Journal of Pharmaceutical Analysis 9 (2019) 9–19

Contents lists available at ScienceDirect

Journal of Pharmaceutical Analysis

journal homepage: www.elsevier.com/locate/jpa
www.sciencedirect.com

Review Paper

Recent advances in electrogenerated chemiluminescence biosensing methods for pharmaceuticals

Yu Zhang [a], Rui Zhang [b,*], Xiaolin Yang [c], Honglan Qi [c], Chengxiao Zhang [c]

[a] Medpace Bioanalytical Laboratories, 5365 Medpace Way, Cincinnati, OH 45227, USA
[b] School of Informatics, Computing, and Engineering, Indiana University, Bloomington, IN 47405, USA
[c] Key Laboratory of Analytical Chemistry for Life Science of Shaanxi Province, School of Chemistry and Chemical Engineering, Shaanxi Normal University, Xi'an 710062, China

ARTICLE INFO

Article history:
Received 20 June 2018
Received in revised form
20 November 2018
Accepted 21 November 2018
Available online 22 November 2018

Keywords:
Electrogenerated chemiluminescence
Pharmaceutical analysis
Immunoassay
Biosensors

ABSTRACT

Electrogenerated chemiluminescence (electrochemiluminescence, ECL) generates species at electrode surfaces, which undergoes electron-transfer reactions and forms excited states to emit light. It has become a very powerful analytical technique and has been widely used in such as clinical testing, biowarfare agent detection, and pharmaceutical analysis. This review focuses on the current trends of molecular recognition-based biosensing methods for pharmaceutical analysis since 2010. It introduces a background of ECL and presents the recent ECL developments in ECL immunoassay (ECLIA), immunosensors, enzyme-based biosensors, aptamer-based biosensors, and molecularly imprinted polymers (MIP)-based sensors. At last, the future perspective for these analytical methods is briefly discussed.

1. Introduction

Electrogenerated chemiluminescence (also called Electrochemiluminescence and abbreviated ECL) generates species at electrode surfaces, which then undergoes electron-transfer reactions to form excited states that emit light [1–3]. The first detailed ECL study by Hercules [4], Visco and Chandross [5] and Santhanam and Bard [6] was reported in the mid-1960s, although early reports about light emission during electrolysis can be dated back to the 1920s [7,8]. ECL possesses several advantages over chemiluminescence (CL), photoluminescence (PL), and electrochemistry. First, the electrochemical reaction in ECL allows the controls of the time and position of the light-emitting reaction through applied potential. Second, ECL is more selective than CL, since the generation of excited states in ECL can be selectively controlled by changing the electrode potentials. Third, ECL is a nondestructive technique, such that, in many cases, ECL emitters can be regenerated after emission [2]. Compared to PL method, ECL method does not need a light source. Therefore, it offers many advantages to avoid issues such as scattered light and luminescent impurities. Compared to electrochemical methods, ECL method is more selective and has less electrode fouling. ECL method has become popular in analytical chemistry and sensor technology, owing to its wide versatility, simple instrumentation, low background signal, wide linear working range, and high sensitivity. ECL method has been widely used in immunoassay, food and water testing, biowarfare agent detection, and pharmaceutic analysis.

There are numerous reviews on ECL analytical applications [1,9–11], specifically, in biosensing [2], immunoassays and DNA probe assays for clinical diagnostics [3], capillary electrophoresis coupling with ECL detection [12,13], immunosensors [14], immunosensing and biological and pharmaceutic analysis [15], and nanomaterials applications in ECL biosensors and biosensing [16–19]. However, particular issues about recent advances in ECL biosensing methods for pharmaceuticals have not yet been fully reviewed. Therefore, this review focuses on trends of ECL biosensing methods for pharmaceuticals since 2010. It introduces a general background of ECL and presents the recent ECL developments in molecular recognition-based biosensing methods for pharmaceuticals, including immunosensors, enzyme-based biosensors, aptamer-based biosensors, and molecularly imprinted polymers (MIP) sensors. A perspective on potential developments in ECL biosensing is also briefly provided.

2. Typical ECL systems

ECL systems can be classified into two kinds based on their ECL reaction mechanisms (i.e., ion annihilation or a coreactant ECL process) [1,2]. Although modern ECL applications are almost

Peer review under responsibility of Xi'an Jiaotong University.
* Corresponding author.
E-mail address: rz20@iu.edu (R. Zhang).

https://doi.org/10.1016/j.jpha.2018.11.004

exclusively based on coreactant ECL, the early ECL studies were originated from ion annihilation ECL. Ion annihilation ECL involves the formation of an excited state as a result of an exergonic electron transfer between electrochemically generated species, often radical ions, at the surface of an electrode. Electrolytic generation of both the oxidized and reduced ECL precursors is required [1]. Ion annihilation ECL systems mainly involve organic compounds in organic solvents [1,2]. Donor-acceptor conjugated molecules have been recognized as promising organic molecules for applications in organic light-emitting diodes and photovoltaic devices, nonlinear optics, field-effect transistors and fluorescence imaging [20–25]. However, only a few of these organic regents could be used as labelers in ECL biosensors [25].

Coreactant ECL systems are widely employed in analytical chemistry, since the ECL reagents have a high ECL efficiency (so that the developed ECL methods have a high sensitivity). Unlike ion annihilation ECL, coreactant ECL is frequently generated with one directional potential scanning at an electrode, and in a solution containing luminophore species ("emitter") in the presence of a deliberately added reagent (coreactant). Depending on the polarity of the applied potential, both the luminophore and the coreactant species can be first oxidized or reduced at the electrode to form radicals. Intermediates formed from the coreactant then decompose to produce a powerful reducing or oxidizing species. The species reacts with the oxidized or reduced luminophore to produce the excited states that emit light. Based on luminophores used as ECL labels in coreactant ECL systems, ECL reagents (materials) can be mainly classified into three types including metal ion (Ru, Ir) complexes [3,11–13], luminol and its derivatives [9,10], and nanomaterials [17–19].

2.1. $Ru(bpy)_3^{2+}$/TPA system

The majority of reported ECL applications involve $Ru(bpy)_3^{2+}$ or its derivatives as an emitter (or label) and tripropylamine (TPA) as a coreactant. The $Ru(bpy)_3^{2+}$/TPA system exhibits the highest ECL efficiency and thus this system constitutes the basis of commercial systems for immunoassay and DNA analysis [3,11]. The ECL mechanism of this system is very complicated in aqueous solution and has been extensively investigated [2,26]. In general, this system gives ECL emission that is a function of applied potential and consists of two waves. The first wave ($\sim +0.8$ V, vs SCE) occurs with the direct oxidation of TPA at the electrode, and is often merged into the foot of the second wave ($+1.1$ V vs. SCE) when relatively high concentration of $Ru(bpy)_3^{2+}$ (~millimolar) is used. The second wave appears where $Ru(bpy)_3^{2+}$ is oxidized at $+1.1$ V vs. SCE. Both waves are associated with the emission from $Ru(bpy)_3^{2+*}$ ($\lambda_{ECL} =$ 642 nm) [2]. The relative ECL intensity from the first wave is significant, especially in diluted $Ru(bpy)_3^{2+}$ solutions (less than approximately micromolar) containing ~0.1 M TPA. Thus, for the low concentrations of analytes such as in immunoassays and detection of drugs and biomakers with $Ru(bpy)_3^{2+}$ as an ECL label, the bulk of the ECL signal obtained in this system probably originates from the first ECL wave at Au electrode or from the second ECL wave at carbon electrode [27–32]. In recent years, cyclometalated iridium (III)-complexes have received much attention, since these complexes have high ECL efficiencies and low emission potentials compared with Ru(II) complexes [33,34].

2.2. Luminol-H_2O_2 system

Luminol (2,3-aminophthalhydrazide) is another classic ECL reagent. Light emission from luminol at electrodes was first reported in 1929 upon application of $+2.8$ V in aqueous alkaline solution [8]. Luminol ECL is often produced in alkaline solution with hydrogen peroxide, when anodic electrochemical oxidation from the electrogenerated excited monoanionic form of 3-aminophthalic acid ($\lambda_{ECL} = 420$ nm) occurs at a Pt or carbon electrode ($+0.6$ V vs SCE) [1]. The coreactant H_2O_2 can be generated from many oxidase catalytic systems. In ECL biosensing, the ECL reagents luminol and its derivatives [35,36] can be coupled with various oxidases, such as horseradish peroxidase (HRP), acetylcholinesterase (AChE), cholesterol oxidase (ChOx), and choline oxidase (ChOX) for the determination of specific proteins and small drugs.

2.3. Semiconductor nanoparticle system

Since the first report of ECL of silicon quantum dots (2–4 nm in diameter) in 2002 [37], the use of nanomaterials in ECL has become one of the most active research areas. The ECL of semiconductor nanoparticles (NPs, also known as nanocrystals, quantum dots) was generated from both annihilation and coreactant oxalate ($C_2O_4^{2-}$) and persulfate ($S_2O_8^{2-}$) systems in MeCN. The visible light can be produced by electron transfer reactions between positively and negatively charged nanocrystals (or between charged nanocrystals and molecular redox-active coreactants) that lead to electron and hole annihilation. The ECL spectra of SiNPs exhibited a peak maximum at 640 nm, a significant red shift from the PL maximum (420 nm) of the same Si NP solution. In addition to elemental semiconductors (e.g., Si and Ge), many compound semiconductors (e.g., CdS, CdSe, and CdTe) can produce ECL [17–19]. Recently, nanosheets such as graphite-like carbon nitride (g-C_3N_4) have been reported [38–42]. Nanomaterials-based ECL biosensors have been shown as promising potentials compared with traditional ones in analytical applications [17,42].

A variety of pharmaceuticals have been determined by ECL methods self-molecular ECL [43], enhancing typical ECL systems [44–46], or coupling with separation methods such as capillary/microchip electrophoresis [12,13]. ECL methods based on molecular-recognition have received much attention, since they offer high selectivity and accuracy. In this review, biosensing methods will be presented in three sections based on the difference of molecular-recognition materials, including biological materials (e.g. enzymes, antibodies, and nucleic acids), biologically derived materials (e.g. recombinant antibodies, and aptamers) and biomimic (e.g. biomimetic catalysts and imprinted polymers). The review for ECL biosensing pharmaceuticals reported recently is focused on the description of the molecular recognition materials including antibodies, aptamers, enzymes and imprinted polymers as well as the ECL signals.

3. ECL immunoassays/immunosensors

Immunoassays are powerful analytical methods based on the high bio-affinity of the antigen and antibody reaction, which forms the foundation of radio-immunoassays (RIAs). This revolutionary concept was first introduced for measuring endogenous plasma insulin by Yalow and Berson in 1960 [47]. The remarkable selectivity of antibodies is based on the stereo-specificity of the antigen binding site that provides large binding constants (ranging from 10^5 to 10^9 L/mol). Immunoassay mainly includes sandwich type format and competition assay format. The sandwich type format is perhaps the most commonly used means of ECL detection of an antigen [2]. In sandwich type format, primary antibody (Ab1) is generally immobilized on the surface of electrode as capture probe, and then bound with the target antigen and the ECL reagent-labeled second antibody as signal probe, which is detected using ECL technology. In a competition type format, unlabeled analyte (usually antigen) in the test sample is measured by its ability to compete with a labeled antigen in the immunoassay. The unlabeled antigen blocks the ability of the labeled antigen to bind

because that binding site on the antibody is already occupied. Thus, in a competitive immunoassay, less label measured in the assay means more of the unlabeled (test sample) antigen is present. The amount of antigen in the test sample is inversely related to the amount of label measured in the competitive format. In the past 8 years, many advances in the ECL immunoassays for quantitation of pharmaceuticals have been made, including evaluation of the ECL immunoassays by commercially available and newly-developed ECL immunoassays/immunosensors (Table 1 and Table 2).

3.1. Evaluation of the ECL immunoassays commercially available

Evaluation of the ECL immunoassay (ECLIA) established with HPLC/MS and other immunoassays is an important issue, since it can provide important information of the evaluated ECL immunoassays for laboratories. Brandhorst et al. [48] reported an evaluation of new ECL testosterone immunoassay (Elecsys® Testo II) compared to HPLC/MS. The new Elecsys® Testo II assay showed an improved agreement with a validated HPLC/MS assay for testosterone, particularly regarding testosterone concentrations in samples from female patients and may be a suitable alternative for laboratories with no access to mass spectrometry techniques.

Jafri et al. [49] reported a comparison of ECL immunoassay (Roche Modular E-170) with HPLC (Perkin Elmer series 200 with ultraviolet detection), and radio immunoassay (kit from DiaSorin) for quantification of serum 25 hydroxy vitamin D (25OHD). Acceptable correlation was observed among HPLC and RIA and also with RIA and ECLIA in quantification of 25OHD. The ease and overall consistency with which 25OHD can be measured with automated methods makes it suitable to large commercial laboratories where high throughput is required.

Johnson-Davis et al. [50] reported an evaluation of the Abbott ARCHITECT i2000 sirolimus assay and compared it with the Abbott IMx sirolimus microparticle enzyme immunoassay as well as LC-MS/MS method for sirolimus. The ARCHITECT i2000 assay demonstrated good linearity, low imprecision, and was free of common interferences. Results obtained from both immunoassay methods were biased slightly high, compared with those of LC-MS/MS method. Moreover, the agreement between the two immunoassays was better for frozen patient samples. Both immunoassays methods exhibited a $>100\%$ cross-activity with sirolimus.

Eshratkhah et al. [51] reported a comparative study on determination of plasma thyroid hormones including thyroxine (T4), tri-iodothyronine (T3), free thyroxine (fT4) and free tri-iodothyronine (fT3) in sheep by using DiaSorin chemiluminescence immunoassay (CLIA) kits and Cobas ECLIA kits (Roche Boeringer-Mannheim, USA). Results obtained from both immunoassay methods indicated that the ECLIA method is more suitable than the CLIA method for clinical endocrinology laboratory investigations especially thyroid hormones in sheep.

Shipkova et al. [52] reported a multicenter evaluation of the new ECLIA developed by Roche Diagnostics that uses a cobas immunoassay analyzer for tacrolimus, in five European laboratories with experience in the TDM of ISDs. The Elecsys Tacrolimus assay has good linearity, functional sensitivity and intermediate imprecision and is comparable to LC–MS/MS methods. The overall performance of ECLIA demonstrates a modern generation TAC assay that meets the demands of monitoring drug concentrations in current immunosuppressive regimens.

Vogeser et al. [53] reported a multicenter analytical evaluation of the automated ECLIA for cyclosporine (Elecsys cyclosporine; Roche Diagnostics, Switzerland) with chemiluminescent microparticle immunoassay (CMIA) on the Abbott Architect platform and LC–MS/MS. The data obtained from this multicenter evaluation indicated that the new ECLIA-based cyclosporine assay is fit for the therapeutic monitoring of CsA.

Miura et al. [54] assessed the inter-hospital laboratory variability of immunoassay methods for tacrolimus. The results were obtained by immunoassays including an affinity column-mediated immunoassay (ACMIA) on a Dimension analyzer, an enzyme-multiplied immunoassay technique (EMIT) on a Viva-E analyzer, a CLIA on the Architect system, and the ECLIA on a cobas analyzer. The 20% CVs values from the CLIA, ACMIA, EMIT, and ECLIA assays in the hospital laboratories were 1.82, 5.36, 4.59 and 0.89 ng/mL, respectively. CLIA and ECLIA provide adequate precision at the target tacrolimus concentration of 3.0 ng/mL, whereas ACMIA and EMIT appear unable to analyze target concentrations between 3.0 and 5.0 ng/mL. Appropriate assessment of tacrolimus concentration by an assay with higher sensitivity, precision, and accuracy is required to ensure long-term survival of transplant recipients administered tacrolimus.

Sasanoa et al. [55] reported an evaluation of the Elecsys® Cyclosporine and Elecsys® Tacrolimus assays on the cobas e411 analyzer compared to an affinity chrome-mediated immunoassay (ACMIA) for cyclosporine and a CLIA for tacrolimus. The analytical performances of the Elecsys® Cyclosporine and Elecsys®Tacrolimus assays were acceptable, and cyclosporine and tacrolimus concentrations may be simultaneously measured using a single pretreatment which is of benefit if patients have to undertake conversion between these two drugs. Additionally, it benefits the workflow in the clinical laboratory. Thus, the Elecsys® Cyclosporine and Elecsys® Tacrolimus assays may be suitable for routine therapeutic drug monitoring.

3.2. Newly-developed ECL immunoassays/immunosensors

Extensive efforts have been devoted to developing and improving the ECL immunoassays/immunosensors for pharmaceutical analysis, including exploring new ECL reagents, immobilization methods, molecular recognition elements, and searching signal-enhanced approaches.

Lowe et al. [56] developed a novel ECL immunoassay for quantitation of ranibizumab in human serum. In this assay, ruthenium (II) complex-labeled affinity-purified rabbit anti-

Table 1
ECL immunoassays commercially available.

Drugs/analytes	Systems	Linear ranges	LOD (S/N = 3)	Samples	Ref.
Testosterone	Elecsys® Testo II Roche diagnostics, Germany	0.12–15.0 μg/L	–	Serum	[48]
25-Hydroxy-vitamin D	Roche Modular E-170	4.9–214.6 nM	–	Serum	[49]
Sirolimus	Abbott ARCHITECT i2000 sirolimus assay	/	–	Whole blood	[50]
Thyroid hormone	Cobas ECLIA kits, Roche Boeringer-Mannheim, USA	/	–	Blood	[51]
Tacrolimus (TAC)	Elecsys tacrolimus assay, Roche diagnostics	0.5–40 ng/mL	1 ng/mL	Whole blood	[52]
Cyclosporine (CsA)	Elecsys cyclosporine assay, Roche diagnostics	30–2000 mcg/L	6.8 mcg/L	Whole blood	[53]
Tacrolimus (TAC)	Cobas®, Roche, Tokyo, Japan	0.0–26.0 ng/mL	0.89 ng/mL	Whole blood	[54]
Cyclosporine (CsA)	Elecsys® Cyclosporine assay kits	94–1238 ng/mL CsA	16 ng/mL CsA	Whole blood	[55]
Tacrolimus (TAC)	Elecsys® Tacrolimus assay kits	2.1–17.8 ng/mL TAC	0.95 ng/mL TAC		

Table 2
Newly-developed ECL immunoassays/immunosensors.

Drugs/Analytes	Systems	Linear ranges	LOD (S/N = 3)	Samples	Ref.
Ranibizumab	Sandwich-type, biotinylated rhVEGF/antigen/ruthenium-labeled antibodies, captured by streptavidin-coated paramagnetic beads	300–24,000 pg/mL	300 pg/mL	Human serum	[56]
Clenbuterol (CLB)	CLB-BSA-CHIT-AuNPs/GCE, Competitive immunoassay, $Ru(bpy)_3^{2+}$-labeled antibody	0.010–1.0 ng/mL	0.0050 ng/mL	Swine urine	[57]
Thyroid stimulating hormone (TSH)	Sandwich-type, enhancing ECL of $S_2O_8^{2-}$-O_2 system	0.05–20 μIU/mL	0.02 μIU/mL	Human serum	[58]
Morphine	Signal-off, label-free, Ab/AuNPs/APTMS/ITO in PBS (pH 8.0)-0.1 mM H_2O_2	2–200 ng/mL	0.82 ng/mL	Spiked urine	[59]
Morphine	Signal-off, label-free, Ab/CdS-PAMAM/AuNPs/Au in 0.1 M PBS-0.1 M $K_2S_2O_8$ (pH74)	0.2–180 ng/mL	67 pg/mL	Human blood plasma	[60]
Phenylethanolamine A (PA)	Competitive immunoassay, signal-off, PA-Ag/CHIT/CdSe QDs/GCE, PA-Ab/GaR IgG-HRP in 0.1 M Tris–HCl buffer (pH 9.0)-0.1 M KNO_3-0.5 mM HQ	0.05–1000 ng/mL	0.015 ng/mL	Pork and liver from swine	[61]
Brombuterol (Bro)	Competitive immunoassay, signal-off, Bro-Ag/SWCNTs-PAMAM-Ag@Au/GCE, signal probe (PAMAM-Au-CdSe QDs) in 0.1 M PBS (pH 9.0)-0.1 M $K_2S_2O_8$	0.001–1000 ng/mL	37 fg/mL	Pork and feed	[62]
Ketamine	Signal-off, Ab/PAMAM-carbon dots/AuNPs/GCE) in 0.1 M PBS (pH 7.4)- 0.1 M $K_2S_2O_8$	0.2–200 ng/mL	0.067 ng/mL	Spiked human plasma	[63]
Insulin	Sandwich-type, $ZnCd_{14}S$ ECL-RET, Ab1/Au-$ZnCd_{14}S$/NH_2-NMCs/GCE, Ab_2/Au-Cu alloy nanocrystals in 0.1 M PBS (pH 7.4)-60 mM $K_2S_2O_8$	0.1 pg/mL to 30 ng/mL	0.03 pg/mL	–	[64]

ranibizumab antibodies and biotinylated rhVEGF are added to serum samples. During overnight incubation, these two labeled molecules bind to ranibizumab, and the resulting immune complex is then captured by streptavidin-coated paramagnetic beads and analyzed for ECL. The ranibizumab PK ECLIA has a reporting range of 300–24,000 pg/mL, based on accuracy and precision parameters. It showed high precision for both intra- and inter-assay analyses. Recovery of ranibizumab from 10 individual donors averaged between 100% and 119% of nominal concentration. There was no cross-reactivity observed in the assay to other recombinant humanized antibodies (whole molecules or monoclonal antibody fragments) or human IgG. This assay's lower limit of quantitation is 300 pg/mL ranibizumab in neat serum, achieving a 67-fold improvement in sensitivity relative to a conventional ELISA-based PK method. Li et al. [57] reported an ultrasensitive ECL immunosensor for assay of trace amount of clenbuterol (CLB) residue in swine urine. The ECL immunosensor was fabricated by drop-coating the immunosensing composite including chitosan, AuNPs and the CLB–BSA on the surface of GCE. To perform the competitive immunoassay, the immunosensor was immersed into 60 μL of the mixture solution composed of the sample and the $Ru(bpy)_3^{2+}$-labeled CLB antibody as signal probe. After an incubation and washing, ECL measurements using the washed immunosensor were accomplished in 7.0 mL of TPA solution. The proposed method is suitable for screening of trace amount of CLB residue, due to its simple manipulation, short assay time, and high detection sensitivity, and exhibits a great promise in food safety and agonist monitoring.

An increasing attention has been made to develop label-free ECL biosensors for the determination of pharmaceuticals. This approach is based on the changes of the interfacial properties related to biorecognition events occurring at the modified surfaces, and hence the label process of ECL signal reagent to the biorecognition element is not required. Liu et al. [58] reported an ECL immunosensor for thyroid stimulating hormone (TSH) based on polyamidoamine-norfloxacin functionalized Pd-Au core-shell hexoctahedrons as signal enhancers. Norfloxacin(NFLX) was decorated abundantly on the surface of polyamidoamine(PAMAM) dendrimer via amide linkage of ormPAMAM-NFLX complex. The resultant PAMAM-NFLX was served as a novel co-reactant to efficiently amplify the ECL signal of $S_2O_8^{2-}$-O_2 system. Pd@Au HOHs were used as nano-carriers to assemble detection antibody (Ab2) and the PAMAM-NFLX complex and to intermediate free radical HO● during the ECL reaction of $S_2O_8^{2-}$-O_2 system. The proposed immunosensor successfully achieved the detection of TSH in practical human blood serum with desirable results. Yang et al. [59] designed a label-free immunosensor for morphine based on the luminol ECL on AuNPs/ITO. The ECL immunosensor was fabricated by electrostatic adsorbing morphine antibody on the surface of AuNPs/APTMS/ITO. After the immunosensor was incubated with morphine sample solution, the ECL measurement was carried out in PBS (pH 8.0) −0.1 mM H_2O_2, resulted in decreased ECL. The immunosensor was used to determine the concentration of morphine in spiked urine samples with a satisfactory recovery.

Several recent papers have addressed signal nanomaterials such as quantum dots (QDs) and carbon dots. Fei et al. [60] designed a signal-off ECL immunosensor for morphine using CdS QDs as signal and AuNPs as signal enhancers. The ECL immunosensor was fabricated by drop-coating polyamidoamine (PAMAM)-CdS QDs on the surface of the electrodeposited Au NP/Au electrode and finally covalent coupling anti-morphine antibody on the surface of the PAMAM-CdS using glutaraldehyde as well as blocking using BSA for non-specific absorption. The designed immunosensor was successfully applied to the determination of morphine in blood plasma. This kind of assay is expected to pave new avenues in label-free drug assays. Tang et al. [61] designed an ECL

immunosensor for the β-adrenergic agonist phenylethanolamine A (PA) using CdSe QDs as signal material and enzymatic amplification. The ECL immunosensor was fabricated by drop-coating CdSe QDs and chitosan solution on the surface of GCE, and then covalent coupling synthesized PA antigen on the surface of CdSe QDs using glutaraldehyde as well as blocking the remaining binding sites using BSA. After the immunosensor was incubated with the mixture solution of different concentrations of PA and fixed PA antibody at 37 °C for 1 h, and then bound with HRP-GaR IgG, the ECL measurement was carried out in 0.01 M Tris–HCl buffer (pH 9.0)- 0.5 mM HQ from −0.2 to −1.25 V, resulted in decreased ECL. The immunosensor was used to determine the concentration of morphine in spiked urine samples with a satisfactory recovery. Hu et al. [62] designed an ECL immunosensor for the illicit β-adrenergic agonist brombuterol (Brom) by applying a multiple signal amplification strategy based on a PAMAM-gold nanoparticle conjugate as the bioprobe and Ag@Au core shell nanoparticles as a substrate. The ECL immunosensor was fabricated by successively drop-coating PAMAM-SCNW, Ag@Au core/shell NPs, and Brom coating antigen on the surface of GCE. To perform the competitive immunoassay, a mixture solution, composed of different concentrations of Brom and a fixed of PAMAM-Au-CdSe QDs-Brom-antibody ECL signal probes on the surface of the ECL immunosensor, was prepared. The ECL measurement was carried out in 0.1 M PBS (pH 9.0) containing 0.1 M $K_2S_2O_8$. The unique nanostructure of PAMAM has numerous functional amino groups that can assemble a large number of CdSe QDs to develop the amplified PAMAM-Au-CdSe QDs ECL signal probe.

Li et al. [63] designed a signal-off ECL immunosensor for ketamine detection based on PAMAM-coated carbon dots (CDs) as signal material. The CDs about 2 nm was synthesized by a one-step hydrothermal synthetic route using PAMAM as platform and passivant. The ECL immunosensor was fabricated by electrochemical depositing AuNPs on the surface of GCE and then drop-coating PAMAM-CDs on the surface of the electrodeposition AuNPs film and finally covalent coupling anti-ketamine antibody on the surface of the PAMAM-CDs using glutaraldehyde as well as blocking the remaining binding sites using BSA for non-specific absorption. The designed immunosensor was successfully applied to the determination of ketamine in blood plasma.

Zhu et al. [64] reported an efficient ECL resonance energy transfer (RET) system between $ZnCd_{14}S$ as a donor and Au-Cu alloy nanocrystals as an acceptor for highly sensitive and excellent specific detection of target insulin. $ZnCd_{14}S$ showed the stable and strong ECL emission in the presence of coreactant $K_2S_2O_8$, which was enhanced by AuNPs and mesoporous carbons, and further quenched by Au-Cu alloy nanocrystals. The ECL immunosensor was fabricated by drop-coating Ab1/Au-ZnCd14S/NH_2-NMCs composites and BSA. After the immunosensor was incubated with insulin sample and Ab_2/Au-Cu solution, the ECL measurement was performed in PBS- 60 mM $K_2S_2O_8$ (pH 7.4). Much low limit of detection (0.03 pg/mL) was achieved using this ECL-RET immunosensor.

4. ECL aptamer-based biosensors

In the past three decades, antibody was widely utilized as molecular recognition substance with high affinity and specificity in ECL immunoassays/immunosensors. However, antibodies have limitations such as production in vivo, limited target analytes, limited shelf lives, and temperature-sensitive to undergo denaturation. Aptamers are oligonucleotides (DNA or RNA) selected from combinatorial libraries using SELEX (systemic evolution of ligands by exponential enrichment) that can bind with high affinity and specificity to a wide range of target molecules, such as inorganic ions, organic molecules, drugs, and proteins [65–67]. Aptamers as molecular recognition substances appear to be excellent alternatives to antibodies due to their ease of production in vitro, wide target range, modification ease, reversible thermal denaturation, and unlimited shelf life. Recently, a number of ECL aptamer-based biosensors for the determination of a small molecule drug have been designed employing aptamers as molecular recognition elements and Ru(II) complex served as an ECL label [68–73], as listed in Table 3.

Sun et al. [68] designed a highly sensitive and reusable aptamer-based biosensor for the detection of cocaine using a double covalent coupling method for the fabrication of the biosensor. The ECL sensor was constructed by covalent coupling of amino containing $Ru(bpy)_3^{2+}$ derivative-tagged cocaine aptamer to the surface of a paraffin-impregnated graphite electrode that had been covalently modified with a monolayer of 4-aminobenzene sulfonic acid via electrochemical oxidations. The ECL aptamer-based biosensor showed an extremely low detection limit of 10 pM for cocaine, and offered a good selectivity toward cocaine, heroin, and caffeine. Cai et al. [69] also reported an ECL “sandwich” aptamer-based biosensor for the detection of cocaine. The biosensor was fabricated by self-assembling a capture probe on the surface of Au electrode. After the biosensor was incubated in a mixture solution containing the RuSiNPs-labeled signal probe and cocaine, the ECL measurement was performed in PBS (pH 7.4) and (dibutylamino) ethanol. The biosensor was applied to detect trace amounts of cocaine on banknotes with satisfactory results.

Wang et al. [70] developed a label-free bifunctional ECL aptasensor for parallel detection of small molecule (adenosine) and protein (lysozyme) based on switching structures of aptamers from DNA/DNA duplex to DNA/target complex. Thiolated single-stranded DNA (DNA1) containing adenosine aptamer and sequence complementary to DNA2 was first immobilized on the Au electrode via sulfur–gold affinity, and DNA2 containing lysozyme aptamer and sequence complementary to DNA3 was bound to the electrode via forming DNA/DNA duplex. Gold nanoparticles functionalized with DNA3 (DNA3-AuNPs) were used as signal amplifiers to enhance the sensitivity of the aptasensor. With $Ru(phen)_3^{2+}$ as the signal transducer, the aptasensor exhibited high sensitivity and specificity. This proposed method provided a promising platform for ECL parallel detection of small molecules and protein.

Li et al. [71] developed a simple and highly sensitive ECL adenosine aptasensor by adsorbing ruthenium complex-tagged aptamer on single-walled carbon nanotubes (SWCNTs). Ru1-tagged aptamer utilized as an ECL probe and the ECL probe was non-covalently assembled on the surface of the SCWNTs to form the ECL probe/SWCNTs composite. Analyte adenosine was bound with the aptamer of the ECL probe on the SWCNTs so that the ECL probe was moved away or dropped from the SWCNTs, resulting in the decrease of ECL signal. This work demonstrated that the strategy of simply adsorbing ECL probe/SWCNTs composites as a biosensing platform is a promising approach to design ECL aptasensors with high sensitivity and selectivity.

Feng et al. [72] developed a “dual-potential” ECL aptasensor array for simultaneous detection of malachite green (MG) and chloramphenicol (CAP) in one single assay. The screen printed carbon electrode (SPCE) substrate consisted of a common Ag/AgCl reference electrode, a carbon counter electrode and two carbon working electrodes (WE1 and WE2). In this system, CdS QDs were modified on WE1 as cathode ECL emitters and luminol-gold nanoparticles (Lu-AuNPs) were modified on WE2 as anode ECL emitters. Then the MG aptamer complementary strand and CAP aptamer complementary strand were attached on CdS QDs and Lu-AuNPs, respectively. The cDNA would hybridize with the corresponding aptamer that was respectively tagged with

Table 3
ECL aptamer-based sensors (aptasensor).

Drugs/analytes	Systems	Linear ranges	LOD (S/N = 3)	Samples	Ref.
Cocaine	Signal-on, Ru1-tagged aptamer/4-ABSA/ PIGE in 0.10 M PBS-0.10 M TPA (pH 7.40)	50 pM to 5 nM	10 pM	–	[68]
Cocaine	Capture apt-probe assembled on Au electrode, signal apt-probe labeled with RuSiNPs in PBS (pH7.4)-1.0 M dibutylamino-ethanol	0.01–1.0 nM	3.7 pM	Banknotes	[69]
Adenosine	Sandwich type, Au electrode, $Ru(phen)_3^{2+}$ as interactor	0.5–7 nM	0.15 nM	–	[70]
Adenosine	Signal off, Ru1-tagged aptamers/SWCNTs/GCE	0.1–500 nM	0.05 nM	–	[71]
Malachite green (MG) Chloramphenicol (CAP)	Aptasensor array using CdS QDs and luminol-AuNPs as labels, 0.1 M PBS (pH 8.0)-12 mM H_2O_2	0.1–100 nM MG 0.2–150 nM CAP	0.03 nM MG 0.07 nM CAP	Fish samples	[72]
Biowarfare agents	SPC electrode array (ss-DNA), $Ru(bpy)_3^{2+}$-labeled DNA reporter, sandwich-type hybridisation assay in 300 mM PB-100 mM TPA-0.02% Triton X-100 (pH 7.6)	0–16 nM	0.6–1.2 nM for six targets	–	[73]

cyanine dye (Cy5) (as quenchers of CdS QDs) and chlorogenic acid (CA) (as quenchers of Lu-AuNPs) using poly(ethylenimine) (PEI) as a bridging agent. PEI could lead to a large number of quenchers on the aptamer, which increased the quenching efficiency. Upon MG and CAP adding, the targets could induce strand release due to the high affinity of analytes toward aptamers. Meanwhile, it could release the Cy5 and CA, which recovered cathode ECL of CdS QDs and anode ECL of L-AuNPs simultaneously. This "dual-potential" ECL strategy could be used to detect MG and CAP in real fish samples.

Spehar-Délèze et al. [73] developed an ECL DNA sensor array for multiplex detection of six biowarfare agents. Aminated-DNA capture probes were covalently immobilised on activated-carbon electrodes and subsequently hybridized to target strands. Detection was achieved via a sandwich-type assay after $Ru(bpy)_3^{2+}$-labeled reporter probes were hybridized to the formed probe–target complexes. The assay was performed in an automated microsystem in a custom designed ECL detection box with integrated fluidics, electronics, and movable photomultiplier detector. Detection of six targets on a single chip was achieved with subnanomolar detection limits.

5. ECL enzyme-based biosensors

Enzymes are proteins that catalyze chemical reactions in living systems. Such catalysts are not only efficient but also extremely selective. Hence, enzymes combine the recognition and amplification steps, as needed, for many sensing applications. Enzyme-based biosensors (enzyme electrodes) are based on the coupling of a layer of an enzyme with an appropriate electrode. Such electrodes combine the specificity of the enzyme for its substrate with the ECL reagents. As a result of such coupling, ECL enzyme-based sensors have shown to be extremely useful for monitoring a wide variety of analytical importance substrates in clinical, environmental and food samples. Several recent papers have addressed the application of nanomaterials in the typical luminol-H_2O_2 system [74–81], where luminol as ECL reagents was coupled with oxidases such as glucose oxidase (GOD), acetylcholinesterase (AChE), cholesterol oxidase (ChOx), and choline oxidase (ChOX) for the determination of glucose and organophosphate pesticides, as listed in Table 4.

In ECL GOD-based sensor for glucose, several nanoparticles have been developed to increase the renewability and sensitivity, and design a non-enzyme sensor. Xiong et al. [74] reported a GOD-based ECL biosensing system for glucose using Fe_3O_4 magnetic nanoparticles. GOD was covalently cross-linked to the surface of 3-(aminopropyl) triethoxysilane-coated magnetic Fe_3O_4 nanoparticles using glutaraldehyde as a linker regent. The composite particles of Fe_3O_4/GOD were adhered onto solid paraffin carbon paste electrode surface by magnetic force to act as an ECL sensor. ECL could be obtained by the reaction between the immobilized luminol and hydrogen peroxide produced by enzymatic reaction of GOD. The electrode surface was easily renewable. The proposed biosensor has been applied for the determination of glucose in plasma samples. Tian et al. [75] also reported a GOD-based ECL biosensing system for glucose. The biosensor was fabricated by drop-coating a mixture platinum nanoflowers, graphene oxide, Nafion and GOD onto the surface of GCE, and displayed a high electrocatalytic activity towards generated hydrogen peroxide. Thus, the sensitivity of the designed biosensor was improved. The fabricated biosensor was applied to determine glucose concentration in glucose injection, glucose functional drink, and blood serum.

Liu et al. [76] reported a signal-off ECL non-enzyme biosensor for the determination of glucose based on the integration of chitosan, CdTe QDs and AuNPs on GCE. Chitosan displays high water permeability, hydrophilic property, strong hydrogelability and good adhesion to load the double nanoparticles to the glassy carbon electrode surfaces. AuNPs are efficient GOD-mimic to catalytically oxidize glucose, similar to the natural process. Upon the addition of glucose, the AuNPs catalyzed glucose to produce gluconic acid and hydrogen peroxide (H_2O_2) based on the consumption of dissolved oxygen (O_2), which resulted in a quenching effect on the ECL emission. The proposed biosensor has been employed for the detection of glucose in human serum samples with satisfactory results.

Guan et al. [77] reported ECL imaging methods combining microfluidic cloth-based analytical devices (μCADs) for the determination of glucose. Wax screen-printing is employed to make cloth-based microfluidic chambers which are patterned with carbon SPE to create truly disposable, simple, and inexpensive sensors which can be read with a low-cost, portable charge coupled device (CCD) imaging sensing system. This work demonstrates that μCADs with ECL detection could provide a new sensing platform for point-of-care testing, public health, food safety detection and environmental monitoring in remote regions, developing or developed countries.

Organophosphates very often refer to a group of insecticides (pesticides) that act on the enzyme acetylcholinesterase (AChE). Today, organophosphates make up about 50% of the killing agents in chemical pesticides. Organophosphates pesticides (OPs) exposure is a growing global health problem, posing a highly severe threat on human life. High-level exposure to OPs results in the inhibition of AChE activity, which may cause respiratory paralysis and death. Several pieces of work have addressed ECL AChE/ChOx-based sensors for organophosphate pesticides [78–80]. Liang et al. [78] developed a signal-on ECL AChE-based

Table 4
Enzyme-based ECL sensors.

Drugs/analytes	Systems	Linear ranges	LOD (S/N = 3)	Samples	Ref.
Glucose	GOD/Fe_3O_4/SPCE in 0.1 M borate buffer-0.5 mM luminol (pH 8.0)	10 μM to 10 mM	1.0 μM	Serum	[74]
Glucose	GOD/GO/Pt NFs/GCE in 0.1 M PB-0.1 mM luminol (pH 7.4)	5–80 μM, 80–1000 μM	2.8 μM	Blood serum	[75]
Glucose	Signal-off, CdTe QDs/AuNPs/CHIT/GCE in 0.1 M PBS (pH8.0)	0.01–10 mM	5.28 μM	Human blood	[76]
Glucose	Microfluidic cloth-based analytical devices (μCADs), CCD camera, GOD/SPEs in PB (pH 7.4)-2.5 mM luminol	0–10 mM	0.038 mM	Artificial urine	[77]
Methyl parathion	Signal-on, AChE/CdTe QDs/GNs/GCE in 0.1 M PBS (pH 8.0)-1.0 mM acetylthiocholine chloride	0.2–10 ng/mL, 20–150 ng/mL	0.06 ng/mL	Cabbages	[78]
Malathion (MA) Methylparathion (ME) Chlorpyrifos (CH) Pesticide dufulin (PD)	Enhancing ECL of luminol biosensor AChE-ChOx/PtNPs-AuNPs/MWCNTs/GCE in PBS (pH 8.0)-2.0 mM ATCI-0.4 mM luminol	0.1–50 nM MA, 0.1–50 nM ME, 0.1–50 nM CH, 50–500 nM PD	0.16 nM MA, 0.09 nM ME, 0.08 nM CH, 29.7 nM PD	Cabbages	[79]
Ethyl paraoxon	AChE/C-g-C_3N_4-PEI/GCE in 0.1 M PBS (pH 7.0) with 0.05 M $K_2S_2O_8$ and 0.12 mM ATCl	1.0 pM to 5.0 μM	0.3 pM	Cabbages	[80]
Acetylcholine	Fe_3O_4-TiO_2-AChE-ChOx-GR-AuNPs-CS/GCE, luminol ECL system	6.7 nM to 0.92 mM	2.2 nM	Human serum	[81]

sensor for the detection of OPs. The electrochemically synthesized graphene nanosheets (GNs) were selected as a supporting material to anchor CdTe QDs, exhibiting a significantly amplified ECL signal of QDs. On the basis of the effect of OPs on the ECL signal of AChE-QDs-GNs modified GCE, a highly sensitive GNs-anchored-QDs-based signal-on ECL biosensor was developed for sensing OPs, combined with the enzymatic reactions and the dissolved oxygen as coreactant. The conditions for OPs detection were optimized by using methyl parathion (MP) as a model OP compound. Under the optimized experimental conditions, such a newly designed system shows remarkably improved sensitivity and selectivity for the sensing of OPs. Miao et al. [79] also developed an ECL biosensing system for individual detection of different OPs in food samples. Bi-metallic Pt-Au nanoparticles were electrodeposited on MWNTs-modified GCE to increase the surface of electrode and ECL signals of luminol. Biocomposites of enzymes from acetylcholinesterase and choline oxidase (AChE and ChOx) were immobilized onto the electrode surface to produce massive H_2O_2, thus amplifying ECL signals. Based on the dual-amplification effects of nanoparticles and H_2O_2 produced by enzymeaticre-actions, the proposed biosensor exhibits highly sensitivity. The proposed biosensing approach was then used for detecting OPs by inhibition of OPs on AChE. The resulting biosensor was further validated by assessment of OPs residues in cabbage, which showed a fine applicability for the detection of OPs in the realistic samples. Wang et al. [80] reported a signal-on ECL biosensor for the detection of OPs based on a novel composite of carboxylated graphitic carbon nitride-poly (ethylenimine) (C-g-C_3N_4-PEI) and AChE. The C-g-C_3N_4-PEI nanocomposite which was prepared through covalent bonding between the COOH of C-g-C_3N_4 and the NH_2 of PEI exhibited significantly enhanced ECL efficiency and stability. $K_2S_2O_8$ as the coreactant of C-g-C_3N_4-PEI could be consumed by thiocholine, produced by the hydrolysis of acetylthiocholine (ATCl) in the presence of AChE. Since OPs are one of AChE inhibitors the consumption of coreactant $K_2S_2O_8$ decreased with the increasing concentration of OPs, thus enhancing ECL signal. This novel strategy has the advantages of fine practicality, good stability and reproducibility, which might provide a new promise for OPs detection in real-life samples.

Acetylcholine is one of the most important neurotransmitters. An abnormal level of the metabolites leads to some neuropsychiatric disorders. Blocking, hindering or mimicking the action of acetylcholine has many uses in medicine. Acetylcholine itself does not have a therapeutic value as a drug for intravenous administration because of its multi-faceted action (non-selective) and rapid inactivation by cholinesterase. However, it is used in the form of eye drops to cause constriction of the pupil during cataract surgery, which facilitates quick post-operational recovery. Wu et al. [81] developed an ECL biosensing system for the detection of acetylthiocholine chloride (ATCI). First, graphene-AuNPs-chitosan (GR-AuNPs-CS) nanocomposite, which possesses the property of intensification effect on the ECL of luminol, was electrochemically deposited on the bare GCE. Then, a biofunctional Fe_3O_4-TiO_2-AChE-ChO biocomposite was unprecedentedly prepared which exhibited the mimic peroxidase activity of Fe_3O_4 and the enhancement effect of TiO_2 on the ECL intensity of luminol. Subsequently, the substrate ATCl was hydrolyzed by AChE of AChE-ChO multiple enzymes to generate thiocholine, which was then catalyzed by ChO to produce H_2O_2 in situ. H_2O_2, as the coreactant of luminol-ECL system, can enhance the ECL intensity of luminol for detection of ATCI. The resulting biosensor showed high sensitivity, good selectivity, reproducibility and ideal stability for the detection of ATCl in real samples. Thus the strategy could provide a promising avenue to develop efficient biosensors to determine biologically significant compounds in clinical application.

6. ECL chiral sensors and MIP sensors

ECL chiral sensor is a new type of ECL biosensors using chiral elements to recognize chiral drugs. β-cyclodextrin (β-CD), a cyclic oligosaccharide consisting of seven glucose units, has been widely used as a chiral selector to construct chiral materials for specific recognition of guest molecules for the enantioselective detection of amino acids enantiomers or chiral drugs. Three papers on ECL chiral sensors using different ECL reagents have been reported in recent years [82–84], listed in Table 5. Dai et al. [82] developed a new ECL sensing platform utilizing β-CD functionalized carbon nanohorns (β-CD/CNHs) as an ECL amplification and sensing element for sensitive detection of naringin with good specificity and excellent stability. The biosensor was fabricated by modifying β-CD/CNHs onto GCE. The ECL measurement was performed in PBS (pH 8.0) containing luminol and naringin. This β-CD/CNHs-based ECL biosensor could selectively and sensitively detect naringin down to nanomolar level. Wang et al. [83] reported a stereo-selective ECL sensor for discriminating proline enantiomers. The

sensor was fabricated by immobilizing $Ru(bpy)_3^{2+}$–gold nanoparticles and β-cyclodextrin–reduced graphene oxide (β-CD–rGO) on GCE. When the developed sensor interacted with proline enantiomers, obvious difference of ECL intensities towards L- and D-proline was observed, and a larger intensity was obtained from D-proline. Lin et al. [84] reported a stereo-selective ECL sensor for specific recognition of penicillamine (Pen) enantiomers. The sensor was fabricated by immobilizing hemoglobin (Hb) as chiral selector and gold nanoparticles functionalized graphite-like carbon nitride nanosheets composite (AuNPs-g-C_3N_4NHs) luminophore onto GCE. After L-Pen or D-Pen was bounded on the surface of the sensor, and the ECL signal decreased gradually with increasing the concentration of the Pen enantiomer. The sensitivity of the sensor for L-Pen is higher than that for D-Pen.

Molecular imprinted polymer (MIP) can enrich and separate target molecules in complex samples, which can not only eliminate the interferences within samples, but also further improve the sensitivity of the detection methods. In this case of the antibodies and aptamers unavailable for some drugs, MIP could be used as molecular recognition elements. Even if the binding constants of MIP are generally lower than that of antibodies and aptamers, and they have a low electric conductivity, MIP have great chemical and thermal stability and are easy to be prepared. Much effort has been made to develop ECL MIP sensors for pharmaceuticals in recent years [85–93], listed in Table 5.

Wu et al. [85] reported a MIP-based ECL sensor for the detection of isoniazid (INH). The sensor was fabricated by electrochemical copolymerization of acrylamide and N,N′-methylene diacrylamide in the presence of INH template molecules onto the surface of $Ru(bpy)_3^{2+}$/AuNPs/MWCNTs-Nafion modified GCE. The ECL measurement was performed in 0.1 M PBS (pH7.4) containing isoniazid as a co-reactant. The concentration of isoniazid was quantified by the increased ECL intensity. The sensor has been applied to the determination of INH in human urine and pharmaceutical samples.

Han et al. [86] reported a MIP ECL sensor for the detection of the hidden drugs (methamphetamine). The sensor was fabricated by coating Nafion-MWCNTs on the surface of GCE, then adsorbing $Ru(bpy)_3^{2+}$ on Nafion/MWCNTs/GCE, and finally modifying MIP with methamphetamine as the template molecule using sol-gel technology. The as-prepared sensor exhibited a very high sensitivity and excellent selectivity toward methamphetamine. The sensor was applied to detect the drug odors in a closed container.

Li et al. [87] reported a signal-on MIP ECL sensor for the detection of thifensulfuronmethyl (TFM) herbicide using core–shell imprinted nanoparticles. The core–shell imprinted nanoparticles were prepared by a surface monomer-directing strategy for imprinting TFM at the surface of 3-methacryloxypropyl trimethoxysilane modified silica particles. The ECL sensor was prepared by depositing the core–shell imprinted nanoparticles/chitosan composite film on the surface of GCE and further removing silica cores from the composite film. The ECL intensity of the sensor was enhanced by the adsorbed TFM molecules in the composite film in 0.1 M KCl-5.0×10^{-5} M luminol (pH = 12.0).

Shang et al. [88] reported a MIP ECL sensor for the detection of heroin. The sensor was prepared by re-modifying the MIP film onto $Ru(bpy)_3^{2+}$ modified GCE. The as-prepared sensor displayed high sensitivity and excellent selectivity for heroin and was applied for the determination of heroin in urine and saliva with the recovery rates in the range of 97%–104%.

Jiang et al. [89] developed a signal-on ECL sensor for the detection of diethylstilbestrol (DES) using magnetic surface magnetic molecular imprinting polymers (MMIPs) as capture probe and an aptamer-labeled CdS QDs as signal probe. When the target DES, MMIPs and CdS-apt were incubated together, a sandwich MMIPs-

Table 5
ECL Chiral sensors and MIP sensors.

Drugs/analytes	Systems	Linear ranges	LOD (S/N = 3)	Samples	Ref.
Naringin	Decreasing ECL of luminol sensor β-CD/carbon nanohorns/GCE in PB (pH 8)-2.0×10^{-5} M luminol	1 nM to 1 μM	0.8 nM	Cough mixture	[82]
L-/D-proline	Enhancing ECL of $Ru(bpy)_3^{2+}$ chiral sensor β-CD-rGO/$Ru(bpy)_3^{2+}$/AuNPs/Nafion/GCE	0.1–5.0 mM	25 μM L-pro 33 μM D-pro	–	[83]
Penicillamine enantiomers	Hb/Au-g-C_3N_4/GCE (−1.1 V∼0 V) in 0.1 M PBS (pH 7.4)-1.0 mM $K_2S_2O_8$	0.10–5.0 mM	31 μM L-pen 33 μM D-Pen	Tablet	[84]
Isoniazid	Enhancing ECL of $Ru(bpy)_3^{2+}$ sensor MIP/$Ru(bpy)_3^{2+}$/ AuNPs/MWCNTs/GCE in 0.1 M PBS (pH 7.0)	0.1–110 μg/mL	0.08 μg/mL	Human urine and tablets	[85]
Methamphetamine	Enhancing ECL of $Ru(bpy)_3^{2+}$ sensor MIP/$Ru(bpy)_3^{2+}$ /Nafion/MWCNTs/GCE in 0.1 M PBS (pH 8.0)	0.01 pM to 0.1 nM	4.0 fM	Hidden drugs	[86]
Thifensulfuronmethyl	Enhancing ECL of luminol Sensor MIP@SiO_2 NPs/ CHIT/GCE in 0.1 M KCl-0.5 μM luminol (pH 12.0)	0.5 nM to 0.1 μM	0.32 nM	Sulfonylurea herbicides mixture	[87]
Heroin	Enhancing ECL of $Ru(bpy)_3^{2+}$ sensor MIP/$Ru(bpy)_3^{2+}$/Nafion/GCE in 0.1 M PBS (pH 7.0)	0.01 pM to 0.1 nM	4.0 fM	Urine and saliva	[88]
Diethylstilbestrol	Signal-on, MIPs-CdS-Apt/SPCE (−1.1 V) in 5 mM H_2O_2	30–100 ng/mL	0.1 pg/mL	Spiked fish	[89]
2-methyl-4-chlorophenoxyacetic acid	MIP/MoS_2-graphene QDs/GCE in 0.01 M PBS (pH 7.4)-100 mM $K_2S_2O_8$	10 pM to 0.1 μM	5.5 pM	Tap water, lake water, food sample oat	[90]
Melamin	Enhancing ECL of $Ru(bpy)_3^{2+}$@SiO_2 NPs/GCE in 0.1 M PB (pH 9.0)	1 pM to 0.1 μM	0.5 pM	Milk	[91]
Clenbuterol	MIP ECL sensor UCNPs/rGO/GCE in 0.1 M PB (pH 7.4)-0.1 M $K_2S_2O_8$	10 nM to 100 μM	6.3 nM	Pig samples including pork, liver and kidney	[92]
Ochratoxin A	MIP/$Ru(bpy)_3^{2+}$-doped SiNPs/GCE in 0.1 M PBS (pH 7.0) containing 2 mM TPA	0.1 pg/mL to 14.76 ng/mL	0.027 pg/mL	Corn	[93]

DES-CdS-apt composite was constructed. After this conjugated composite was adsorbed on the surface of a SPCE by external magnetic field, the ECL signal at potential of − 1.1 V was recorded. The signal intensity was in proportion to the logarithm of DES concentration. Several fish samples were tested by the sensor which showed high selectivity and good recoveries between 80% and 120% with consistent results as that of conventional ELISA.

Yang et al. [90] developed a MIP ECL sensor for the detection of 2-methyl-4-chlorophenoxyacetic acid (MCPA) which is a widely used phenoxy herbicide. The sensor was fabricated by electrochemically synthesizing the MIP film on a modified MoS_2-G QDs/GCE using repetitive CV scans in a deoxygenated acetate buffer (pH 5.2) containing 5 mM o-phenylenediamine and MCPA. The ECL intensity of the MoS_2-G QDs hybrid nanocomposite on GCE was enhanced in 13, 185 and 596-folds larger than that of G QDs, MoS_2 modified electrodes and bare electrode, respectively. The developed sensor exhibited high sensitivity, good selectivity, reproducibility and stability, suggesting the potential for detecting pesticides and veterinary drugs at trace levels in food safety and environmental control.

7. Conclusion and perspectives

The rapid development of ECL biosensing methods over the past years has ushered in ECL as a powerful tool for pharmaceutical analysis. The ECL biosensors such as immunoassays, immunosensors, aptamer-based biosensors, enzyme-based biosensors, chiral sensors and MIP-based sensors were designed for the pharmaceutical quantification. Nanomaterials such as semiconductor quantum dots and carbon dots were adopted in ECL biosensing methods as a novel ECL emission materials and enhancers in typical ECL systems. ECL aptamer-based biosensors, chiral sensors and MIP-based sensors have shown to be competitive to conventional approaches of ECL immunosensors and enzyme-based biosensors. ECL imaging biosensing methods have been demonstrated for being capable of multiplexing detection of pharmaceuticals. The ECL biosensors that are capable of multiplexing detection with high sensitivity, low detection limit, and good selectivity and stability will be the ongoing interest of the research community.

In future work, the development of novel ECL reagents/materials for ECL biosensing methods in pharmaceutical analysis will be highly demanded. Developing of new molecular recognition elements from aptamers or peptides can be useful, since they are more stable and can be reproducibly synthesized in vitro. To meet the requirement of quality control for pharmaceuticals and clinic testing, new ECL biosensing methods such as highly sensitive and label-free immunoassays with nanotechnology should be developed and improved. The fast, standardized, and commercialized methods will be the primary goal. Bearing in mind that most drugs bind to red blood cells and plasma proteins, only the unbound drug can interact with the targets. Therefore, the determination of the free drug concentration may provide more valuable information compared to that in whole blood. The hybrid of ECL and other techniques could be a promising approach to developing new instruments and providing valuable tools. A new trend of point-of-care testing by using smartphone read-out is emerging [94–98], which may inspire designing novel ECL biosensing methods for pharmaceutical analysis. We believe that ECL biosensing methods will be widely applied in pharmaceutical analysis.

References

[1] M.M. Richter, Electrochemiluminescence (ECL), Chem. Rev. 104 (2004) 3003–3036.
[2] W. Miao, Electrogenerated chemiluminescence and its biorelated applications, Chem. Rev. 108 (2008) 2506–2553.
[3] G.F. Blackburn, H.P. Shah, J.H. Kenten, et al., Electrochemiluminescence detection for development of immunoassays and DNA probe assays for clinical diagnostics, Clin. Chem. 37 (1991) 1534–1539.
[4] D.M. Hercules, Chemiluminescence resulting from electrochemically generated species, Science 145 (1964) 808–809.
[5] R.E. Visco, E.A. Chandross, Electroluminescence in solutions of aromatic hydrocarbons, J. Am. Chem. Soc. 86 (1964) 5350–5351.
[6] K.S.V. Santhanam, A.J. Bard, Chemiluminescence of electrogenerated 9,l0-diphenyl- anthracene anion radical, J. Am. Chem. Soc. 87 (1965) 139–140.
[7] R.T. Dufford, D. Nightingale, L.W. Gaddum, Luminescence of Grignard compounds in electric and magnetic fields, and related electrical phenomena, J. Am. Chem. Soc. 49 (1927) 1858–1864.
[8] N. Harvey, Luminescence during electrolysis, J. Phys. Chem. 33 (1929) 1456–1459.
[9] G.M. Greenway, Analytical applications of electrogenerated chemiluminescence, Analyst 9 (1994) 200–203.
[10] A.W. Knight, A review of recent trends in analytical applications of electrogenerated chemiluminescence, TrAC Trends Anal. Chem. 18 (1999) 47–62.
[11] X. Yin, S.J. Dong, E.K. Wang, Analytical applications of the electrochemiluminescence of tris (2,2′-bipyridyl) ruthenium and its derivatives, TrAC Trends Anal. Chem. 23 (2004) 432.
[12] X.B. Yin, E.K. Wang, Capillary electrophoresis coupling with electrochemiluminescence detection: a review, Anal. Chim. Acta 533 (2005) 113–120.
[13] M. Su, W. Wei, S. Liu, Analytical applications of the electrochemiluminescence of tris(2,2′-bipyridyl)ruthenium(II) coupled to capillary/microchip electrophoresis: a review, Anal. Chim. Acta 704 (2011) 16–32.
[14] K. Muzyka, Current trends in the development of the electrochemiluminescent immunosensors, Biosens. Bioelectron. 54 (2014) 393–407.
[15] Z. Fu, W. Wei, C. Li, et al., Electrochemiluminescent immunosensing and its application in biological and pharmaceutic alanalysis, Sci. China-Chem. 41 (2011) 773–784.
[16] H.L. Qi, Y. Peng, Q. Gao, et al., Application of nanomaterials in electrogenerated chemiluminescence biosensors, Sensors 9 (2009) 674–695.
[17] J.P. Lei, H.X. Ju, Fundamentals and bioanalytical applications of functional quantum dots as electrogenerated emitters of chemiluminescence, TrAC Trends Anal. Chem. 30 (2011) 1351–1359.
[18] Y.H. Xu, J.Q. Liu, C.L. Gao, et al., Applications of carbon quantum dots in electrochemiluminescence: a mini review, Electrochem. Commun. 48 (2014) 151–154.
[19] Y. Chen, S.W. Zhou, L.L. Li, et al., Nanomaterials-based sensitive electrochemiluminescence biosensing, Nano Today 12 (2017) 98–115.
[20] H.L. Qi, J. Chang, S.H. Abdelwahed, et al., Electrochemistry and electrogenerated chemiluminescence of π-stacked poly(fluorenemethylene) oligomers. Multiple, interacting electron transfers, J. Am. Chem. Soc. 134 (2012) 16265–16274.
[21] H.L. Qi, J.J. Teesdale, R.C. Pupillo, et al., Synthesis, electrochemistry, and electrogenerated chemiluminescence of two BODIPY-appended bipyridine homologues, J. Am. Chem. Soc. 135 (2013) 13558–13566.
[22] H. Qi, Y.H. Chen, C.H. Cheng, et al., Electrochemistry and electrogenerated chemiluminescence of three phenanthrene derivatives, enhancement of radical stability, and electrogenerated chemiluminescence efficiency by substituent groups, J. Am. Chem. Soc. 135 (2013) 9041–9049.
[23] H.L. Qi, C.X. Zhang, Z. Huang, et al., Electrochemistry and electrogenerated chemiluminescence of 1,3,5-tri(anthracen-10-yl)-benzene-centered starburst oligofluorenes, J. Am. Chem. Soc. 138 (2016) 1947–1954.
[24] Y. Zhao, Q. Zhang, K. Chen, et al., Triphenothiazinyl triazacoronenes: donor–acceptor molecular graphene exhibiting multiple fluorescence and electrogenerated chemiluminescence emissions, J. Mater. Chem. C 5 (2017) 4293–4301.
[25] Y. Zhao, D. Xue, H.L. Qi, et al., Twisted configuration pyrene derivative: exhibiting pure blue monomer photoluminescence and electrogenerated chemiluminescence emissions in non-aqueous media, RSC Adv. 7 (2017) 22882–22891.
[26] J.K. Leland, M.J. Powell, Electrogenerated chemilumineecence: an oxidative-reduction type ECL reaction sequence using tripropylamine, J. Electrochem. Soc. 137 (1990) 3127–3131.
[27] X.M. Zhou, D.B. Zhu, Y.H. Liao, et al., Synthesis, labelling, and bioanalytical applications of a tris(2, 2′-bipyridyl) ruthenium (II) based electrochemiluminescence probe, Nat. Protoc. 9 (2014) 1146–1159.
[28] H.L. Qi, X.Y. Qiu, D.P. Xie, et al., Ultrasensitive electrogenerated chemiluminescence peptide-based method for the determination of cardiac troponin I incorporating amplification of signal reagent-encapsulated liposomes, Anal. Chem. 85 (2013) 3886–3894.
[29] H.L. Qi, M. Li, M.M. Dong, et al., Electrogenerated chemiluminescence peptide-based biosensor for the determination of prostate-specific antigen based on target-induced cleavage of peptide, Anal. Chem. 86 (2014) 1372–1379.
[30] J. Zhang, H.L. Qi, Z.J. Li, et al., Electrogenerated chemiluminescence bioanalytic system based on bio-cleavage of probes and homogeneous detection, Anal. Chem. 87 (2015) 6510–6515.
[31] X. Liu, M.M. Dong, H.L. Qi, et al., Electrogenerated chemiluminescence bioassay of two protein kinases incorporating peptide phosphorylation and versatile

probe, Anal. Chem. 88 (2016) 8720–8727.
[32] X.F. Wang, H.F. Gao, H.L. Qi, et al., Proximity hybridization-regulated immunoassay for cell surface protein and protein-overexpressing cancer cells via electrochemiluminescence, Anal. Chem. 90 (2018) 3013–3018.
[33] Y.Y. Zhou, H.F. Gao, X.M. Wang, Qi, et al., Electrogenerated chemiluminescence from heteroleptic iridium(III) complexes with multicolor emission, Inorg. Chem. 54 (2015) 1446–1453.
[34] Y. Zhao, X.L. Yang, D.J. Han, et al., Cyclometalated iridium-complex-based label-free supersandwich electrogenerated chemiluminescence biosensor for the detection of micro-RNA, ChemElectroChem 4 (2017) 1775–1782.
[35] C.X. Zhang, H.H. Zhang, M.L. Feng, Homogeneous electrogenerated chemiluminescence immunoassay using a luminol-labeled digoxin hapten, Anal. Lett. 36 (2003) 1103–1114.
[36] H.L. Qi, C.X. Zhang, Homogeneous electrogenerated chemiluminescence immunoassay for the determination of digoxin, Anal. Chim. Acta 501 (2004) 31–35.
[37] Z. Ding, B.M. Quinn, S.K. Haram, et al., Electrochemistry and electrogenerated chemiluminescence from silicon nanocrystal quantum dots, Science 296 (2002) 1293–1297.
[38] C.M. Cheng, Y. Huang, J. Wang, et al., Anodic electrogenerated chemiluminescence behavior of graphite-like carbon nitride and its sensing for rutin, Anal. Chem. 85 (2013) 2601–2605.
[39] J.J. Ji, J. Wen, Y.F. Shen, et al., Simultaneous noncovalent modification and exfoliation of 2D carbon nitride for enhanced electrochemiluminescent biosensing, J. Am. Chem. Soc. 139 (2017) 11698–11701.
[40] Y.Q. Lv, S.Y. Chen, Y.F. Shen, et al., Competitive multiple-mechanism-driven electrochemiluminescent detection of 8-hydroxy-2′-deoxyguanosine, J. Am. Chem. Soc. 140 (2018) 2801–2804.
[41] H. Wang, G.Q. Pu, S. Devaramani, et al., Bimodal electrochemiluminescence of G-CNQDs in the presence of double coreactants for ascorbic acid detection, Anal. Chem. 90 (2018) 4871–4877.
[42] Z.X. Zhou, Y.Y. Zhang, Y.F. Shen, et al., Molecular engineering of polymeric carbon nitride: advancing applications from photocatalysis to biosensing and more, Chem. Soc. Rev. 47 (2018) 2298–2321.
[43] C.X. Zhang, G.J. Zhou, Z.J. Zhang, et al., Highly sensitive electrochemical luminescence determination of thiamine, Anal. Chim. Acta 394 (1999) 165–170.
[44] C.X. Zhang, J.C. Huang, Z.J. Zhang, et al., Flow injection chemiluminescence determination of catecholamines with electrogenerated hypochlorite, Anal. Chim. Acta 374 (1998) 105–110.
[45] J.C. Huang, C.X. Zhang, Z.J. Zhang, Flow injection chemiluminescence determination of isoniazid with electrogenerated hypochlorite, Fresen J. Anal. Chem. 363 (1999) 126–128.
[46] C.X. Zhang, H.L. Qi, Highly sensitive determination of riboflavin based on the enhanced electrogenerated chemiluminescence of lucigenin at a platinum electrode in a neutral aqueous solution, Anal. Sci. 18 (2002) 819–822.
[47] R.S. Yalow, S.A. Berson, Immunoassay of endogenous plasma insulin in man, J. Clin. Investig. 39 (1960) 1157–1175.
[48] G. Brandhorst, F. Streit, J. Kratzsch, et al., Multicenter evaluation of a new automated electrochemiluminescence immunoassay for the quantification of testosterone compared to liquid chromatography tandem mass spectrometry, Clin. Biochem. 44 (2011) 264–267.
[49] L. Jafri, A.H. Khan, A.A. Siddiqui, et al., Comparison of high performance liquid chromatography, radio immunoassay and electrochemiluminescence immunoassay for quantification of serum 25 hydroxy vitamin D, Clin. Biochem. 44 (2011) 864–868.
[50] K.L. Johnson-Davis, S. De, E. Jimenez, et al., Evaluation of the abbott ARCHITECT i2000 sirolimus assay and comparison with the abbott IMx sirolimus assay and an established liquid chromatographytandem mass spectrometry method, Ther. Drug Monit. 33 (2011) 453–459.
[51] B. Eshratkhah, H. Rajabian, D. Namvar, et al., Comparative study on determination of plasma thyroid hormones by chemiluminescence and electrochemiluminescence immunoassay methods in sheep, Comp. Clin. Pathol. 20 (2011) 135–138.
[52] M. Shipkova, M. Vogeser, P.A. Ramos, et al., Multi-center analytical evaluation of a novel automated tacrolimus immunoassay, Clin. Biochem. 47 (2014) 1067–1077.
[53] M. Vogeser, M. Shipkova, R. Rigo-Bonnin, et al., Multicenter analytical evaluation of the automated electrochemiluminescence immunoassay for cyclosporine, Ther. Drug. Monit. 36 (2014) 640–650.
[54] M. Miura, S. Masuda, H. Egawa, et al., Inter-laboratory variability of current immunoassay methods for tacrolimus among Japanese hospitals, Biol. Pharm. Bull. 39 (2016) 1331–1337.
[55] M. Sasanoa, S. Kimura, I. Maeda, et al., Analytical performance evaluation of the Elecsys® Cyclosporine and Elecsys® Tacrolimus assays on the cobas e411 analyzer, Pract. Lab. Med. 8 (2017) 10–17.
[56] J. Lowe, M. Maia, E. Wakshull, et al., Development of a novel homogenous electrochemiluminescence assay for quantitation of ranibizumab in human serum, J. Pharm. Biomed. 52 (2010) 680–686.
[57] Z.Y. Li, Y.H. Wang, W.J. Kong, et al., Ultrasensitive detection of trace amount of clenbuterol residue in swine urine utilizing an electrochemiluminescent immunosensor, Sens. Actuators B 174 (2012) 355–358.
[58] Y.T. Liu, Q.Q. Zhang, H.J. Wang, et al., An electrochemiluminescence immunosensor for thyroid stimulating hormone based on polyamidoamine-norfloxacin functionalized Pd-Au core-shell hexoctahedrons as signal enhancers, Biosens. Bioelectron. 71 (2015) 164–170.
[59] Y. Yang, X.S. Wan, Q. Da, et al., Label-free immunosensor for morphine based on the electrochemiluminescence of luminol on indium–tin oxide coated glass functionalized with gold nanoparticles, Anal. Methods 7 (2015) 4502–4507.
[60] W.J. Fei, F.F. Chen, L. Sun, et al., Ultrasensitive electrochemiluminescent immunoassay for morphine using a gold electrode modified with CdS quantum dots, polyamidoamine, and gold nanoparticles, Microchim. Acta 181 (2014) 419–425.
[61] Q.H. Tang, F.D. Cai, A.P. Deng, et al., Ultrasensitive competitive electrochemiluminescence immunoassay for the β-adrenergic agonist phenylethanolamine A using quantum dots and enzymatic amplification, Microchim. Acta 182 (2015) 139–147.
[62] L.Y. Hu, T.T. Dong, K. Zhao, et al., Ultrasensitive electrochemiluminescent brombuterol immunoassay by applying a multiple signal amplification strategy based on a PAMAM-gold nanoparticle conjugate as the bioprobe and Ag@Au core shell nanoparticles as a substrate, Microchim. Acta 184 (2017) 3415–3423.
[63] Q.H. Li, W.J. Tang, Y. Wang, et al., Electrochemiluminescence immunosensor for ketamine detection based on polyamidoamine-coated carbon dot film, J. Solid State Electrochem. 19 (2015) 2973–2980.
[64] W.J. Zhu, C. Wang, X.J. Li, et al., Zinc-doping enhanced cadmium sulfide electrochemiluminescence behavior based on Au-Cu alloy nanocrystals quenching for insulin detection, Biosens. Bioelectron. 97 (2017) 115–121.
[65] D. Ellington, J.W. Szostak, In vitro selection of RNA molecules that bind specific ligands, Nature 346 (1990) 818–822.
[66] C. Tuerk, L. Gold, Systematic evolution of ligands by exponential enrichment: RNA ligands to bacteriophage T4 DNA polymerase, Science 249 (1990) 505–510.
[67] D. Ellington, J.W. Szostak, Selection in vitro of single-stranded DNA molecules that fold into specific ligandbinding structures, Nature 355 (1992) 850–852.
[68] B. Sun, H.L. Qi, F. Ma, et al., Double covalent coupling method for the fabrication of highly sensitive and reusable electrogenerated chemiluminescence sensors, Anal. Chem. 82 (2010) 5046–5052.
[69] Q.H. Cai, L.F. Chen, F. Luo, et al., Determination of cocaine on banknotes through an aptamer-based electrochemiluminescence biosensor, Anal. Bioanal. Chem. 400 (2011) 289–294.
[70] H.Y. Wang, W. Gong, Z.A. Tan, et al., Label-free bifunctional electrochemiluminescence aptasensor for detection of adenosine and lysozyme, Electrochim. Acta 76 (2012) 416–423.
[71] Z.J. Li, H.L. Qi, H.Y. Yang, et al., Simple and highly sensitive electrogenerated chemiluminescence adenosine aptasensor by adsorbing ruthenium complex-tagged aptamer on single-walled carbon nanotubes, Anal. Methods 6 (2014) 1317–1323.
[72] X.B. Feng, N. Gan, H.R. Zhang, et al., A novel “dual-potential" electrochemiluminescence aptasensor array using CdS quantum dots and luminol-gold nanoparticlesas labels for simultaneous detection of malachite green and chloramphenicol, Biosens. Bioelectron. 74 (2015) 587–593.
[73] A. Spehar-Délèze, R. Gransee, S. Martinez-Montequin, et al., Electrochemiluminescence DNA sensor array for multiplex detection of biowarfare agents, Anal. Bioanal. Chem. 407 (2015) 6657–6667.
[74] Z.G. Xiong, J.P. Li, L. Tang, et al., A novel electrochemiluminescence biosensor based on glucose oxidase immobilized on magnetic nanoparticles, Chin. J. Anal. Chem. 38 (2010) 800–804.
[75] X.T. Tian, S. Lian, L.M. Zhao, et al., A novel electrochemiluminescence glucose biosensor based on platinum nanoflowers/graphene oxide/glucose oxidase modified glassy carbon electrode, J. Solid State Electrochem. 18 (2014) 2375–2382.
[76] L.L. Liu, Q. Ma, Y. Li, et al., A novel signal-off electrochemiluminescence biosensor for the determination of glucose based on double nanoparticles, Biosens. Bioelectron. 63 (2015) 519–524.
[77] W.R. Guan, M. Liu, C.S. Zhang, Electrochemiluminescence detection in microfluidic cloth-based analytical devices, Biosens. Bioelectron. 75 (2016) 247–253.
[78] H. Liang, D.D. Song, J.M. Gong, Signal-on electrochemiluminescence of biofunctional CdTe quantum dots for biosensing of organophosphate pesticides, Biosens. Bioelectron. 53 (2014) 363–369.
[79] S.S. Miao, M.S. Wu, L.Y. Ma, et al., Electrochemiluminescence biosensor for determination of organophosphorous pesticides based on bimetallic Pt-Au/multi-walled carbon nanotubes modified electrode, Talanta 158 (2016) 142–151.
[80] B.X. Wang, X. Zhong, Y.Q. Chai, et al., Ultrasensitive electrochemiluminescence biosensor for organophosphate pesticides detection based on carboxylated graphitic carbon nitride-poly(ethylenimine) and acetylcholinesterase, Electrochim. Acta 224 (2017) 194–200.
[81] X.P. Wu, X. Zhong, Y.Q. Chai, et al., Electrochemiluminescence acetylcholine biosensor based on biofunctional AMs-AChE-ChO biocomposite and electrodeposited graphene-Au-chitosan nanocomposite, Electrochim. Acta 147 (2014) 735–742.
[82] H. Dai, C.P. Yang, X.L. Ma, et al., A highly sensitive and selective sensing ECL platform for naringin based on β-Cyclodextrin functionalized carbon nanohorns, Chem. Commun. 47 (2011) 11915–11917.
[83] Q.H. Wang, X. Lin, D.M. Guo, et al., A novel chiral electrochemiluminescence

sensor that can discriminate proline enantiomers, RSC Adv. 5 (2015) 94338–94343.
[84] X. Lin, S. Zhu, Q.H. Wang, et al., Chiral recognition of penicillamine enantiomers using hemoglobinand gold nanoparticles functionalized graphite-like carbon nitridenanosheets via electrochemiluminescence, Colloid Surf. B 148 (2016) 371–376.
[85] B.W. Wu, Z.H. Wang, Z.H. Xue, et al., A novel molecularly imprinted electrochemiluminescence sensor for isoniazid detection, Analyst 137 (2012) 3644–3652.
[86] C.F. Han, Z.Y. Zhang, H.H. Zhang, et al., Detection of hidden drugs with a molecularly imprinted electrochemiluminescence sensor, Anal. Methods 5 (2013) 6064–6070.
[87] H.F. Li, C.G. Xie, X.C. Fu, Electrochemiluminescence sensor for sulfonylurea herbicide with molecular imprinting core–shell nanoparticles/chitosan composite film modified glassy carbon electrode, Sens. Actuators B 181 (2013) 858–866.
[88] Z.Y. Shang, C.F. Han, Q.J. Song, An electrochemiluminescence sensor with molecularly imprinted polymer for heroin detection, Chin. J. Anal. Chem. 42 (2014) 904–908.
[89] Q.L. Jiang, D.N. Zhang, Y.T. Cao, et al., An antibody-free and signal-on type electrochemiluminescence sensor for diethylstilbestrol detection based on magnetic molecularly imprinted polymers-quantum dots labeled aptamer conjugated probes, J. Electroanal. Chem. 789 (2017) 1–8.
[90] Y.K. Yang, G.Z. Fang, X.M. Wang, et al., Electrochemiluminescent graphene quantum dots enhanced by MoS_2 as sensing platform: a novel molecularly imprinted electrochemiluminescence sensor for 2-methyl-4-chlorophenoxyacetic acid assay, Electrochim. Acta 228 (2017) 107–113.
[91] S. Lian, Z.Y. Huang, Z.Z. Lin, et al., A highly selective melamine sensor relying on intensified electrochemiluminescence of the silica nanoparticles doped with $[Ru(bpy)_3]^{2+}$/molecularly imprinted polymer modified electrode, Sens. Actuators B 236 (2016) 614–620.
[92] X.C. Jin, G.Z. Fang, M.F. Pan, et al., A molecularly imprinted electrochemiluminescence sensor based on upconversion nanoparticles enhanced by electrodeposited rGO for selective and ultrasensitive detection of clenbuterol, Biosens. Bioelectron. 102 (2018) 357–364.
[93] Q.L. Wang, M.M. Chen, H.Q. Zhang, et al., Solid-state electrochemiluminescence sensor based on RuSi nanoparticles combined with molecularly imprintedpolymer for the determination of ochratoxin A, Sens. Actuators B 222 (2016) 264–269.
[94] H. Cui, F. Paolucci, N. Sojic, et al., Analytical electrochemiluminescence, Anal. Bioanal. Chem. 408 (2016) 7001–7002.
[95] J.L. Delaney, E.H. Doeven, A.J. Harsant, et al., Use of a mobile phone for potentiostatic control with low cost paper-based microfluidic sensors, Anal. Chim. Acta 790 (2013) 56–60.
[96] E.H. Doeven, G.J. Barbante, A.J. Harsant, et al., Mobile phone-based electrochemiluminescence sensing exploiting the 'USB On-The-Go' protocol, Sens. Actuat. B 216 (2015) 608–613.
[97] L. Chen, C.S. Zhang, D. Xing, Paper-based bipolar electrode-electrochemiluminescence (BPE-ECL) device with battery energy supply and smartphone read-out: a hand held ECL system for biochemical analysis at the point-of-care level, Sens. Actuat. B 237 (2016) 308–317.
[98] W.Y. Gao, M. Saqib, L.M. Qi, et al., Recent advances in electrochemiluminescence devices for point-of-care testing, Curr. Opin. Electrochem. 3 (2017) 4–10.