Anal. Bioanal. Electrochem., Vol. 14, No. 9, 2022, 891-903

Analytical & Bioanalytical Electrochemistry

2022 by CEE
www.abechem.com

Review

Electroanalytical Techniques used in Determination of Ethinylestradiol

Leila Hajiaghababaei

Department of Chemistry, Yadegar-e-Imam Khomeini (RAH) Shahre Rey Branch, Islamic Azad University, Tehran, Iran

*Corresponding Author, Tel.: + +982133999022

E-Mail: lhajiaghababaei@iausr.ac.ir

Received: 17 May 2022 / Received in revised form: 30 August 2022 / Accepted: 5 September 2022 / Published online: 30 September 2022

Abstract- Ethinylestradiol (EE2) is classified as an estrogenic pharmaceutical compound. It is an agonist of the estrogen receptors and is approved for treating estrogen deficiency and osteoporosis prophylaxis in the short term when a choice of medicines is limited. EE2 is administered for the treatment of female hypogonadism and menstrual disorders. This strong estrogen is known to cause nausea, fluid retention, and thrombosis as side effects. It is, therefore, necessary to monitor EE2 levels in biological, environmental, and pharmaceutical samples. Various ultra-sensitive analytical methods were introduced during the last decades but modified electrochemical methods have great attention due to their undeniable properties. Here, all electrochemical analytical methods used for the determination of EE2 are considered and discussed.

Keywords- Ethinylestradiol; Estrogen medication; Electrochemical determination; Sensor; Biosensor

1. INTRODUCTION TO ETHINYLESTRADIOL

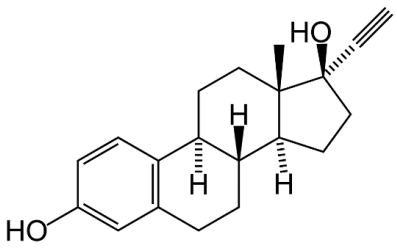
Steroidal estrogens, also known as, female reproductive hormones influence growth, reproduction, and sexual behaviors, and are known to be the cause of the development of

female secondary sex characteristics. The hormones offer advantages like lowering the risk of heart attacks and osteoporosis in women. Analysis of estrogen content of urine samples is a key tool in monitoring women's health.

Further to what was said, estrogens are also administered as drugs in conditions such as menopausal hormone therapy or hormone-based birth control [1–3].

Estrogens are categorized into natural (estrone (E1), estradiol (E2), and estriol (E3)) or synthetic (ethinylestradiol called EE2) hormones [4], among which E2 is the most important, due to its key role in the development and maintenance reproductive tissues in women, animal growth promotion and milk yield increment. E2 is produced by all mammalian species, mainly by the ovaries and partially by the adrenal cortex and the testes. Age and menstrual cycle determine normal E2 levels in serum, plasma, saliva, and hair, which constitute valuable information in the clinical endocrinological investigation of women while studying ovarian function or early pregnancy, treating hormonal infertility, or diagnosing tumors [1-4]. Estradiol deficiency especially that of E2 can lead to hyperandrogenism, osteoporosis, menopausal symptoms, and breast cancer in older women [5-7].

17α−Ethinylestradiol (EE2), Scheme 1, is a low-cost and soluble synthetic form of estrogen female hormone used as medication. It is derived from the natural estrogen E2 which is a common ingredient of oral birth control pills. It was first synthesized in 1938 and is one of the widely prescribed oral contraceptives for women in the world to prevent ovulation, implantation, and therefore, pregnancy. EE2 is readily absorbed upon oral administration, reaching a peak concentration in plasma within 1 to 2 hours. EE2 is also used to reduce the symptoms of menopause, to treat polycystic ovarian syndrome or prostate and breast cancers, in physiological hormonal replacement, or in alopecia lotions [8]. Although E2 and EE2 are both quickly absorbed orally, E2 is readily inactivated by the liver, while the EE2 molecule, due to the substitution of the hydrogen on carbon−17 with a –CCH is very resistant against degradation by the liver, which has opened horizons for developing oral contraceptives.



Scheme 1. Ethinylestradiol (EE2)

2. IMPORTANCE OF DETERMINATION OF EE2

E2 is a non-synthetic high potent endocrine disruptor compound (EDC), which is regarded as an environmental pollutant, even at ppt levels. It can enter water systems directly from

human or animal urine or feces. The compound is known to disturb the endocrine system, affecting the reproductive, immune, and cardiovascular systems of species [9-12]. Various studies have illustrated that EDCs can mimic natural hormones or meddle with the function of endogenous hormones, causing different conditions such as male reproductive system abnormalities [13-20].

Since E2 is an EDC, EE2 which is a derivative of E2 family can be also an EDC. Even when EE2 concentration is low as 0.1 ng L^{-1}, it leads to feminization in some male wild fish [18].

In the face of grave concern about the occurrence of EE2 in water samples, there exist a limited number of reports on its electrochemical determination in aqueous media [21,22].

Regardless of their therapeutic properties, EE2 can cause serious side-effects such as cardiovascular, dermatological, gastrointestinal, genitourinary system, hematological, hepatic, metabolic, nervous system, ocular, oncologic, and psychiatric disorders [23]. EE2 is also suspected as a female carcinogen, leading specifically to breast, endometrial, and ovarian cancers. Research studies have illustrated that exposure to estrogenic compounds even at very low levels (ng/L) can cause serious disorders including infertility, development of tumors, reproductive alteration, and feminization of waterborne organisms [24-26].

While the level of EE2 in birth control medications has been lowered to evade potential side effects, the large global consumption rates by women have increased the chance of occurrence in the environment, which can pose serious risks to humans and wildlife [27]. Replacement of C-17 with an acetylene group releases large quantities of the compounds in the environment since the undegraded hormones are discharged into the surface waters by humans and animals via municipal and industrial sewage, as well as through agricultural [28] and aquacultural wastes [29]. E2 and EE2 are released as glucuronide and sulfate conjugated forms, yet microbial processes can deconjugate them, allowing for their biotransformation into active estrogens in the environment [30-33].

Oral contraceptives were first introduced in 1960 in the US. The first commercial sample contained 150 mg/day of mestranol, which changed to EE2 through metabolic reactions. The majority of OCs available contain EE2 below a quarter of that dose. This is to address the concern arising from the results of studies arguing that lower levels of estrogen decrease the chances of cardiovascular disease [34,35] while having almost similar contraceptive efficiency. Low-dose OC containing 50µg of estrogens was introduced in the 1970s, while the commercial product containing 20µg of EE entered the market in 1973 for women at high cardiovascular risk [36].

The amount of estrogen in an OC should be determined based on a tradeoff since although higher doses more satisfactorily support the endometrium, leading to better cycle control, they bring about bloating, breast tenderness, and nausea, acting as strong predictors of adherence to routine administration by a woman [37,38].

Due to the importance of estrogenic compounds, various analytical methods have been aimed at detecting natural and synthetic estrogens in biological fluids and water samples in the past decade [39-44]. Among all of them, publications reporting the determination of synthetic estrogen (EE2) are scarce. The most common technique, which is used to detect EE2 is HPLC [41,43,45] using UV, fluorescence, electrochemical, MS, or MS/MS detectors. Other techniques are GC−MS [46,47], microextraction [48-51], fluorimetry [52] and molecular imprinting [40]. Urinary E2 levels are clinically analyzed using biological assays [53-56], like enzyme- or radio-immunoassay. Electroanalysis [21,39,57-63] based on various modified electrodes is a robust alternative with key advantages like simplicity, cost-effectiveness, satisfactory sensitivity and selectivity, portability, low response time, and a capability to directly determine the target species in complex matrices. Satisfactory results were carried out by choosing suitable working electrodes.

The development of electrochemical sensors and biosensors as tools for the detection and quantification EE2 in different samples has attracted some researchers in recent years. Here we are going to consider these reports.

3. ELECTROCHEMICAL DETERMINATION OF EE2

The phenolic -OH group of E2 and EE2, which can be electrochemically oxidized or reduced, using electrodes, bestows these species with electroactivity. EE2 also includes an active ethinyl group in its structure.

However, the electrochemical activity of EE2 is too poor, which allowed its low-level concentrations to be analyzed on bare electrodes. Hence, various kinds of modified electrodes have been introduced for EE2 detection over the years. Figure 1 shows typical cyclic voltammetry (CV) plot obtained for EE2 on a boron-doped diamond electrode (BDD) electrode in Britton-Robinson (BR) buffer [22]. The oxidation of EE2 is reflected by the irreversible peak at 0.65 V vs. Ag/AgCl in pH=8.0.

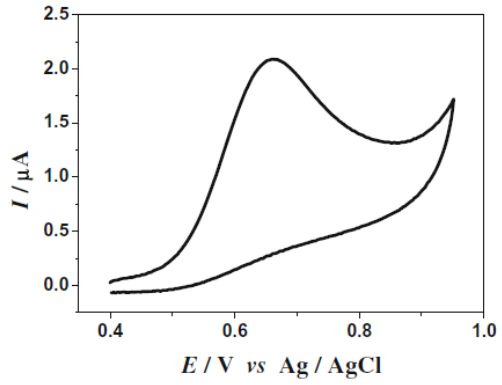


Figure 1. EE2 electrochemical behavior. Cyclic voltammograms of 1.96×10^{-5} M EE2 in BR (pH 8) at 100 mV s^{-1} on BDD electrode [22]

Table 1. Details of EE2 determination by various electroanalytical methods

Working and Ref Electrodes	**Electrolyte and pH**	**Tech.**	**Real sample**	**LR**	**LOD**	**Year [Ref]**
Dual thin GCE vs. Ag/AgCl	40% methanol-25% acetonitrile-35% PBS pH 6.0	LC-Amperometry	Human and Rabbit urine	250–1000 ng/ml	23.55 nM	2005 [57]
HMDE vs. Ag/AgCl/KCl	Universal BR pH 7.0	SWAdCS	Ethinyl-oestradiol® tablets, human serum and plasma	1.9×10^{-9}-6×10^{-7} M	0.59 nM	2006 [58]
CPB modified CPE vs. SCE	1.15 M PBS pH 8.04	LSV	Levonorgestrel and Etinylestradiol tablets	5.0×10^{-8}-2.0×10^{-5} M	0.03 µM	2007 [59]
anti-EE2/ MWCNTs/GCE vs. Ag/AgCl	0.01 M PBS pH 7.2	SWV	Water samples	0.035-70 ng/L	0.03 pM	2012 [21]
FTO-Chi/CNT electrode vs. Ag/AgCl	0.01 M PBS pH 7.0	SWV	Synthetic human urine	0.05-20 µM	0.09 µM	2015 [60]
Anti-EE2/ AgNPs/SiO_2/GO/ GC vs. Ag/AgCl	0.1 M PBS pH 6.0	CV	Urine	0.1-50 ng/mL	0.21 nM	2016 [61]
BDD vs. Ag/AgCl	0.1 M BR pH 8.0	SWV	Water sample	9.9×10^{-7}- 5.2×10^{-6} M	0.24 µM	2016 [22]
rGO/RuO_2NPs/ GCE vs. Ag/AgCl	PBS pH 7.0	DPV	Urine sample	5.50×10^{-8}-1.20×10^{-6} M	2.04 nM	2017 [62]
FTO/PVP/Chi /rGO vs. Ag/AgCl	PBS pH 7.0	Amperometry	Synthetic and human urine	0.25 -20 pM	0.15 pM	2018 [63]
HDME SPCE SPCNTE	0.03 M BR pH 10.0	SWAdSV SWV SWV	Urine sample	-	0.05 µM 4.6 µM 3.75 µM	2019 [39]
Solid amalgam electrode fabricated with Ag NPs vs. Ag/AgCl	0.04 M BR pH 7.0	SCV	Pharmaceutical and urine sample	6.4×10^{-7}M-7.8×10^{-6}M	0.103 µM	2020 [64]
Au/Fe_3O_4@TA/ MWNT/GCE vs. Ag/AgCl	0.1 M PBS pH 9.0	DPV	Human serum, urine specimens, natural water, and wastewater	0.01–120 µM	3.3 nM	2020 [65]
Ni deposited-SPCE	0.1 M BR pH 8.0	SWV	Organic fertilizers	0.23-30 µM	0.052 µM	2021 [66]
mag@MIP)-GQDs-FG-NF/SPE	0.2 M PBS pH 7.0	SWV	River water, serum, and urine	0.001-2.5 µM	2.6 nM	2021 [67]
Immunosensor AntiEE2-Au NPs/rGO/SPE	0.1 M PBS pH 7.0	EIS	Water sample	5-100 ng L^{-1}	1.5 ng L^{-1}	2021 [68]

Here, we are going to consider these modifiers and electrochemical techniques reported for the electrochemical analysis of EE2 in various real samples. The electroanalytical techniques reported for the determination of EE2 in different samples have been considered and listed in Table 1. Our search till 2021 from Scopus database resulted 15 reports [21,22,39,57-68].

In all reports, E2 was electrochemically determined on a modified electrode. The type of working and reference electrodes, modifiers, used electrochemical technique, analyzed matrix, linear ranges, and limit of detections of each report was shown in Table 1.

The first report on the electrochemical determination of EE2 dates back to 2015. Wang and Jseng [57] developed a liquid chromatography procedure using an amperometric detection system for the analysis of estrone, estradiol, estriol, 2-methoxyestrone, and ethinylestradiol in urine samples obtained from humans and rabbits. The detection system included a dual thin-layer glassy carbon electrode. The electrolyte/was the mobile phase contained 40% methanol-25% acetonitrile-35% in phosphate buffer (pH=6.0).

Ghoneim et al [58] determined EE2 on a hanging mercury drop electrode (HMDE) in BR. Li [59] used a carbon paste electrode (CPE) modified with cetyl pyridine bromide (CPB) to study the electrochemical behavior of EE2. The oxidation peak potential of EE2 showed a negative shift on the modified CPE while the peak current had a significant increase, which was attributed to the improved accumulation of EE2 on the modified electrode due to the electrostatic interaction with cetyl pyridine bromide.

Martínez et al [21] described the developing a precise, sensitive and selective sensor for preconcentrating and analysis of EE2 presented in aqueous media. The biosensor was based on magnetic particles (MPs) having anti-EE2 antibodies (anti-EE2 Ab) were fixed on a glassy carbon electrode (GCE) modified with multi-walled carbon nanotubes (MWCNTs/GCE) to create the required bioaffinity for preconcentration of EE2. The desorption of the analyte was performed using a H_2SO_4 solution. The determination of the pre-concentrated analyte was conducted via square wave voltammetry (SWV). The biosensor showed promising results in the direct trace analysis of EE2.

Pavinatto et al [60] reported a fluorine-doped tin oxide (FTO) electrode, which was coated Layer-by-Layer (LbL) films of chitosan/multi-walled carbon nanotubes (Chi/MWCNTs). CV plots were recorded using three bilayer electrode (FTO-$(Chi/CNTs)_3$) as a selective probe for the analysis of EE2, via an irreversible adsorption-controlled electrochemical oxidation process. The square wave voltammograms (SWV) led to a linear response in EE2 screening. The responses obtained using FTO-$(Chi/CNTs)_3$ had relative standard deviations (RSD) of 3.2% and 6.6% to intra- and inter-electrode modes, and the response of the electrode was not subject to significant interference effects from common interfering compounds.

Cincotto et al [61] developed a competitive amperometric immunoassay for analysis of EE2 based on peroxidase-labeled ethinylestradiol (HRP-EE2). The amperometric measurement were performed at -200 mV in the presence of hydroquinone as redox mediator,

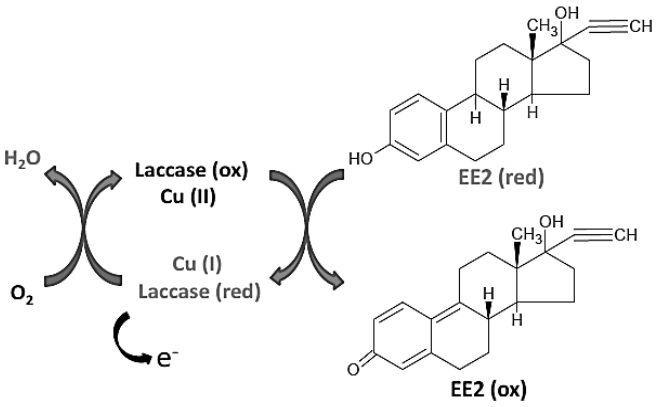
and the response had an RSD of 4.5% (n=10). The response obtained using a single bio-electrode, which was modified with anti-EE2, did not considerably change for at least for 15 days. During this period, the electrode was stored at 4°C in a humid atmosphere between the measurements.

Prado et al [62] electrochemically immobilized ruthenium nanoparticles on the surface of a GCE and the resulting electrode was used for simultaneous electroanalysis of EE2 and Amoxicillin (AMX).

Comparison of rGO/RuNP/GCE with rGO/GCE and GCE revealed better stability and separation of anodic peak currents, which is desirable for the determination of organic molecules in environmental, clinical and food specimens.

In 2018, Pavinatto et al [63] reported a very sensitive electrochemical EE2 biosensor based on electrospun nanofibers of polyvinylpyrrolidone (PVP)/chitosan (Chi)/ reduced graphene oxide (rGO), which was further grafted with Laccase. The hybrid nanofibers were deposited on a fluorine-doped tin oxide (FTO) electrode. The mechanism proposed for detecting EE2 is presented in Scheme 2.

The PVP/Chi/rGO-Laccase electrode was used in the amperometric detection of EE2 and showed a detection limit of 0.15 pM (3.3 σ/S), further to favorable intra- and inter-electrode RSD value of 4.29% and 8.44% for. The PVP/Chi/rGO-Laccase electrode also had a selective behavior towards EE2 in samples containing several common interfering species.



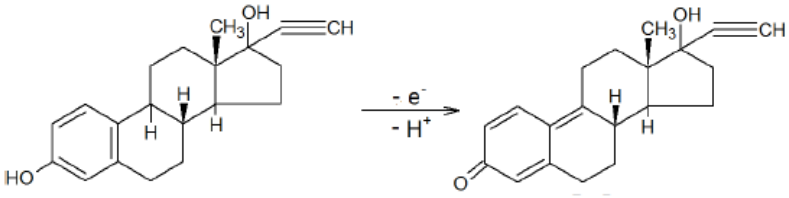
Scheme 2. Reaction mechanism of the Laccase biosensor for indirect determination of EE2 [63]

In a report in 2019 by Triviño et al [39] a robust method was proposed for the analysis of E2 and EE2 in medicinal formulations and urine specimens using an HMDE, screen printed carbon electrodes (SPCE), and screen-printed carbon nanotube electrodes (SPCNTE). The method involved adsorbing the analytes on the working electrodes, and the optimal values for different parameters included pH= 10.0; E_{ads}= -0.60 and t_{ads}= 30 s, for HMDE. Under optimal conditions, a reduction signal was observed at -1.31 V for E2 and two reduction signals

appeared at -0.23 V and -1.20 V for EE2. The limits of detection were 0.3 μg L^{-1} for E2, 14.8 μg L^{-1} for EE2 at -0.23 V, and 9.7 μg L^{-1} for EE2 at -1.20 V.

In the case of the screen-printed electrodes, on the other hand, E2 and EE2 samples led to oxidation signals for phenolic -OH groups at 0.30, 0.31, 0.32, and 0.33 V (pH=10) with respective detection limits of 242, 277; 182, and 191 μg L^{-1} for SPCE and SPCNTE. The method was used to determine the target species in Primaquin® (E2), Gynera® (EE2), spiked urine (with EE2), and urine samples of women using Tinelle®.

As results of these reports, E2 electro-oxidation can be perfrmed through a one-step process as shown in Scheme 3.



Scheme 3. Single electron transfer electrooxidation of EE2 [62]

As it can be seen from the chemical structure of EE2, it can be concluded that it has a suitable moiety to be electrooxidized generally at potential from +0.6 to +0.7 V depends on pH of the electrolyte or the type of electrode setup and the modifiers on working electrodes surfaces. However, its oxidation on the electrode surface is not sensitive enough to allow low-level concentration measurements through electrochemical methods. As discussed earlier, detecting low-level concentrations of EE2 in pharmaceutical, environmental and biological samples up to pM or less is required. As reports shown in Table 1, several researchers have tried to determine EE2 electrochemically. Martínez et al [21] and Pavinatto et al [63] reached the best detection limit up to pM. In both works, two biosensors were designed. Electrode modifications and suitable bioreceptors improve performance of the method.

In a report in 2020 [64], staircase voltammetry (SCV) was employed for the simultaneous determination of EE2 and cyproterone acetate (CPA). SCV was used instead of sensitive DPV or SWV because the researchers found that these techniques are not successful in separation of EE2 and CPA voltammetric signals.

A real sample, organic fertilizers containing EE2 was analyzed by a portable sensor [66]. In this work Silva et al, modified a screen-printed carbon electrode with nickel film which was electrochemically deposited on the surface of the electrode.

In 2021, Santos et al [67] also reported another portable sensor based on a screen-printed electrode modified with functionalized graphene, graphene quantum dots and molecularly imprinted polymers (MIP) for the sensitive detection of EE2.

An immunosensor was reported for the determination of EE2 through electrochemical impedance spectroscopy in 2021 [69]. This biosensor was made by direct modification of a screen-printed carbon electrode. Initially, electrochemically reduced graphene was coated on the surface of the electrode. Next, a porous gold nanoparticle was placed on the reduced graphene SPCE through electrodeposition. Finally, specific anti-EE2 antibodies immobilized on the Au surface by a covalent bonding using α-lipoic acid. The designed biosensor was then incubated for 30 min in an EE2 sample solution. After interacting the antigen–antibody binding, the charge transfer resistance of a redox probe $[Fe(CN)_6]^{4-/3-}$ on the electrode surface was recorded which was proportioned to the amount of EE2 molecules be captured.

4. CONCLUSION AND SUMMARY

17α−Ethinylestradiol (EE2) is a widely used synthetic estrogen. It is also an endocrine disruptor compound (EDC), which can enter as pollutant in environment. Even low level of this compound can affect the human life and causes serious metabolic disorders or other illness. Thus, monitoring its concentration is vital. Sine EE2 can be an electroactive compound; it is possible to be analyzed through some electrochemical methods. There are several electrochemical methods for determination of EE2, while there are many reports on detection or monitoring of EE2 with other analytical techniques. Designing sensors or biosensors based on various materials, (nanomaterials) give a chance to find a better way to analysis EE2 through fast and simple methods.

List of Abbreviations

Abbreviation	**Full name**
BDD	Boron Doped Diamond
BR	Briton-Robinson (buffer)
CPE	Carbon Paste Electrode
CPB	Cetyl Pyridine Bromide
CV	Cyclic Voltammetry
DPV	Differential Pulse Voltammetry
E1	Estrone
E2	Estradiol
E3	Estriol
EDC(s)	Endocrine Disrupter Compound(s)
EIS	Electrochemical Impedance Spectroscopy
FTO	Fluorine doped Tin Oxide
GC	Gas Chromatography
GCE	Glassy Carbon Electrode
HME	Hanging Mercury Electrode
HMDE	Hanging Mercury Drop Electrode
HPLC	High-performance liquid chromatography
LbL	Layer-by-Layer
LC	Liquid Chromatography

LOD	Limit of Detection
LR	Linear Range
LSV	Linear Sweep Voltammetry
MIP	Molecularly Imprinted Polymer
MP(s)	Magnetic Particles
MS	Mass Spectroscopy
OC	Oral Contraceptives
PBS	Phosphate Buffer Solution/Saline
rGO	Reduced Graphene Oxide
RSD	Relative Standard Deviation
SWV	Square Wave Voltammetry
SWAdSV	Square Wave Adsorptive Striping Voltammetry
SCE	Standard Calomel Electrode
SCV	Staircase voltammetry
SPE	Screen Printed Carbon Electrode
SPCNTE	Screen Printed Carbon Nanotube Electrode

REFERENCE

[1] Y.C. Wang, P. Su, X.-X. Zhang, and W.B. Chang, Anal. Chem. 73 (2001) 5616.

[2] J. Geisler, D. Ekse, H. Helle, N.K. Duong, and P.E. Lønning, The Journal of Steroid Biochemistry and Molecular Biology 109 (2008) 90.

[3] T. Kokko, L. Kokko, T. Lövgren, and T. Soukka, Anal. Chem. 79 (2007) 5935.

[4] S.S.C. Tai, and M.J. Welch, Anal. Chem. 77 (2005) 6359.

[5] A. Stopforth, B.V. Burger, A.M. Crouch, and P. Sandra, J. Chromat. B 856 (2007) 156.

[6] L. Havlíková, L. Nováková, L. Matysová, J. Šícha, and P. Solich, J. Chromatogr. A 1119 (2006) 216.

[7] B.C. Janegitz, F.A. dos Santos, R.C. Faria, and V. Zucolotto, Mater. Sci. Eng. C 37 (2014) 14.

[8] J.H. Kim, S.Y. Lee, H.J. Lee, N.Y. Yoon, and W.S. Lee, Annals of dermatology 24 (2012) 295.

[9] Y. Li, J. Xu, M. Jia, Z. Yang, Z. Liang, J. Guo, Y. Luo, F. Shen, and C. Sun, Mater. Lett. 159 (2015) 221.

[10] M. Kawaguchi, R. Ito, N. Sakui, N. Okanouchi, K. Saito, and H. Nakazawa, J. Chromatogr. A 1105 (2006) 140.

[11] Z. Lin, L. Chen, G. Zhang, Q. Liu, B. Qiu, Z. Cai, and G. Chen, Analyst 137 (2012) 819.

[12] M. Murugananthan, S. Yoshihara, T. Rakuma, N. Uehara, and T. Shirakashi, Electrochim Acta 52 (2007) 3242.

[13] S.R. Greytak, A.M. Tarrant, D. Nacci, M.E. Hahn, and G.V. Callard, Aquatic Toxicology 99 (2010) 291.

[14] J. Corcoran, M.J. Winter, and C.R. Tyler, Critical Reviews in Toxicology 40 (2010) 287.

[15] K.L. Howdeshell, J. Furr, C.R. Lambright, V.S. Wilson, B.C. Ryan, and L.E. Gray Jr, Toxicological Sci. 102 (2007) 371.

[16] M. Vosges, J.C. Braguer, and Y. Combarnous, Reproductive Toxicology 25 (2008) 161.
[17] J.P. Sumpter, and A.C. Johnson, Journal of Environmental Monitoring 10 (2008) 1476.
[18] L.L. Johnson, D.P. Lomax, M.S. Myers, O.P. Olson, S.Y. Sol, S.M. O'Neill, J. West, and T.K. Collier, Aquatic Toxicology 88 (2008) 29.
[19] C. Purdom, P. Hardiman, V. Bye, N. Eno, C. Tyler, and J. Sumpter, Chemistry and Ecology 8 (1994) 275.
[20] Å. Bergman, J.J. Heindel, S. Jobling, K. Kidd, and T.R. Zoeller, W.H. Organization, State of the science of endocrine disrupting chemicals (2012) World Health Organization2013.
[21] N.A. Martínez, S.V. Pereira, F.A. Bertolino, R.J. Schneider, G.A. Messina, and J. Raba, Anal. Chim. Acta 723 (2012) 27.
[22] C. Perez, F.R. Simões, and L. Codognoto, J. Solid State Electrochem. 20 (2016) 2471.
[23] [Online] avialable at: https://www.drugs.com/sfx/ethinyl-estradiol-side-effects.html; accessed 20.10.1019
[24] Z. Yan, G. Lu, J. Liu, and S. Jin, Ecotoxicology and Environmental Safety 84 (2012) 334.
[25] J.P. Sumpter, and A.C. Johnson, Environmental Sci. Technol. 39 (2005) 4321.
[26] M. La Farre, S. Pérez, L. Kantiani, and D. Barceló, TrAC Trends in Anal. Chem. 27 (2008) 991.
[27] R.W. Reis Filho, J.d. Araújo, and E.M. Vieira, Quim. Nova 29 (2006) 817.
[28] F.F. Sodré, I.C. Pescara, C.C. Montagner, and W.F. Jardim, Microchem. J. 96 (2010) 92.
[29] L.S. Shore, and M. Shemesh, Pure Appl. Chem. 75 (2003) 1859.
[30] R. Guedes-Alonso, Z. Sosa-Ferrera, and J.J. Santana-Rodríguez, J. Anal. Methods Chem. 2013 (2013) 210653.
[31] M. Jürgens, K. Holthaus, A. Johnson, J. Smith, M. Hetheridge, and R. Williams, Environ. Toxicol. Chem. 21 (2002) 480.
[32] S. Saini, Anal. Chem. Lett. 8 (2018) 6061.
[33] T. Martimiano do Prado, F. Cincotto, F. Moraes, and S.A.S. Machado, Electroanalysis (2017).
[34] L. Rosenberg, J.R. Palmer, M.I. Sands, D. Grimes, U. Bergman, J. Daling, and A. Mills, American J. Obstetrics and Gynecology 177 (1997) 707.
[35] L. Chasan-Taber, and M.J. Stampfer, Annals of Internal Medicine 129 (1998) 747.
[36] M.J. Rosenberg, A. Meyers, and V. Roy, Contraception 60 (1999) 321.
[37] M.J. Rosenberg, M.S. Waugh, and T.E. Meehan, Contraception 51 (1995) 283.
[38] M.J. Rosenberg, and M.S. Waugh, American J. Obstetrics and Gynecology 179 (1998) 577.
[39] J.J. Triviño, M. Gómez, J. Valenzuela, A. Vera, and V. Arancibia, Sens Actuators B (2019).
[40] J. Tang, J. Wang, L. Yuan, Y. Xiao, S. Wang, and X. Wang, Food Anal. Methods (2019).
[41] S. Seidi, L. Alavi, A. Jabbari, and M. Shanehsaz, J. Iran. Chem. Soc. 16 (2019) 1007.

[42] N.M.F.M. Sampaio, N.D.B. Castilhos, B.C. Da Silva, I.C. Riegel-Vidotti, and B.J.G. Silva, Molecules 24 (2019).
[43] V.L. Louros, D.L.D. Lima, J.H. Leitão, V.I. Esteves, and H.G. Nadais, J. Sep. Sci. 42 (2019) 1585.
[44] S.N. do Carmo, J. Merib, and E. Carasek, J. Chromatogr. B Anal. Technol. Biomed. Life Sci. 1118-1119 (2019) 17.
[45] M.S. Daniel, and E.C. de Lima, Rev. Ambiente Agua 9 (2014) 688.
[46] Y. Zuo, K. Zhang, and S. Zhou, Environ. Sci. Process. Impacts 15 (2013) 1529.
[47] Y.q. Zhou, Z.j. Wang, and N. Jia, J. Environ. Sci. 19 (2007) 879.
[48] M. Alimzhanova, Y. Nurzhanova, D. Onglasynkyzy, K. Ashimuly, and S. Batyrbekova, International Multidisciplinary Scientific Geoconference (2016) pp. 209.
[49] Q. Xu, S.Y. Wu, M. Wang, X.Y. Yin, Z.Y. Wen, W.N. Ge, and Z.Z. Gu, Chromatographia 71 (2010) 487.
[50] N.A. Martínez, R.J. Schneider, G.A. Messina, and J. Raba, Biosens. Bioelectron. 25 (2010) 1376.
[51] P. Braun, M. Moeder, S. Schrader, P. Popp, P. Kuschk, and W. Engewald, J. Chromatogr. A, 988 (2003) 41.
[52] L.N. Hordge, K.L. McDaniel, D.D. Jones, Jr., and S.O. Fakayode, Talanta 152 (2016) 401.
[53] M.L. Scala-Benuzzi, E.A. Takara, M. Alderete, G.J.A.A. Soler-Illia, R.J. Schneider, J. Raba, and G.A. Messina, Microchem. J. 141 (2018) 287.
[54] H. Kanso, N. Inguimbert, G. Istamboulie, L. Barthelmebs, C. Calas-Blanchard, and T. Noguer, Anal. Biochem. 537 (2017) 63.
[55] H. Kanso, L. Barthelmebs, N. Inguimbert, and T. Noguer, Anal. Chem. 85 (2013) 2397.
[56] T. Hintemann, C. Schneider, H.F. Schöler, and R.J. Schneider, Water Res. 40 (2006) 2287.
[57] L.H. Wang, and S.F. Jseng, J. Liq. Chromatogr. Relat. Technol. 28 (2005) 1367.
[58] E.M. Ghoneim, H.S. El-Desoky, and M.M. Ghoneim, J. Pharm. Biomed. Anal. 40 (2006) 255.
[59] C. Li, Bioelectrochemistry 70 (2007) 263.
[60] A. Pavinatto, L.A. Mercante, C.S. Leandro, L.H.C. Mattoso, and D.S. Correa, J. Electroanal. Chem. 755 (2015) 215.
[61] F.H. Cincotto, G. Martínez-García, P. Yáñez-Sedeño, T.C. Canevari, S.A.S. Machado, and J.M. Pingarrón, Talanta 147 (2016) 328.
[62] T.M. Prado, F.H. Cincotto, F.C. Moraes, and S.A.S. Machado, Electroanalysis 29 (2017) 1278.
[63] A. Pavinatto, L.A. Mercante, M.H.M. Facure, R.B. Pena, R.C. Sanfelice, L.H.C. Mattoso, and D.S. Correa, Appl. Surf. Sci. 458 (2018) 431.

[64] V. O. Jesus, V. S. Ferreira, and B. G. Lucca, Talanta 210 (2020) 120610.
[65] M. Nodehi, M. Baghayeri, R. Ansari, and H. VeisiMater. Chem. Phys. 244 (2020) 122687.
[66] L. R.G. Silva, J.G.A. Rodrigues, J.P. Franco, L.P. Santos, E. D'Elia, W. Romao, and R.Q. Ferreira, Ecotoxicology and Environmental Safety 208 (2021) 111430.
[67] A.M. Santos, A. Wong, T.M. Prado, E.L. Fava, O. Fatibello-Filho, M.D.P. T. Sotomayor, and F. C. Moraes, Talanta 224 (2021) 121804.
[68] M. L.,Scala-Benuzzi, G. J. A. A. Soler-Illia, J. Raba, F. Battaglini, R.J. Schneider, S. V. Pereira, and G.A. Messina, J. Electroanal. Chem. 897 (2021) 115604.

Copyright © 2022 by CEE (Center of Excellence in Electrochemistry)

ANALYTICAL & BIOANALYTICAL ELECTROCHEMISTRY (http://www.abechem.com**)**

Reproduction is permitted for noncommercial purposes.