Electrochemical analysis of speedball-like polydrug samples

ARTICLE

Electrochemical Analysis of Speedball-like Polydrug Samples

Mats de Jong[a], Anca Florea[a,b], Devin Daems[a,b], Joren Van Loon[a,c], Nele Samyn[d] and Karolien De Wael[*a,b]

Received 00th January 20xx,
Accepted 00th January 20xx

DOI: 10.1039/x0xx00000x

Increasing global production, trafficking and consumption of drugs of abuse cause an emerging threat to people's health and safety. Electrochemical approaches have proven to be useful for on-site analysis of drugs of abuse. However, few attention has been focused on the analysis of polydrug samples, despite these samples causing severe health concerns, certainly when stimulants and depressants are combined, as is the case for Speedball, a mixture of cocaine and heroin. In this work, we provide solutions for the selective detection of cocaine (stimulant) in polydrug samples adulterated with heroin and codeine (depressants). The presence of either one of these compounds in cocaine street samples leads to an overlap with the cocaine signal in square-wave voltammetry measurements at unmodified carbon screen-printed electrodes, leading to inconclusive screening results in the field. The provided solutions to this problem consist of two parallel approaches: (i) cathodic pretreatment of the carbon screen-printed electrode surface prior to measurement in both alkaline and neutral conditions; (ii) electropolymerization of orthophenylenediamine on graphene modified carbon screen-printed electrodes prior to measurement in neutral conditions. Both strategies allow simultaneous detection of cocaine and heroin in speedball samples as well as simultaneous detection of cocaine and codeine. Implementing these strategies in portable devices holds great potential for significantly improved accuracy of on-site cocaine screening in polydrug samples.

Introduction

Drugs of abuse are becoming a globally greater threat every year. The United Nations estimated that 275 million people used drugs in 2017, which is an increase of 32 % compared to the situation in 2007. More alarmingly, global death rates directly caused by drug (ab)use have been dramatically rising from 105,000 deaths in 2000 to 585,000 in 2017, emphasizing the urgent problem of drug (ab)use.[1, 2] Polysubstance (ab)use has a widespread pattern, which is observed in different regions and has emerged lately due to both the increasing range of drugs available and the willingness of different groups of (young) people to experiment with illicit psychoactive substances.[3] The most frequently reported drug combination among European patients entering treatment was heroin (opioid) and cocaine (stimulant), which is also referred to as speedball.[4] Opioids depress the central nervous system while cocaine stimulates it. Combined use of these induces an alleviation of their own specific side-effects and severity of withdrawal symptoms,[1, 5, 6] but also causes additional aggravated health risks.[4, 6-8] For instance, the negative cardiovascular effects of cocaine are amplified when it is co-administered with opioids. Moreover, cocaine can initially mask the sedative effects of opioids, therefore increasing the risk of a later overdose.[1, 3]

Color tests are mainly used for on-site drug screenings. Usually, these tests are selective for one certain drug, but have a poor accuracy, generating many false positive and false negative results. There are also a few variants to detect multiple drugs in one test, but given the problems already existing with the single compound tests, the accuracy of the multi-drug tests are even lower.[9-12] Therefore, identification of multiple drugs in polydrug samples using these tests is nearly impossible on-site. A different method for on-site detection of (poly)drug samples, such as speedball, is, thus, necessary.

Electrochemical approaches offer attractive options for the on-site analysis of drugs of abuse. They are accurate, low-cost and robust. The equipment can easily be miniaturized to be applied in field settings for portable analysis.[13] Several amperometry and voltammetry approaches were proposed in the literature concerning the analysis of drugs of abuse. However, many of these publications mainly focus on the detection of one compound of interest and involve extensive electrode modification procedures.[14-19] Voltammetry approaches are ideal for the detection of multiple compounds, and several publications reported on the simultaneous detection of a certain drug and its main cutting agents or metabolites.[14, 20-23] However, the simultaneous electrochemical detection of

[a] AXES Research Group, University of Antwerp, Groenenborgerlaan 171, 2020 Antwerp, Belgium. E-mail: karolien.dewael@uantwerpen.be
[b] NANOlab Centre of Excellence, University of Antwerp, Groenenborgerlaan 171, 2020 Antwerp, Belgium.
[c] Product Development Research Group, University of Antwerp, Ambtmanstraat 1, 2000 Antwerp, Belgium.
[d] Drugs and Toxicology Department, National Institute for Criminalistics and Criminology, Vilvoordsesteenweg 100, 1120 Brussels, Belgium.
Electronic Supplementary Information (ESI) available: see DOI: 10.1039/x0xx00000x

cocaine and heroin or codeine in polydrug samples has not been explored yet. Garrido *et al.* discussed this topic briefly but rendered the simultaneous voltammetric detection of cocaine and heroin impossible because of convolution of the peaks.[24]

Recently, in our previous work, the performance of electrochemical approaches showed a great advantage over the use of classical color tests for the on-site screening of cocaine (COC).[9] Unfortunately, mixtures containing heroin (HER) or codeine (COD) (both opioids) presented problems in electrochemical COC detection due to an overlap of their signals with the signal of COC. In this work, we evaluate pretreatment strategies and polymer platforms towards the simultaneous detection of stimulant (COC) and depressant (HER or COD) for the purpose of polydrug screening.

Experimental

Cocaine HCl, heroin HCl and codeine HCl standards were purchased from Lipomed (Arlesheim, Switzerland). Potassium monophosphate, potassium chloride, potassium hydroxide, o-phenylenediamine (OPD), acetic acid and sodium acetate were purchased from Sigma-Aldrich (Overijse, Belgium). A solution of 20 mM phosphate buffer containing 100 mM KCl was always used as supporting electrolyte. All aqueous solutions were prepared using double distilled water. The reagents were of analytical grade and used without further purification. The pH of the freshly made buffer solutions was adjusted by adding 100 mM KOH solution, monitoring the pH using a CyberScan 510 pH-meter from Eutech Instruments (Landsmeer, The Netherlands) connected to a HI-1131 glass bodied pH electrode from Hanna Instruments (Bedfordshire, United Kingdom).

Electrochemical measurements, including square wave voltammetry (SWV) and cyclic voltammetry (CV), were carried out with an Autolab PGSTAT302N potentiostat/galvanostat controlled by NOVA 2.1 software from Metrohm (Herisau, Switzerland). Disposable carbon ItalSens IS-C Screen Printed Electrodes (SPE) were purchased from PalmSens (Utrecht, The Netherlands) and were used during all electrochemical measurements apart from the polymer modified measurements. These SPEs have a diameter of 3 mm. Graphene modified DropSens DRP-110GPH carbon SPEs were purchased from Metrohm (Antwerp, Belgium) and were used for all polymer modified experiments since graphene improves the conductivity of the polymer layers and thus the sensitivity of detection, offering at the same time a higher surface area with more possible binding sites for the analytes.[25] These SPEs have a diameter of 4 mm. Both SPEs contain a silver pseudo reference electrode and a carbon counter electrode.

SWV measurements were carried out with a step potential of 5 mV, amplitude of 25 mV and frequency of 10 Hz. The used potential window was from -0.1 V to +1.3 V. These parameters correspond to a total analysis time of 28 s. All results obtained by SWV were presented after baseline correction using the mathematical algorithm “moving average” (peak width = 1) contained within NOVA software, which improves the visualization and identification of the peaks over the baseline. All electrochemical experiments were performed at 22 °C.

Poly(orthophenylenediamine) (OPD) was electropolymerized on graphene-SPE by performing cyclic voltammetry (parameters: -0.3 to +0.8 V, 5 cycles, 50 mV/s; 100 µL OPD solution 1mM in acetate buffer pH 5.2 containing 0.1M KCl). The modified electrodes were incubated for 10 min with 100

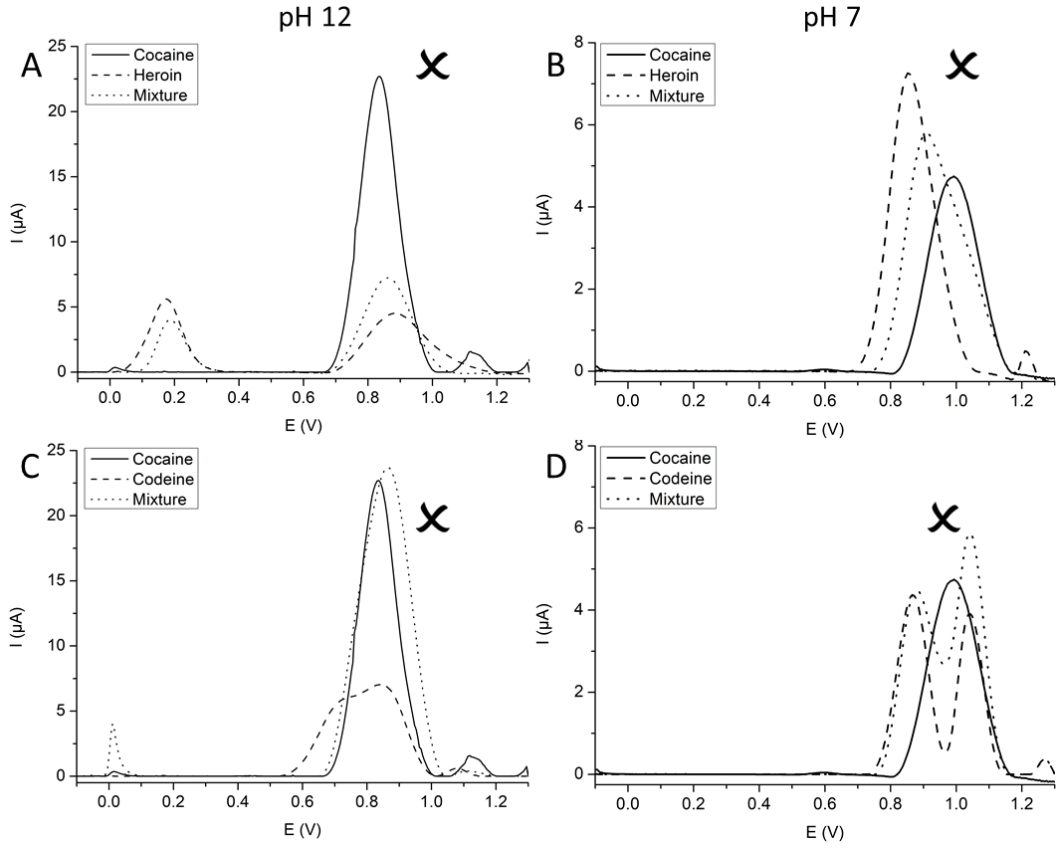

Figure 1: SWVs (pH 12) of 1 mM COC, 0.5 mM interfering agents HER (A) and COD (B), and a binary mixture (1:0.5 mM) of COC and each interfering agent. ItalSens carbon SPEs were used.

µL solutions of COC (0.5 mM), COD/HER (0.5 mM), and COC with COD/HER (0.5:0.5mM) in phosphate buffer pH 7, washed and subjected to SWV in phosphate buffer pH 7.

Results and discussion

Impossibility to detect COC in the presence of HER or COD

Extensive research has been performed on the detection of COC in the presence of its cutting agents and a few other drugs of abuse in previous work. Strategies were proposed and applied using pH 7 and pH 12 phosphate buffer. It was shown that the pH 12 approach was superior to the pH 7 method, due to electrochemical peak suppression of COC in pH 7, in the presence of several cutting agents.[9, 26, 27] However, two compounds, i.e. COD and HER, hinder the detection of COC in street samples in pH 12 conditions as well (chemical structures shown in Figure S-1 in the *Supporting Information*).

Figure 1 displays the electrochemical signal for COC (solid line), its peak potential situated at +0.83 V (pH 12) or +0.99 V (pH 7) is related to the oxidation of the tertiary amine group to an iminium ion, releasing two electrons and one proton in the process. This ion is then easily converted further into amine group, creating norheroin in the case of HER and norcodeine in the case of COD.[13, 15, 16] One cannot distinguish between COC and HER or COD based on the signal of the mixture (Figure 1 dotted lines), although the total fingerprint of HER (in pH 12) and COD is different from COC with the additional signal at +0.18 V for HER and +0.72 V for COD, respectively. These signals are related to the presence of a phenolic group in 6-monoacetylmorphine and morphine, in-situ generated by the fast hydrolysis of HER[13] in alkaline media and to the presence of the 6-hydroxy group in the COD structure, which is oxidized to a keton after the release of two electrons and a proton.[13, 15, 16, 28] While these distinctions allow to discriminate between the drugs, they do not provide a solution for analysis of mixtures with COC, i.e. polydrug samples. Therefore, alternative strategies were discovered, proposed and evaluated.

Electrochemical pretreatment strategies

Cathodic electrochemical pretreatment is a straightforward, controllable and reproducible way to pretreat carbon electrodes, also cleaning them during this process.[29] Indeed, the electrocatalytic properties of carbon-based electrodes can

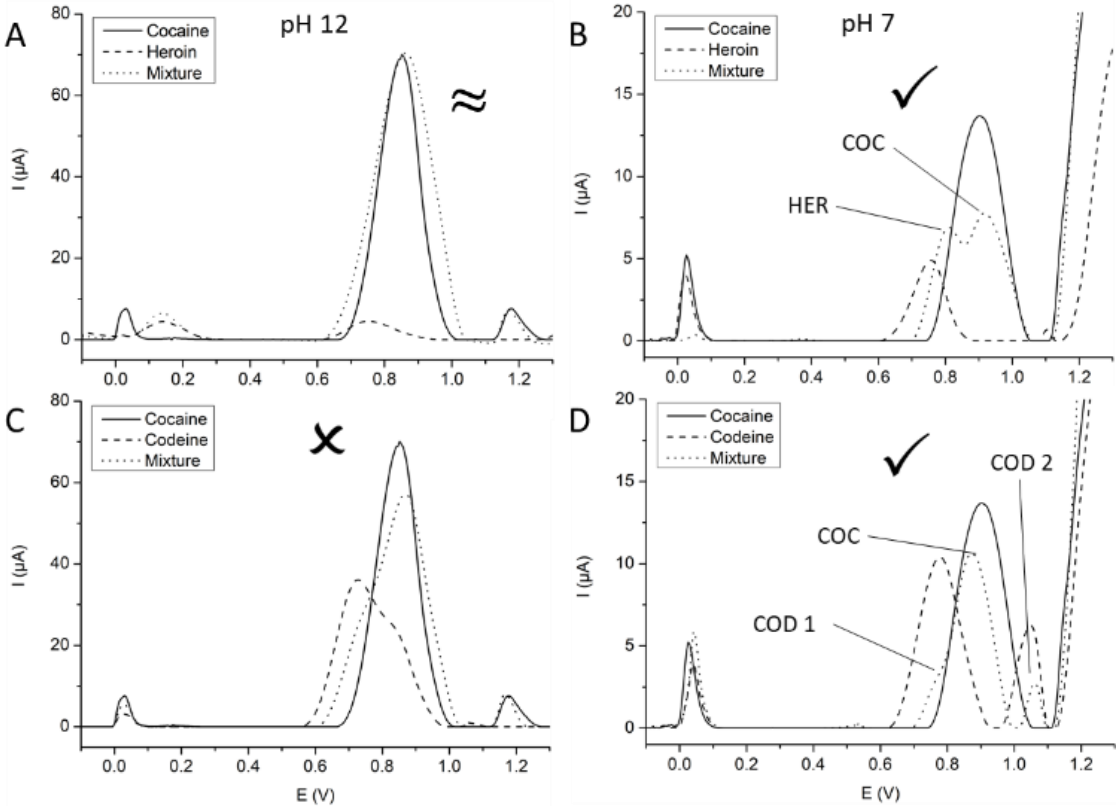

Figure 2: SWVs of 1 mM COC (solid line), 0.5 mM interfering agents (dashed line), and a binary mixture (1:0.5 mM) of COC and interfering agent (dotted line) for interfering agents HER (A and B) and COD (C and D) after a cathodic pretreatment at -0.8 V for 360 s in pH 12 (A and C) and pH 7 (B and D) buffer. ItalSens carbon SPEs were used.

norcocaine in the presence of water.[17, 27] The COC signal overlaps with the signal of HER (Figure 1A and 1B dashed line) and COD (Figure 1C and 1D dashed line) in both pH 7 and pH 12 conditions. HER and COD contain a similar tertiary amine group, which is after oxidation converted to a secondary

be improved by electrochemical pretreatment, facilitating the electron transfer exchange between the analyte and the electrode surface.[26, 30-32] Therefore, we studied the effect of an electrochemical pretreatment of the electrode surface on the detection of COC in mixture with HER and COD.

The effect of a cathodic surface pretreatment on the aforementioned polydrug samples was evaluated by applying various potentials for various times, both in pH 12 and pH 7 buffer solutions. Pretreatment potentials ranging from -0.4 to -1.2 V were studied, while the time was varied from 5 to 360 s. Changing these parameters allows to find optimal conditions for each specific case, since longer pretreatment times will increase the cleaning effect and the amount of reactive surface defects created. In addition, performing pretreatment at more negative potentials will amplify the intensity of these effects.[26, 29-32]

It was observed for a pure COC solution that the COC peak potential values deviate from their original values when a pretreatment is performed (Figure S-2 in the *Supporting Information*). These potential shifts must be taken into account for the detection in binary mixtures of COC and the other compounds.

For a 2:1 binary mixture of COC with HER, the pretreatment approach does not allow the selective detection of COC using most of the studied parameters. However, using a pretreatment potential of -0.8 V or -1.2 V combined with a pretreatment time of 360 s in pH 12 conditions does show a significant increase of the COC current compared to the HER current (Figure 2A) in respect to the situation when no pretreatment is performed (Figure 1A). Quantitatively, the COC signal intensity has increased by over three times (70.1 µA vs 22.7 µA), whereas the main HER signal intensity did not change after pretreatment (4.5 µA with and without pretreatment), making the pretreatment approach specifically more sensitive for COC. Based on this significant difference in current response and the obtained difference in peak potential for the COC (+0.85 V) and HER (+0.75 V) signal, one could proof the presence of COC in the mixture (dotted line). However, this approach does not deliver definite proof on the presence or absence of HER in the mixture.

Definite proof for both COC and HER presence could be achieved using pH 7 pretreatment conditions: using -0.8 or -1.2 V combined with a pretreatment time of at least 60 s is already sufficient. In addition, -0.6 V could also be used as pretreatment potential to simultaneously detect the compounds, as long as the pretreatment time is higher than 120 s. These findings are shown in Figure S-3 in the *Supporting Information*. The remaining softer conditions always result in a sustaining overlap of the COC signal, comparable to the reference system without pretreatment. In the case of COD, the electrochemical pretreatment approach does not work for pH 12 conditions. However, a single solution is found for pH 7 at a pretreatment potential of -0.8 V for 360 s. Optimal conditions were acquired both in HER and COD mixtures using pretreatment parameters -0.8 V and 360 s in pH 7. The SWVs using these optimal conditions are shown in Figure 2B and 2D for pure COC (solid lines), pure HER or COD (dashed lines) and mixtures of COC with HER or COD (dotted lines). In one case (mixture of COC and COD in pH 12, Figure 2C), the selective detection of COC remains impossible.

In summary, this data shows that using -0.8 V and 360 s as pretreatment parameters provides the best solution for all cases, providing stable, intense and (using pH 7) distinguishable signals for COC in mixtures.

Polymer strategies

Alternatively, the use of functionalized electro-active polymers might improve the detectability of COC in presence of interfering agents such as HER and COD. Covering the electrode surface with such a layer changes the interactions between the compounds in the measuring solution and the electrode surface. Moreover, a polymer with specific functional groups can induce enhanced binding (adsorption) affinity towards COC, resulting in an improved intensity of the SWV signal.

Two promising electro-active monomer materials for COC, i.e. o-phenylenediamine and p-aminobenzoic acid (PABA) (Figure S-4 in the *Supporting Information*), were selected, electropolymerized and optimized by Florea *et al.*.[33] In this work, OPD is studied because this polymer does, in contrast to PABA, not contain any oxygen-containing functional groups. It holds only two primary amine groups in the monomer, which are converted to secondary amine and tertiary amine groups during the electropolymerization process.[34, 35] Here,

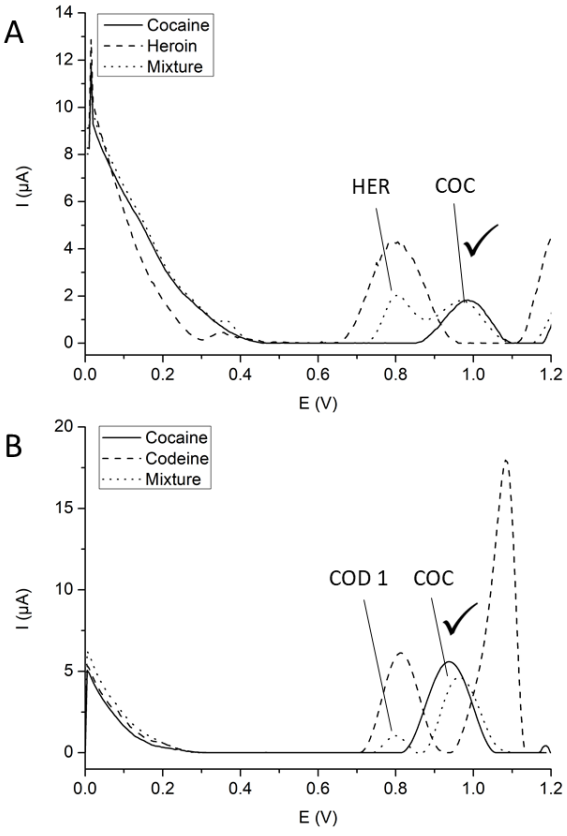

Figure 3: SWVs (pH 7) of 0.5 mM COC (solid line), 0.5 mM interfering agents (dashed line), and a binary mixture (0.5:0.5 mM) of COC and interfering agent (dotted line) for interfering agents HER (A) and COD (B) on a OPD-GPH modified carbon electrode (DropSens).

interaction between the amine groups is the reason for affinity of COC towards the polymer layer. HER and COD possess the same tertiary amine group, but these structures are a lot

bulkier and more rigid, causing this group to be more shielded. Furthermore, both HER and COD contain more accessible ester groups compared to COC, which would cause strong interference using PABA. Therefore, OPD is a more suitable polymer layer for COC detection in the presence of HER and COD.

The SWV results for mixtures of COC with HER and COD on OPD modified electrodes are shown in Figure 3, whilst the CVs showing the successful electropolymerization process of OPD are shown in Figure S-5 of the *Supporting Information*, yielding thin and transparent polymer films deposited on the electrode surface. Figure 3 (dotted line) shows the COC signal is clearly detectable and distinguishable from both the HER and COD signal when using the OPD-modified SPE. The use of the OPD layer causes the COC signal to be more prominently present in mixture compared to the HER and COD signals. This indicates a competition is occurring at the surface of the OPD layer between COC and the other compounds, but that COC tends to adsorb stronger onto the layer. This is also shown by the difference in relative intensities of the signals. Figure 3 shows the HER and COD signals lose, respectively, over 50 % and over 80 % of their intensities (dotted line) compared to their intensity in pure solution (dashed line). The intensity of the COC signal remains, however, constant.

Conclusions

We demonstrated two suitable strategies to allow selective detection of COC in the presence of HER and COD adulterated polydrug samples. The first solution was found in the cathodic pretreatment of the SPE's in both alkaline and neutral conditions, allowing either complete distinction of the relevant signals (in pH 7) or suppression of the adulterant signals (in pH 12 for HER). The use of a polymer layer, i.e. OPD, provided a second effective strategy to allow COC detection in the presence of HER and COD in pH 7 conditions. Implementing these strategies in portable electrochemical devices assures greatly improved accuracy for COC on-site screening in polydrug samples and provides suitable platforms for other electrochemical applications.

Conflicts of interest

There are no conflicts to declare.

Acknowledgements

This work was supported by IOF-SBO and IOF-POC from University of Antwerp, Antwerp, Belgium; and VLAIO IM [HBC.2019.2181], Brussels, Belgium.

Notes and references

1 United Nations Office on Drugs and Crime, *World Drug Report 2019*, Vienna, Austria, 2019.
2 United Nations Office on Drugs and Crime, *World Drug Report 2007*, United Nations Piblication, 2007.
3 European Monitoring Center for Drugs and Drug Addiction, *Polydrug Use: Patterns and Responses*, Luxembourg, 2009.
4 L. P. Pattison, S. McIntosh, E. A. Budygin and S. E. Hemby, *J. Neurochem.*, 2012, **122**, 138-146.
5 F. Leri, J. Bruneau and J. Stewart, *Addiction*, 2003, **98**, 7-22.
6 K. A. Trujillo, M. L. Smith and M. M. Guaderrama, *Pharmacol. Biochem. Behav.*, 2011, **99**, 451-458.
7 United Nations, *World Drug Report 2016*, Vienna, Austria, 2016.
8 A. K. Lamonica and M. Boeri, *J Ethnogr Qual Res*, 2012, **6**, 160.
9 M. de Jong, A. Florea, J. Eliaerts, F. Van Durme, N. Samyn and K. De Wael, *Analytical Chemistry*, 2018, **90**, 6811-6819.
10 Y. Tsumura, T. Mitome and S. Kimoto, *Forensic Sci.Int.*, 2005, **155**, 158-164.
11 A. Deakin, *Microgram Journal*, 2003, **1**, 40-43.
12 M.-J. Binette and P. Pilon, *Microgram Journal*, 2013, **10**, 8-11.
13 J. Garrido, C. Delerue-Matos, F. Borges, T. R. A. Macedo and A. M. Oliveira-Brett, *Anal. Lett.*, 2004, **37**, 831-844.
14 L. S. de Oliveira, M. A. Balbino, M. M. T. de Menezes, E. R. Dockal and M. F. de Oliveira, *Microchem J.*, 2013, **110**, 374-378.
15 J. Garrido, C. Delerue-Matos, F. Borges, T. R. A. Macedo and A. M. Oliveira-Brett, *Electroanalysis*, 2004, **16**, 1427-1433.
16 J. Garrido, C. Delerue-Matos, F. Borges, T. R. A. Macedo and A. M. Oliveira-Brett, *Electroanalysis*, 2004, **16**, 1497-1502.
17 M. T. FernandezAbedul and A. CostaGarcia, *Anal. Chim. Acta*, 1996, **328**, 67-71.
18 A. Navaee, A. Salimi and H. Teymourian, *Biosens. Bioelectron.*, 2012, **31**, 205-211.
19 N. Tavakkoli, N. Soltani and F. Mohammadi, *RSC Adv.*, 2019, **9**, 14296-14301.
20 M. de Jong, N. Sleegers, J. Kim, F. Van Durme, N. Samyn, J. Wang and K. De Wael, *Chem. Sci.*, 2016, **7**, 2364-2370.
21 J. M. Freitas, D. L. O. Ramos, R. M. F. Sousa, T. Paixao, M. H. P. Santana, R. A. A. Munoz and E. M. Richter, *Sens. Actuator B-Chem.*, 2017, **243**, 557-565.
22 L. Asturias-Arribas, M. A. Alonso-Lomillo, O. Dominguez-Renedo and M. J. Arcos-Martinez, *Anal. Chim. Acta*, 2014, **834**, 30-36.
23 A. Florea, J. Schram, M. de Jong, J. Eliaerts, F. Van Durme, B. Kaur, N. Samyn and K. De Wael, *Analytical Chemistry*, 2019, **91**, 7920-7928.
24 J. Garrido, M. P. M. Marques, A. M. S. Silva, T. R. A. Macedo, A. M. Oliveira-Brett and F. Borges, *Anal. Bioanal. Chem.*, 2007, **388**, 1799-1808.
25 R. Z. Zhang and W. Chen, *Biosens. Bioelectron.*, 2017, **89**, 249-268.
26 M. de Jong, A. Florea, A. M. de Vries, A. L. N. van Nuijs, A. Covaci, F. Van Durme, J. C. Martins, N. Samyn and K. De Wael, *Analytical Chemistry*, 2018, **90**, 5290-5297.
27 M. de Jong, N. Sleegers, A. Florea, J. Van Loon, A. L. N. Van Nuijs, N. Samyn and K. de Wael, *Analytical Chemistry*, 2019, **91**, 15453-15460.
28 D. A. Barrett, A. L. P. Dyssegaard and P. N. Shaw, *J. Pharm. Pharmacol.*, 1992, **44**, 606-608.
29 Y. X. Gao, D. Wu, Y. X. Yang, W. J. Wang, S. Y. Xie, K. K. Shiu and K. Shi, *J. Electroanal. Chem.*, 2016, **783**, 90-99.
30 H. Zhang, S. Li, F. H. Zhang, M. X. Wang, X. C. Lin and H. X. Li, *J. Solid State Electrochem.*, 2017, **21**, 735-745.
31 X. B. Hu, W. H. Zheng and R. F. Zhang, *J. Solid State Electrochem.*, 2016, **20**, 3323-3330.
32 A. Rana and A. N. Kawde, *J. Chin. Chem. Soc.*, 2016, **63**, 668-676.
33 A. Florea, T. Cowen, S. Piletsky and K. De Wael, *Talanta*, 2018, **186**, 362-367.

34 V. Branzoi, L. Pilan and F. Branzoi, *Mol. Cryst. Liquid Cryst.*, 2004, **416**, 61-72.
35 I. Losito, F. Palmisano and P. G. Zambonin, *Analytical Chemistry*, 2003, **75**, 4988-4995.

