) Agilent Technologies was used. Ultrapure water with 0.1% Formic acid (A) and Acetonitrile (B) were used as mobile phases. The following gradient was used: 0–2.7 min, 5% B; 2.7–10.7 min, 30% B; 10.7–11 min, 35% B; 11–15 min, 50% B; 15–15.5 min, 50% B: 15.5–16 min 30% B; 16–16.5 min 5% B; 16.5–17 min 5% B. The mobile phase flow was 1 mL/min. The column temperature was held at 20 ℃ and the inject ion volume was 5 µL.

2.3. NADES synthesis

NADES synthesis were carried out easily following the heating and stirring method described by Dai et al. [6]. Three different NADES were prepared using inexpensive and natural components, in the following combinations and ratios: Citric acid, Glucose and H_2O (CGH, 1:1:2); Lactic acid, Glucose and H_2O (LGH, 5:1:3); Citric acid, Fructose and H_2O (CFH, 1:1:2). The two-component mixture with calculated amounts of water were placed in a bottle with a stirring bar and cap and heated in a water bath below 80℃ with agitation till a clear liquid is formed (60 min). The stability of synthesized NADES was tested, and they were stable for at least 2 months after its preparation.

2.4. Sample extraction

Red, green and yellow onions were obtained from local market. The samples were cut into small pieces, lyophilized and then the dried samples were homogenized with liquid nitrogen. The homogenates were extracted with 1mL of CGH (1:1:2) or methanol. For this

purpose, 50 mg of the homogenate were transferred to an extraction tube with the solvent, vortexed and then the extraction was accelerated by ultrasonication for 30 min. The onion extract was stored in a refrigerator for further use.

2.5. Electrochemical procedure

The measurements were recorded using a drop of just 50 µL of the solution in order to cover the three electrode system. The electrochemical behavior of QR in working buffer was examined using differential pulse voltammetry (DPV). DPV was performed with a potential range from −0.5 to +1.0 V, with 5 mV step potential, 25 mV pulse potential, 20 mV s^{-1} scan rate, 0.01 s pulse time and 3 s equilibration time.

3. Results and Discussion

3.1. Physicochemical properties of NADES

In this work several combinations of sugars (Sucrose, Fructose, Glucose) and organic acids (Lactic acid, Citric acid, Tartaric acid) in different molar ratios were tested to obtain NADES. The three selected were the ones with better features in terms of stability (at least two months) and low density which facilitates their usage as extracting solvents and detection media. These solvents were CGH, CFH and LGH with densities of 1.46, 1.44 and 1.23 g mL^{-1} and pH of 2.0, 2.0 and 0.78 respectively.

3.2. NADES improved signal in QR determination

In order to explore the performance of NADES as modifiers of background electrolyte for electrochemical detection of QR, three different NADES (CGH, CFH and LGH) were synthesized (as mentioned in section 2.3.) and added to buffer solution at different concentrations. Several buffer agents at different concentrations were tested and the best results were obtained with 5 mM of phosphate, which was selected for further studies. Fig. 1 shows the results obtained in the determination of 17 µM QR in phosphate buffer solution 5 mM (pH 7.6) with different percentage of added NADES.

As can be seen from Fig. 1 A the three studied NADES have a positive effect over the current intensity of QR for concentrations lower than 10% (v/v). The response of this flavonoid increases with the increment of the concentration of NADES until it reaches to 10% (v/v) except for CFH that was 5% (v/v), and then decreases at concentrations of NADES above 10% (v/v) for LGH and CGH and above 5% (v/v) for CFH. This suggests that the interaction of QR with the structure of NADES is favorable at low concentrations of the solvent. Further studies with higher concentrations of CGH have demonstrated that the enhancement effect is lost. Indeed the signal of QR disappears at concentrations upper than 80% (v/v) of CGH (see Fig. 1S).

The above suggests that the interaction of the flavonoid with the structure of NADES provides a suitable environment for electron transference and has a great effect on the kinetics of electrode reaction for QR. In this sense, Zheng et al. [25] demonstrated that using a deep eutectic solvent (DES) electrolyte can effectively reduce the charge transfer resistance and reaction resistance of QR, increasing the electronic exchange rate, which can result in a sensitization effect for this molecule. In addition, the same effect can be observed for ionic liquid in the determination QR and other analytes [26-28]. Supporting this finding, a very interesting work by Dai et al. [3] using FT-IR demonstrated that for QR dissolved in glucose-choline chloride based NADES, the hydroxyl groups in QR donate protons to hydrogen bonds with solvent molecules and QR has a different conformation in GCH from that in the solid state. In addition, they found the existence of multi H-bond interactions between QR and NADES. This demonstrates that this flavonoid has strong interaction with the structure of the natural solvent. In this sense, we tested the effect of the individual components of CGH. While the carbohydrate showed no effect on the analytical signal, the presence of citric acid gave a mild-higher signal compared to the

phosphate solution (data not shown). The latter suggests that analyte interaction with the supramolecular structure of CGH could be ascribed to the organic acid moieties.

Scheme 1 showed NADES Improved Signal (NIS) calculation and Table 1 showed NIS for low concentrations of NADES assayed. As can be seen and in accordance with the above-discussed, almost all low solvent concentrations tested have a positive effect on the signal of QR.

Scheme 1: NADES Improved Signal (NIS) calculation.

$$NIS\% = \left(\frac{QR\ signal\ with\ NADES - QR\ signal\ without\ NADES}{QR\ signal\ without\ NADES}\right)100 \quad (1)$$

Table 1

NADES improved signal.

NADES	% Added	*NIS%*
CGH	5	87.08
	10	379.58
	15	261.72
CFH	5	127.12
	10	36.84
	15	21.21
LGH	5	46.89
	10	92.18
	15	-77.51

Fig. 1 B shows the voltamograms comparing the best results obtained for the different modifications evaluated. A NIS effect up to 380% was obtained for CGH 10% (v/v). In this case the intensity of peak currents for QR increased while the oxidation peak potentials mildly shift to a positive oxidation values compared to peak current of QR in buffer without modification. As can be seen, peak position and shape change; giving evidence that the

enhanced response could be explained by kinetic origin. As stated above, the interaction of the flavonoid with NADES reduces both charge transfer and reaction resistance of QR, and increase the electronic exchange rate, so the peak potential should be shifted negatively. However, as can be seen in Fig. 1B, the potential shifted positively. This effect could be explained by the strong interaction of the flavonoid with the structure of the natural eutectic solvent (H-bond). Moreover, it has previously been demonstrated that QR changes its conformation in NADES [3]. Thus, 10% of CGH was fixed for subsequent experiments.

3.3. Effect of scan rate on the oxidation of QR

The effect of scan rate on the electrooxidation of QR at the CSPE was investigated by cyclic voltammetry to acquire information about electrochemical mechanism from the relationship between peak current and scan rate of potential. The cyclic voltammograms of 17 μM QR in 5mM phosphate buffer with 10% of CGH were recorded at different scan rates from 10 to 200 mV s^{-1} (Fig. 2). As shown in Fig. 2 inset, a linear correlation (R^2 = 0.981) was obtained between the peak current and the scan rate, indicating that the oxidation process is controlled by adsorption in agreement with previous works [11, 25, 29, 30]. The regression equation was I_{pa} (μA) = 12.31υ (mV s^{-1}) – 38.17.

3.4. Effect of pH

To achieve an optimal electrochemical response of QR, the oxidation responses in selected buffer solution (5 mM phosphate buffer with 10% CGH) with different pH values were investigated using DPV on the range of 6 to 11. As can be seen in Fig. 3, the effect of pH was negligible in the assayed range. As already stated, NADES plays a crucial role in the oxidation of this flavonoid that could act reducing the effect of pH over the peak current of QR. Nevertheless as the pH value increased from 6.0 to 7.0, the oxidation peak current of QR gradually increased, suggesting that the oxidation of QR was easier at

higher values of pH. On further increasing the pH value to 8.0, the oxidation peak current of QR gradually decreased. To achieve the highest sensitivity, pH 7.0 buffer solution was subsequently used for the determination of QR.

3.5. Analytical performance

The analytical performance was also evaluated using the modified CGH phosphate buffer in combination with the CSPE. DPV was performed to investigate the relationship between the peak current and concentration of QR due to its higher sensitivity. The DPV responses of QR in different concentrations were recorded as shown in Fig. 4. Table 2 shows the analytical figures of merit of proposed approach. Combination of the high sensitivity of the method with a low noise level resulted in very competitive LODs (S/N=3).

Table 2

Analytical parameters of the proposed methodology.

Parameter	
Regression equation[a]	y= 0.695 + 1.72x
R^2	0.995
Linear range (μM)	0.026-17
LOD (nM)	7.97
LOQ (nM)	26.3
%RSD Potential[b]	1.41
%RSD Intensity[b]	7.49

[a]Regression equation is y=bx+a where y is the amperometric current (μA) and x is the analyte concentration (μM).
[b]n=7

In order to evaluate reproducibility of the method, several CSPEs were tested for the electrochemical measurement of QR. The oxidation peak currents of this analyte decreased continuously, perhaps due to the severe surface sorption and fouling observed. Therefore, the CSPEs were only used for single measurements and the reproducibility

between multiple electrodes was evaluated by the parallel determination of the oxidation peak current of 17 μM of QR. The relative standard deviation (RSD) for peak current was 7.49% (n=7). This acceptable RSD suggested that the reproducibility and the precision of detection were good.

3.6. Interferences effect

The potential interferences on the determination of QR were also evaluated. Representative phenolic compounds commonly present in plant matrices were selected at practical concentrations. The oxidation peak currents of QR were measured individually in pH 7.0 CGH modified phosphate buffer containing the interferents and the peak change in current was then checked. The results indicated that the following molecules did not interfere in the determination of 17 μM QR as the peak current change was <10%: 100 μM Tyrosol, 100 μM Apigenine, 100μM Vanillic acid, 100 μM Oleuropein, 100 μM p-Coumaric acid, 100 μM p-Amino benzoic acid, 100 μM 2,5-dihydroxybenzoic acid, 100 μM Syringic acid, 100 μM 4-hydroxyphenylacetic acid and others non polyphenols like 100 μM Melatonin and 100 μM Triptophan. While 100 μM of Sinapic acid, Galic acid and Rosmarinic acid do interfere with QR determination.

Interestingly, it was discovered that the NIS obtained with QR, also occurs when the developed methodology is applied to other molecules like polyphenols from other families such as Tyrosol, Apigenine, Vanillic acid, Oleuropein, Coumaric acid, and amino acids and hormones like tryptophan and melatonin. This approach could be a valuable tool for biological compound analysis. Nevertheless, further studies are necessary to understand this effect and its applicability.

3.7. Sample analysis

To demonstrate the performance of the proposed method in real samples analysis, the content of QR in onion (yellow, red and green) samples was measured by standard addition method with CGH phosphate buffer at CSPE by DPV. Samples were extracted with both CGH and methanol; the results showed no significant differences at extraction efficiency between both solvents. Thus, CGH was chosen for further extractions. The accuracy of the proposed method was compared traditional HPLC methodology. As can be seen in Table 3, the results obtained by HPLC and by the electrochemical approach were in good agreement and the relative error was <9.5%, indicating that the proposed method is accurate and reliable. In addition, the obtained results demonstrate that the proposed method using CGH phosphate buffer and CSPE is able for the determination of QR in complex food samples. Also, the proposed electrochemical determination method was faster than the chromatographic one, in that it did not demand any previous separation step, and there is an additional advantage of avoiding the consumption of large amounts of toxic organic solvents.

Table 3

Determination of QR in yellow, red and green onions using CGH phosphate buffer and CSPE and HPLC method.

	NADES-CSPE ($\mu g\ g^{-1}$)[a]	HPLC ($\mu g\ g^{-1}$)[a]	Error (%)
Yellow onion	1,75 ± 0,22	1,91 ± 0,08	8,38
Red onion	3,98 ± 0,35	4,39 ± 0,07	9,14
Green onion	ND	ND	-

[a]Values are expressed as mean value±SD

Finally, Table 4 shows a comparison between this method and some previously reported works for determination of QR using electrochemical sensors. The presented method with only a very simple buffer modification and using disposables CSPEs can provide linear range and detection limits comparable with other previous procedures avoiding expensive and time consuming modifications of the electrode. Our system shows acceptable

analytical performance and can be feasible for determination of QR. In addition, compared with other analytical methods such as HPLC, the proposed electrochemical method highlights by its sustainability, simplicity, high sensitivity, good stability, and especially low-cost instrumentation with the additional advantage of being completely portable.

Table 4

Comparison of electrochemical sensors for QR.

Electrode	Modification	Linear Range (μM)	LOD (μM)	Sample	Ref.
GCE	Graphene	0.006-100	0.0036	Apple and Onion	[11]
GCE	Molecularly Imprinted Polymer Incorporated Graphene Oxide	0.6-15	0.048	Apple juice	[31]
CPE	Alumina microfibers	0.025-1.5	0.010	Tea and honeysuckle	[24]
GCE	Graphene nanosheets	0.08-50	0.0011	Human blood, serum and urine	[29]
GCE	electro-deposited grapheme andpolymerized β-cyclodextrin	0.005-20	0.001	Tea and honeysuckle	[30]
CPE	NiO/CNTs and ionic liquid	0.08-400	0.03	Onion, apple and capsule	[32]
GCE	AgNPs-2-aminoethanethiol functionalized graphene oxide	0.01-5	0.0033	Grape wine	[16]
GCE	AuNPs-p-	0.001-0.05	0.0003	Orange and apple	[22]

	aminothiophenol functionalized multi-walled carbon nanotubes			juices	
CSPE	-	0.016-17	0.0079	Onion	This Work

4. Conclusions

In this paper, a new application for Natural Deep Eutectics Solvents was demonstrated. For the first time, Natural Deep Eutectic Solvents have been proved to be enhancer agents for electrochemical detection. A simple and sensitive electrochemical method for QR determination based on NADES electrolyte combined with unmodified screen-printed electrodes was developed. The proposed method exhibited a good performance in terms of sensitivity, detection limit and repeatability. It has proved to be faster and cheaper than other sensors and chromatographic methods available with the additional advantage of avoiding the use of toxic organic solvents besides being completely portable. Furthermore, the approach was successfully applied for the determination of QR in complex plant matrices.

On the other hand, it has been demonstrated that CSPEs electrodes in combination with CGH modified buffer allow the improved detection of QR and the same effect can be achieved with another polyphenols and important biological molecules such as aminoacids and indoleamines. This undoubtedly opens new perspectives for NADES future applications with encouraging novel horizons in sensors development. Noteworthy, that the proposed methodology is entirely green from every point of view, since the extraction and the determination used only NADES as solvents with very good results.

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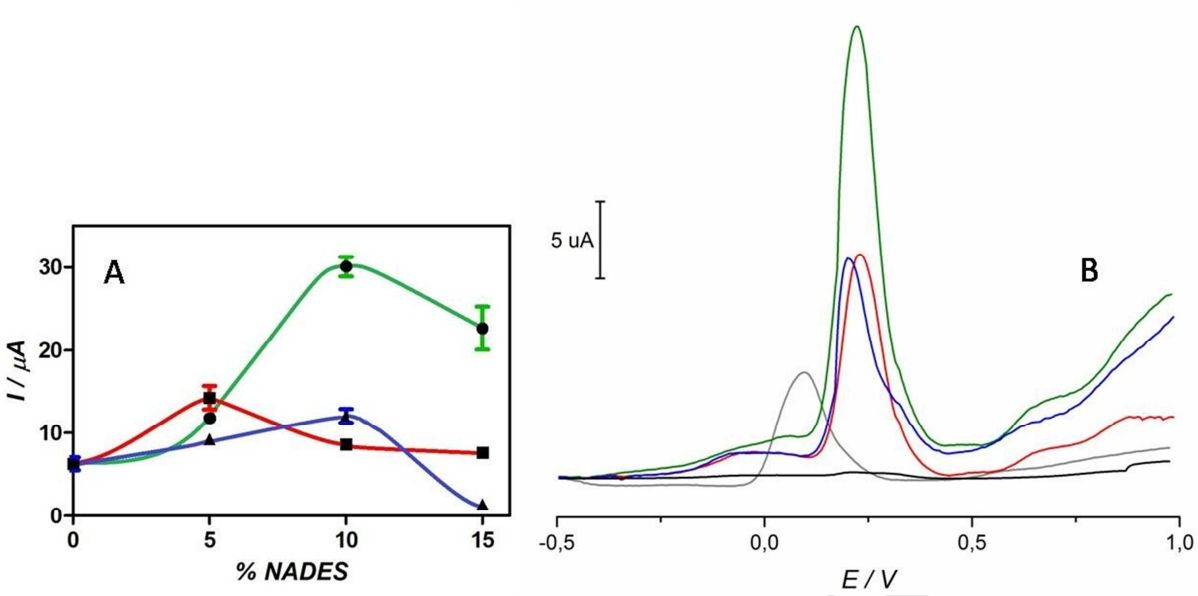
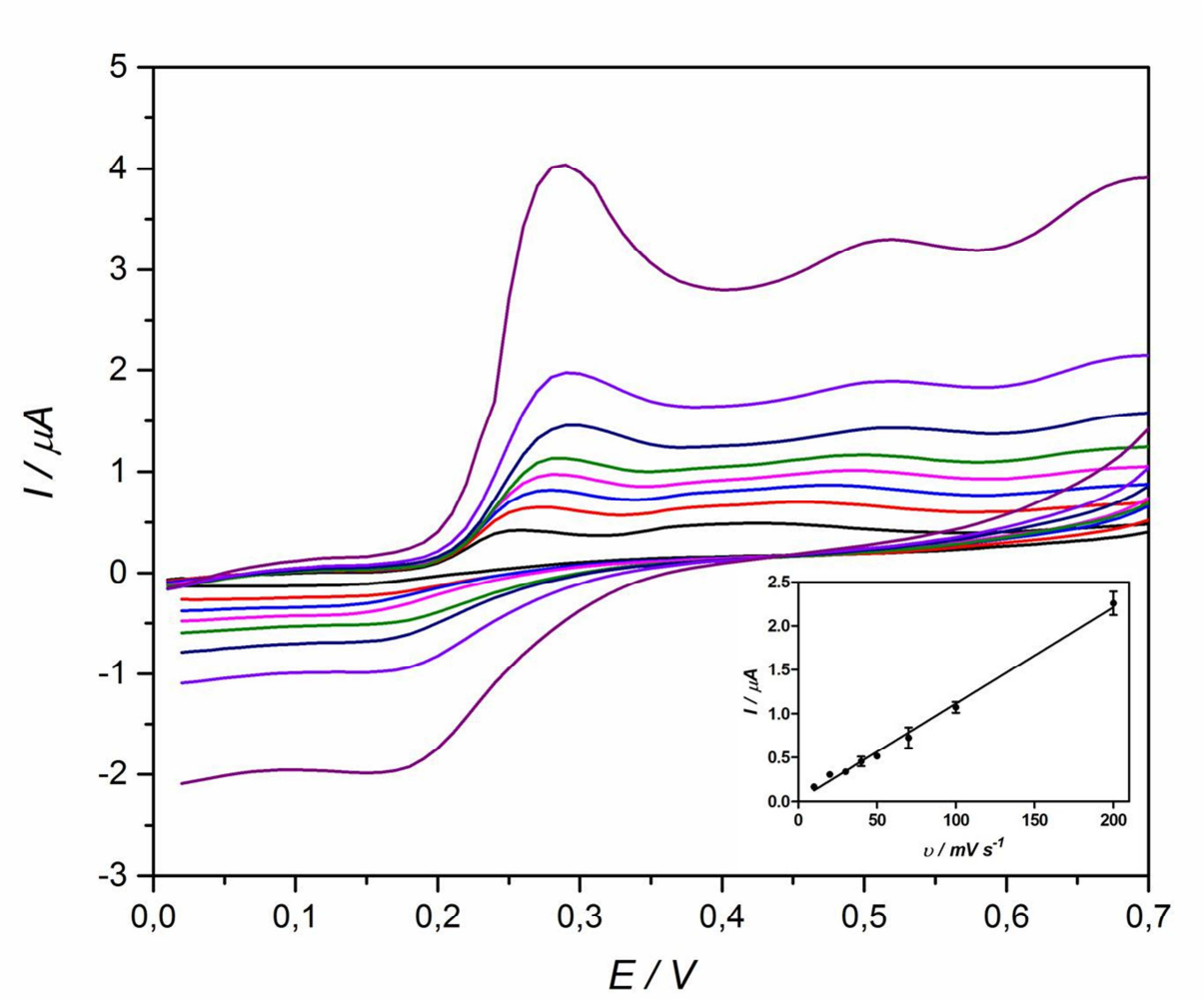
References

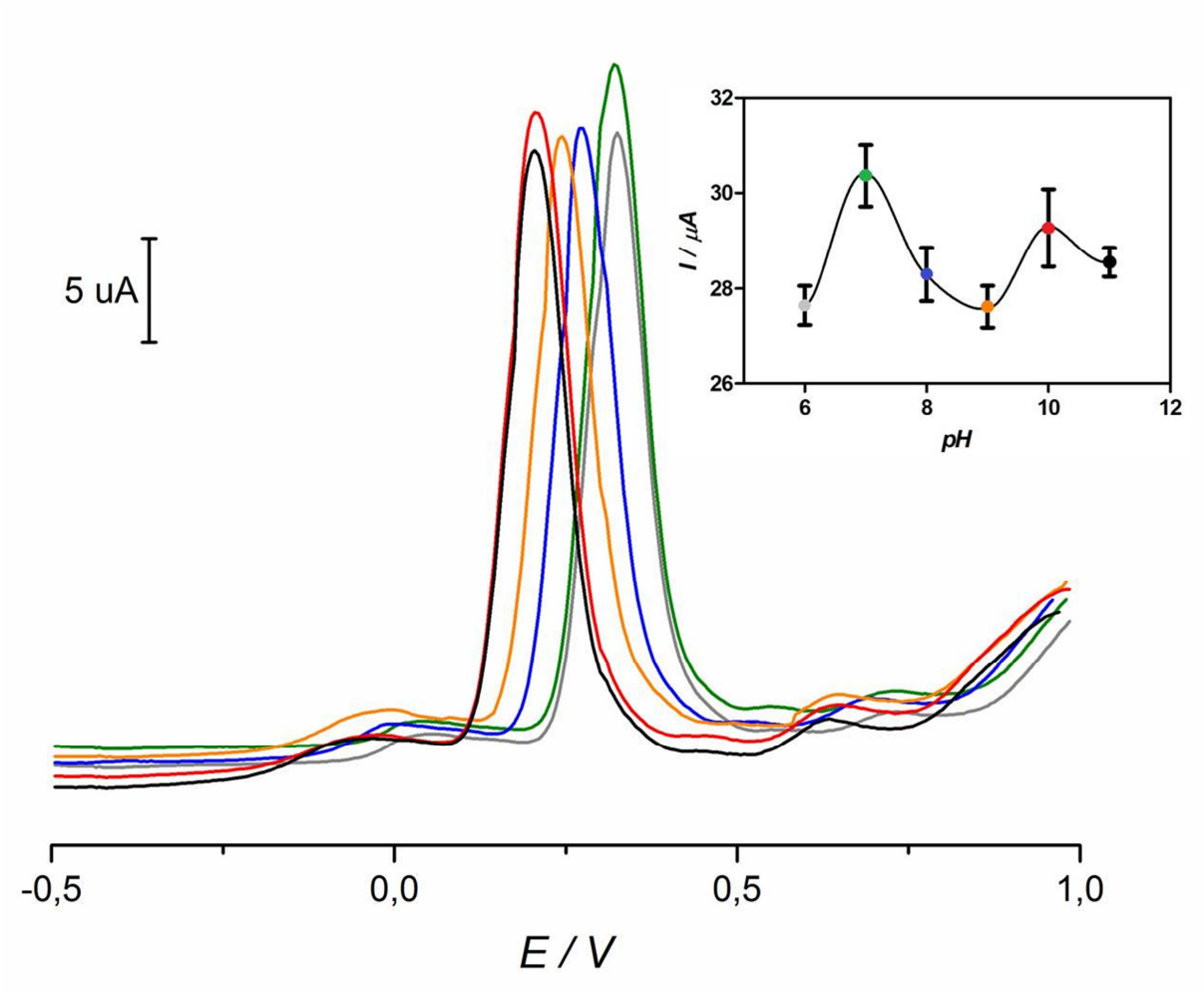
[1] Y.H. Choi, J. van Spronsen, Y. Dai, M. Verberne, F. Hollmann, I.W.C.E. Arends, G.J. Witkamp, R. Verpoorte, Are natural deep eutectic solvents the missing link in understanding cellular metabolism and physiology?, Plant Physiol. 156 (2011) 1701-1705.
[2] M. Espino, M. de los Ángeles Fernández, F.J.V. Gomez, M.F. Silva, Natural designer solvents for greening analytical chemistry, TrAC, Trends Anal. Chem. 76 (2016) 126-136.
[3] Y. Dai, R. Verpoorte, Y.H. Choi, Natural deep eutectic solvents providing enhanced stability of natural colorants from safflower (Carthamus tinctorius), Food Chem. 159 (2014) 116-121.
[4] Y. Dai, G.J. Witkamp, R. Verpoorte, Y.H. Choi, Natural deep eutectic solvents as a new extraction media for phenolic metabolites in carthamus tinctorius L, Anal. Chem. 85 (2013) 6272-6278.
[5] Q. Zhang, K. De Oliveira Vigier, S. Royer, F. Jérôme, Deep eutectic solvents: Syntheses, properties and applications, Chem. Soc. Rev. 41 (2012) 7108-7146.
[6] Y. Dai, J. van Spronsen, G.J. Witkamp, R. Verpoorte, Y.H. Choi, Natural deep eutectic solvents as new potential media for green technology, Anal. Chim. Acta 766 (2013) 61-68.
[7] D. Zielinska, B. Pierozynski, Electrooxidation of quercetin at glassy carbon electrode studied by a.c. impedance spectroscopy, J. Electroanal. Chem. 625 (2009) 149-155.
[8] R. Slimestad, T. Fossen, I.M. Vågen, Onions: A source of unique dietary flavonoids, J. Agric. Food Chem. 55 (2007) 10067-10080.
[9] D. Zielińska, L. Nagels, M.K. Piskuła, Determination of quercetin and its glucosides in onion by electrochemical methods, Anal. Chim. Acta 617 (2008) 22-31.
[10] P. Xiao, F. Zhao, B. Zeng, Voltammetric determination of quercetin at a multi-walled carbon nanotubes paste electrode, Microchem. J. 85 (2007) 244-249.
[11] M. Arvand, M. Anvari, A graphene-based electrochemical sensor for sensitive detection of quercetin in foods, J. Iran. Chem. Soc. 10 (2013) 841-849.
[12] Ž. Nikolovska-Čoleska, L. Klisarova, L. Šuturkova, K. Dorevski, First and second derivative spectrophotometric determination of flavonoids chrysin and quercetin, Anal. Lett. 29 (1996) 97-115.
[13] D.G. Watson, E.J. Oliveira, Solid-phase extraction and gas chromatography–mass spectrometry determination of kaempferol and quercetin in human urine after consumption of Ginkgo biloba tablets, J. Chromatogr. B: Biomed. Sci. Appl. 723 (1999) 203-210.
[14] S.E. Nielsen, L.O. Dragsted, Column-switching high-performance liquid chromatographic assay for the determination of quercetin in human urine with ultraviolet absorbance detection, J. Chromatogr. B: Biomed. Sci. Appl. 707 (1998) 81-89.
[15] M. Careri, L. Elviri, A. Mangia, M. Musci, Spectrophotometric and coulometric detection in the high-performance liquid chromatography of flavonoids and optimization of sample treatment for the determination of quercetin in orange juice, J. Chromatogr. A 881 (2000) 449-460.

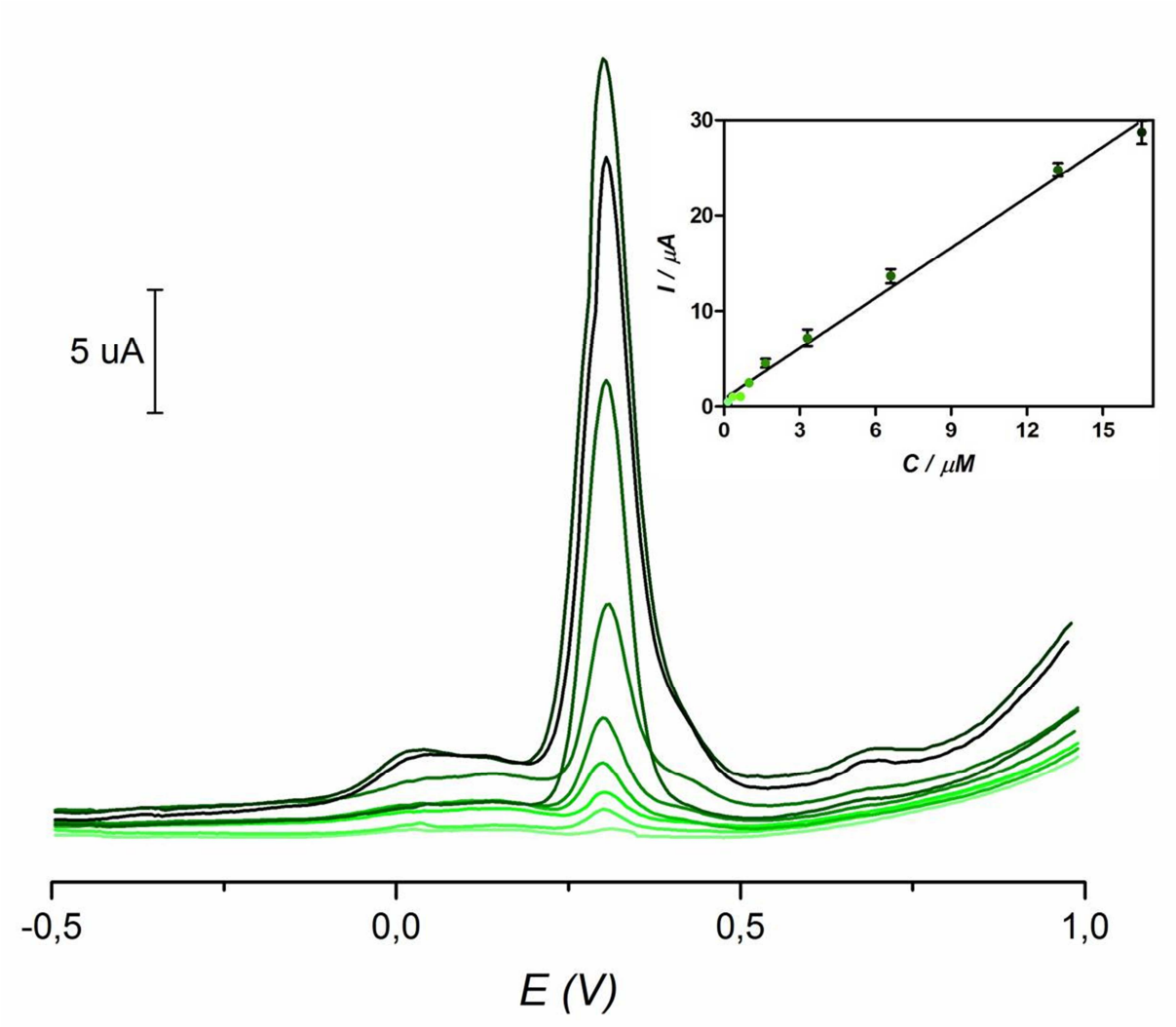
[16] M.L. Yola, V.K. Gupta, T. Eren, A.E. Şen, N. Atar, A novel electro analytical nanosensor based on graphene oxide/silver nanoparticles for simultaneous determination of quercetin and morin, Electrochimica Acta 120 (2014) 204-211.
[17] M.L. Yola, N. Atar, Z. Üstündağ, A.O. Solak, A novel voltammetric sensor based on p-aminothiophenol functionalized graphene oxide/gold nanoparticles for determining quercetin in the presence of ascorbic acid, Journal of Electroanalytical Chemistry 698 (2013) 9-16.
[18] O. Akyildirim, H. Medetalibeyoglu, S. Manap, M. Beytur, F.S. Tokali, M.L. Yola, N. Atar, Electrochemical sensor based on graphene oxide/iron nanoparticles for the analysis of quercetin, International Journal of Electrochemical Science 10 (2015) 7743-7753.
[19] S. Elçin, M.L. Yola, T. Eren, B. Girgin, N. Atar, Highly Selective and Sensitive Voltammetric Sensor Based on Ruthenium Nanoparticle Anchored Calix[4]amidocrown-5 Functionalized Reduced Graphene Oxide: Simultaneous Determination of Quercetin, Morin and Rutin in Grape Wine, Electroanalysis 28 (2016) 611-619.
[20] J.B. He, X.Q. Lin, J. Pan, Multi-wall carbon nanotube paste electrode for adsorptive stripping determination of quercetin: A comparison with graphite paste electrode via voltammetry and chronopotentiometry, Electroanalysis 17 (2005) 1681-1686.
[21] X.Q. Lin, J.B. He, Z.G. Zha, Simultaneous determination of quercetin and rutin at a multi-wall carbon-nanotube paste electrodes by reversing differential pulse voltammetry, Sens. Actuators, B 119 (2006) 608-614.
[22] M.L. Yola, N. Atar, A novel voltammetric sensor based on gold nanoparticles involved in p-aminothiophenol functionalized multi-walled carbon nanotubes: Application to the simultaneous determination of quercetin and rutin, Electrochimica Acta 119 (2014) 24-31.
[23] M. Wang, D. Zhang, Z. Tong, X. Xu, X. Yang, Voltammetric behavior and the determination of quercetin at a flowerlike Co3O4 nanoparticles modified glassy carbon electrode, J. Appl. Electrochem. 41 (2011) 189-196.
[24] Y. Li, W. Huang, Electrode modified with porous alumina microfibers as a highly sensitive electrochemical sensor for quercetin, Anal. Methods 7 (2015) 2537-2541.
[25] Y. Zheng, L. Ye, L. Yan, Y. Gao, The electrochemical behavior and determination of quercetin in choline chloride/urea deep eutectic solvent electrolyte based on abrasively immobilized multi-wall carbon nanotubes modified electrode, International Journal of Electrochemical Science 9 (2014) 238-248.
[26] M. Fouladgar, A new sensor for determination of nalbuphine using NiO/functional single walled carbon nanotubes nanocomposite and ionic liquid, Sensors and Actuators, B: Chemical 230 (2016) 456-462.
[27] L. Daneshvar, G. Rounaghi, Z. E'Shaghi, M. Chamsaz, S. Tarahomi, Electrochemical determination of carbamazepin in the presence of paracetamol using a carbon ionic liquid paste electrode modified with a three-dimensional graphene/MWCNT hybrid composite film, Journal of Molecular Liquids 215 (2016) 316-322.
[28] V.K. Gupta, F. Golestani, S. Ahmadzadeh, H. Karimi-Maleh, G. Fazli, S. Khosravi, NiO/CNTs nanocomposite modified ionic liquid carbon paste electrode as a voltammetric sensor for determination of quercetin, International Journal of Electrochemical Science 10 (2015) 3657-3667.
[29] M. Arvand, M. Anvari, Graphene nanosheets as a sensing platform for amplified electrochemical measurement of quercetin and uric acid in biological fluids, Can. J. Chem. 92 (2014) 1074-1080.
[30] Z. Zhang, S. Gu, Y. Ding, M. Shen, L. Jiang, Mild and novel electrochemical preparation of β-cyclodextrin/graphene nanocomposite film for super-sensitive sensing of quercetin, Biosens. Bioelectron. 57 (2014) 239-244.

[31] S. Sun, M. Zhang, Y. Li, X. He, A molecularly imprinted polymer with incorporated Graphene oxide for electrochemical determination of quercetin, Sensors (Switzerland) 13 (2013) 5493-5506.
[32] V.K. Gupta, F. Golestani, S. Ahmadzadeh, H. Karimi-Maleh, G. Fazli, S. Khosravi, NiO/CNTs nanocomposite modified ionic liquid carbon paste electrode as a voltammetric sensor for determination of quercetin, Int. J. Electrochem. Sci. 10 (2015) 3657-3667.









Natural Deep Eutectic Solvents are enhancers of electrochemical detection for phenolic compounds.

The methodology fully represents the principles of green analytical chemistry.

The approach was successfully applied for the determination of QR in complex plant matrices