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*Review*

# Recent Trends in Biosensors for Environmental Quality Monitoring

**Simona Gavrilaș, Claudiu Ștefan Ursachi, Simona Perța-Crișan and Florentina-Daniela Munteanu ***

Faculty of Food Engineering, Tourism and Environmental Protection, "Aurel Vlaicu" University of Arad, Tourism and Environmental Protection, 2-4 E. Drăgoi Str., 310330 Arad, Romania; simona.gavrilas@uav.ro (S.G.); claudiu.ursachi@uav.ro (C.Ș.U.); simona.perta-crisan@uav.ro (S.P.-C.)
* Correspondence: florentina.munteanu@uav.ro

**Abstract:** The monitoring of environmental pollution requires fast, reliable, cost-effective and small devices. This need explains the recent trends in the development of biosensing devices for pollutant detection. The present review aims to summarize the newest trends regarding the use of biosensors to detect environmental contaminants. Enzyme, whole cell, antibody, aptamer, and DNA-based biosensors and biomimetic sensors are discussed. We summarize their applicability to the detection of various pollutants and mention their constructive characteristics. Several detection principles are used in biosensor design: amperometry, conductometry, luminescence, etc. They differ in terms of rapidity, sensitivity, profitability, and design. Each one is characterized by specific selectivity and detection limits depending on the sensitive element. Mimetic biosensors are slowly gaining attention from researchers and users due to their advantages compared with classical ones. Further studies are necessary for the development of robust biosensing devices that can successfully be used for the detection of pollutants from complex matrices without prior sample preparation.

**Keywords:** environmental quality monitoring; emerging contaminants; detection; biosensing; mimetic biosensors



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## 1. Introduction

The modern world faces a major problem today—environmental pollution, which is caused by the release and accumulation of various harmful substances due to current industries' extreme development, rapid urbanization, and population growth. Pollutants are very diverse, ranging from chemical to physical, biological, and radiological compounds, and are widely spread in the air, soil, and waters, affecting all living systems, especially human health and life [1]. The safety and security of the environment is a major concern worldwide; therefore, prudent monitoring and management of it constitute two of the global and European priorities [2]. Researchers are interested in finding durable solutions to environmental monitoring, as the control of toxic substances is a fundamental condition for pollution remediation. Usually, the classical chromatographic [3–5] and spectroscopic [6–9] methods are used to detect contaminants, which are generally characterized by high sensibility and selectiveness. However, these methods are laborious, need several sample preparation steps, use toxic chemicals, and are time-consuming; and the equipment needs well-qualified operators.

The necessity of using some rapid, selective, sensitive, accurate, and real-time devices for detecting and screening pollutants led to the development of advanced biosensing devices. These must combine the analytical techniques with biotechnology in careful and reliable ways, at a low cost [10–12]. A special use of biosensors is in the evaluation of ecological risks. Biosensors are in such cases essential in complementing the specific chemical analyses [13,14]. For the construction of the biosensors should be considered the complexity of the environmental samples, as their use for technological applications is highly demanded [15–17].

Environmental pollutants can be monitored using specific biosensors. The detection principle must be based on a suitable physical/chemical transducer integrated with a compatible biological or biomimetic element that reversibly binds the analyte. The detector identifies and converts the resulting reactions into qualitative and quantitative sensing signals for the targeted pollutants from the sample [11,16].

The pollutants released from industrial, agricultural, and other intense human activities [11] are organic and inorganic. Biosensors' usage is essential for monitoring actual conditions of soil, water, and air samples to detect pollutants such as pesticides, potentially toxic elements, pathogens, toxins, and endocrine-disrupting chemical compounds [2]. The major and long-lasting environmentally relevant toxicants can be separated into four categories: *organochlorine pesticides* (aldrin, chlordane, DDT (dichlorodiphenyltrichloroethane), dieldrin, endrin, heptachlor, mirex, and toxaphene); *fungicides* (i.e., hexachlorobenzene); *industrial chemicals* (PCBs—polychlorinated biphenyls and their by-products), and *heavy metals*. The possibility of their quantification by using specific biosensors constitutes a significant advantage in controlling them [11]. Even though biosensors have proved their abilities to measure air pollutants in various sample types, their efficiency is often poor [10].

The capacity of these small devices to offer reliable analytical results productively and profitably should be highlighted [18]. Another characteristic that needs to be underlined is the possibility offered by to perform ongoing in-field monitoring of various pollutants [19].

Biosensors are analytical devices that each incorporate a biological sensing element to detect a targeted analyte from complex samples [20]. Biosensors convert a biological signal into a detectable electrical, optical, or thermal signal. They provide high sensitivity even with miniscule analyte concentrations [1,21,22]. A schematic diagram of the typical components of a biosensor is presented in Figure 1.

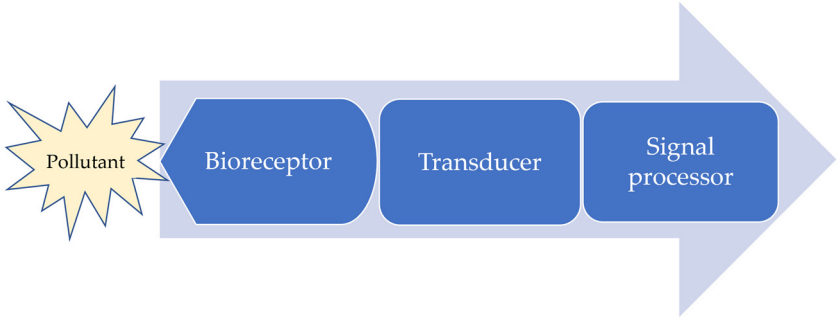


**Figure 1.** Operation of a biosensor.

A biodetection device consists of some distinct components: a bioreceptor, a transducer, a system for signal processing, and a display [16,21]. The entire unit produces a measurable detection signal relating the analyte's concentration in the target [23]. The biochemical receptor is used to recognize biological or chemical elements from the analyzed sample, being intimately associated with the transducing element, which converts the biochemical outcome into quantized electrical, optical, or thermal signal [21,22,24]. The biorecognition element might be a biological material, such as enzymes or a multienzyme system, microbes, recombinant microorganisms, functional nucleic acids, antibodies, antigens, aptamers, or an animal or plant tissue [21,24]. New alternatives use biomimetic materials (biomimetic catalysts, molecularly imprinted polymers, combinatorial ligands, etc.) [25]. Even if the biosensor is a complete, independent unit, the term specifically refers to the component that provides precise, complex bioanalytical measurements in simple formats and in real-time [10,20,24]. Biosensors must allow reuse and not be affected by pH and temperature [26].

Biosensors are classified by the most important components involved in the detection process: the bioreceptor and the transducer. Regarding the bioreceptor type, biosensors can be grouped as follows: the biocatalytic group (enzymatic biosensors), the bioaffinity group (immunosensors, aptasensors, genosensors), and the microbial group (microbial

biosensors) [2,26]. Based on the transducer's physicochemical features and its working principle, biosensors are categorized as: electrochemical (potentiometric, amperometric, impedimetric, conductometric biosensors), optical (fiber-optic, surface plasmon resonance, Raman spectroscopy-based, and FTIR-based biosensors), and mass-based (magnetoelectric and piezoelectric biosensors) (Figure 2) [16].

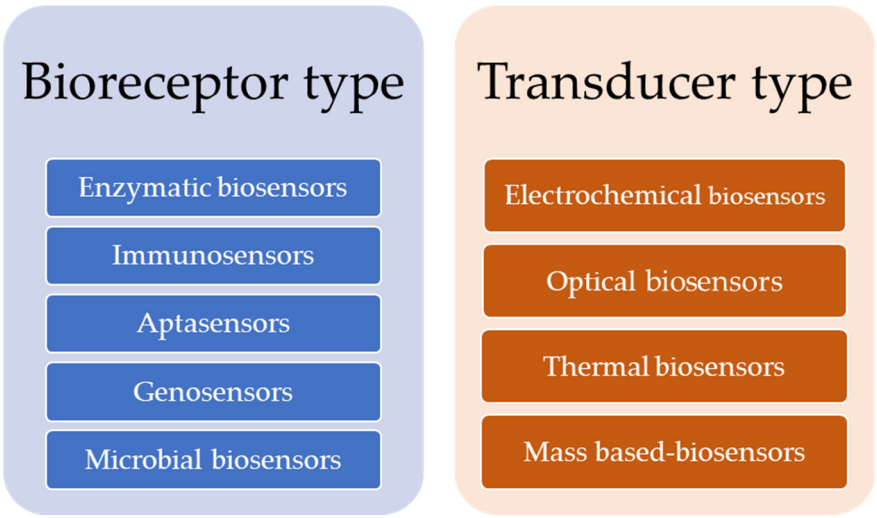


**Figure 2.** Classification of biosensors.

Biosensors present some advantages in analytical chemistry. They expedite the processes of the traditional laboratory and analytical monitoring procedures—that is, taking various analytes from diverse samples. They are small and simple devices with high sensitivity and bioselectivity for targeted analytes, precision, rapidity, and continuity in monitoring. Several factors for users must also be considered when designing them, such as easy manipulation and operation, safety functioning, suitability for in situ detection (no complex sample preparation), real-time detection, cost efficiency, and eco-friendliness [27,28].

Biosensors have seen rapid and varied development in the past few decades [10] due to their ability to identify a wide range of analytes, such as pollutants, bacteria, fungi, drugs, and food additives [16]. Such attributes demonstrate their great applicability in various fields—pharmaceutics, medicine, industry, environmental monitoring, agriculture, food, forensic chemistry, security and defense, robotics, etc. [24,27]. The main uses of a biosensor depend on the specific tasks of the application area. Their utility in the food industry was demonstrated in quality and safety control, by discerning natural and artificial components, monitoring fermentation processes, etc. Their applicability in industry is mainly in control processes. In drug discovery and clinical and medical sciences, their use is recommended for rapidly detecting chemicals or viruses that cause various diseases, including cancer [20,26].

Currently, there is increasing interest in developing highly accurate and efficient systems for identifying and screening environmental pollutants (Figure 3) [29].

Compared to other types of biosensors, e.g., biomedical ones, biosensors for environmental monitoring have a nonaged phase due to the complexity of the analysis, such as the complex ecological matrix, which interferes with pollutant recognition.

A biosensor's characteristics are directly related to its biorecognition element and its transducer's properties. Therefore, the materials used for the construction of the biosensor play an important role. Recently, laminated composites have become of great interest to various industries and applications [30–42]. The development of new composite materials is grabbing researchers' attention, as these materials are characterized by high surface-to-volume ratios, high catalytic activity, good electrical conductivity, and good magnetic properties [43–47]. Yang et al. [47] extensively presented the synthesis of carbon nanotubes

(CNT) (arc discharge, laser ablation, chemical vapor deposition (CVD), etc.) and the possibilities for their functionalization.

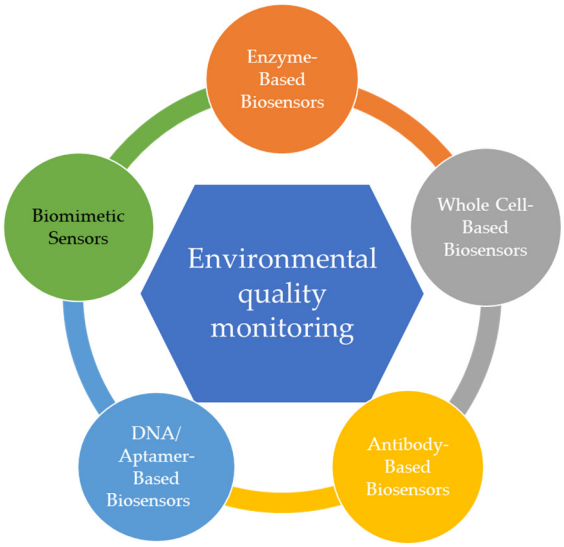


**Figure 3.** Biosensors used for the environmental quality monitoring.

Nanocomposites represent a promising technology that enhances the sensitivity and flexibility of analyses of environmental complex samples. Nanostructures such as tubes, wires, rods, and particles modify biosensors' characteristics toward achieving this goal. However, as Nigam et al. [10] noticed, there is still a real need for innovations in biosensors for environmental purposes, to assure high output of analysis for continuous, automated, and real-time results. Still, accuracy must also be considered the primary priority.

## 2. Sensors Used for Environmental Monitoring Overview

### *2.1. Enzyme-Based Biosensors*

Enzymes are macromolecules with a complex 3D structure consisting of proteins that act as biological catalysts. An enzyme-based biosensor uses a specific enzyme as a biological sensing element, combined with a transducer that converts the signal generated by the enzymatic reaction into a measurable response proportional to the analyte concentration [48]. The enzymatic reaction signal can be generated in different forms: thermal release, proton concentration changes, oxygen emission or uptake, light emission or absorption, etc. The transducer (optical, electrochemical, thermal, piezoelectric) transforms this signal into potential, current, temperature exchange, light absorption, etc.—all of these being measurable by different means [49].

*Enzymatic biosensors have* earned massive interest in the last few years due to their multiple advantages, such as the high specificity and selectivity of enzymatic reactions, their wide range of detectable analytes, flexibility in detection, and the high purity of the available enzymes [50].

Naresh et al. [51] present in their paper the operating principles of enzymatic biosensors. There are two possible categories of mechanism of action: metabolization of the target analyte by the enzyme; or the activation, inhibition, or alteration of the enzyme by the analyte.

The essential requirements of an enzymatic biosensor are the immobilization the enzymes to the transducer's surface and maintenance of their activity after immobilization [48]. The immobilized enzymes are more stable than the mobile versions and can be repetitively and continuously used [52]. The main methods for enzyme immobilization are presented in Figure 4, and in Table 1 are the characteristics of these.

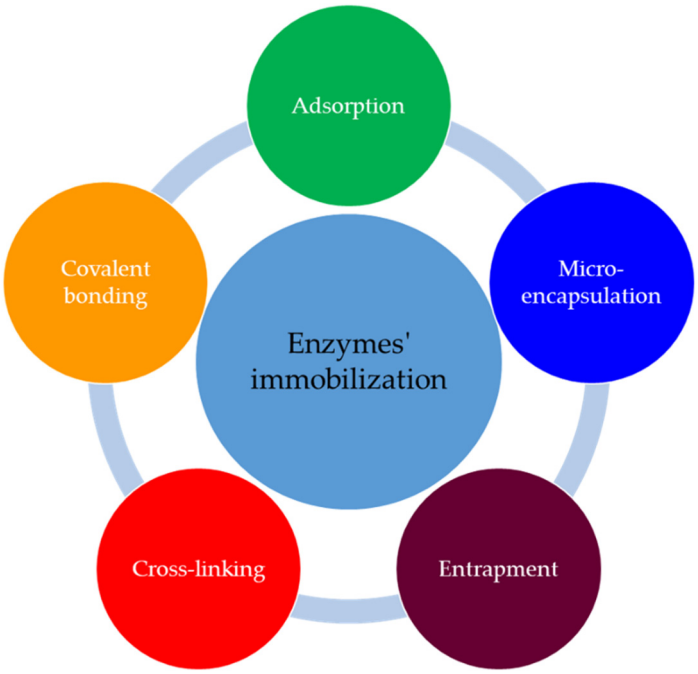


**Figure 4.** Methods for immobilization of the enzymes.

**Table 1.** Methods of enzyme immobilization for biosensors [52,53].

| Immobilization of Enzymes | Method's Characteristics |
|---|---|
| Adsorption | Simple, inexpensive, less destructive to enzymatic activity, no additional reagent necessary |
| Microencapsulation | Preservation of structural and acting integrities of enzymes, due to their protection against environmental conditions |
| Entrapment | High stability conferred to the enzymes |
| Cross-linking | Improved efficiency and stability of enzymes by strong and stable bindings |
| Covalent bondings | More stability for enzymes and enzymes-support complexes, meanwhile stronger bindings than in adsorption case |

Enzyme-based biosensors are widely used in food, medical, agricultural, and environmental fields. As shown in Table 2, the development of enzymatic biosensors for environment monitoring represents a subject of considerable interest.

**Table 2.** Examples of enzyme-based biosensors used for environmental monitoring.

| Analyte | Enzyme(s) | Immobilization Method | Transducer | Target | LOD | Linearity | Reference |
|---|---|---|---|---|---|---|---|
| $Hg^{2+}$, $Cu^{2+}$, $Cd^{2+}$ | Urease | Entrapment in sol-gel matrix | Optical | River water | 10 nM, 50 µM, 500 µM | - | [54] |
| Chromium | GOx | Cross-linking with GA and covering with aniline membrane | Amperometric | Soil | 0.49 µg $L^{-1}$ | 0.49–95.73 $mgL^{-1}$ 95.73–8.05 $mgL^{-1}$ | [55] |
| Paraoxon | AChE | Dropping on the multiwall carbon nanotubes | Amperometric | Water | 0.5 nmol $L^{-1}$ | 6.9 nM | [56] |

**Table 2.** *Cont.*

| Analyte | Enzyme(s) | Immobilization Method | Transducer | Target | LOD | Linearity | Reference |
|---|---|---|---|---|---|---|---|
| Paraoxon-ethyl, diisopropyl fluorophosphates | AChE | Cross-linking with BSA in a saturated glutaraldehyde vapor | Conductometric | Soil | $1 \times 10^{-8}$, $5 \times 10^{-11}$ | - | [57] |
| Atrazine | Tyrosinase | Cross-linking with PVA-SbQ | Amperometric | Spiked drinking water s | 0.3 ppm | 0.5–20 ppm | [58] |
| Atrazine | Tyrosinase | Entrapping in poly(L-DOPA) | Amperometric | Water | 10 ppb | 50 ppb–3.0 ppm | [59] |
| Organophosphorus neurotoxin | AChE | Cross-linking with GA | Piezoelectric | Water | 50 mg/m$^3$ | 0–50 mg/m$^3$ | [60] |
| Captan | Glutathione-S-transferase | Entrapment in gel sodium alginate | Optical | Water | 2 ppm | - | [61] |
| Anatoxin-a | AChE | Entrapment in PVA-SbQ | Amperometric | Water | 1 μg L$^{-1}$ | 0–2.0 ppm | [62] |
| Catechol | Tyrosinase | Chitosan-gold nanoparticles | Amperometric | Environmental monitoring | $27 \times 10^{-6}$ mM | 0.046–50 μM | [63] |
| Methyl salicylate | Alcohol oxidase and peroxidase | Molecular tetherings in carbon nanotube matrix | Amperometric | Environmental monitoring | 0.00098 mM | - | [64] |

Abbreviations: LOD—limit of detection; Gox—glucose oxidase; GA—glutaraldehyde; AchE—acetylcholinesterase; BSA—bovine serum albumin; PVA-SbQ—polyvinyl alcohol bearing styrylpyridinium groups; L-DOPA-l-3,4-dihydroxyphenylalanine.

### *2.2. Whole Cell-Based Biosensors (Microbial)*

Whole-cell-based biosensors use natural or genetically engineered microorganisms (bacteria, fungi, algae, protozoa, or viruses) that can interact with a broad array of analytes and produce a signal detectable and quantifiable by a specific transducer [65]. Several transducers have been integrated with microorganisms, being built on different principles: electrical (amperometric, conductometric, potentiometric), colorimetric, and optical (colorimetric, luminescent, fluorescent) [66–68]. Microbial biosensors operate under a range of working conditions and are more sensitive to environmental signals than conventional ones [15]. They present various advantages: low limits of detection, high selectivity, and high sensitivity. Based on these features, whole-cell bioreceptors are applicable in many fields [51].

Microbial sensors can be considered a developed form of enzyme-based biosensors, as their mechanisms of detection are mostly identical. Both of them require the application of an immobilization technique to fix the biological material onto transducers or support matrices. As in the enzymes case, microorganisms can be immobilized by physical (adsorption and entrapment) and chemical methods (covalent binding and cross-linking). Finally, the chosen immobilization method must ensure mechanical resistance, cell viability, safe handling, and long-term storage [69].

Besides the advantages presented over the conventional methods, namely, high sensitivity, simultaneous detection of several compounds, high potential for on-site examinations, and cost-effectiveness, microbial biosensors are also associated with some drawbacks. Their long response times, the cells' sensitivity to environmental variables (temperature, pH, etc.), and the difficulty of maintaining cell viability for an extended period are some of their limitations [15,65,70].

Numerous recent articles reported on the use of microbial biosensors to detect environmental pollutants, such as pesticides, heavy metals (As, Cu, Hg, Pb, or Cd), phenols, and other toxic compounds, using terrestrial and aquatic biota [15,19,71,72]. Other microbial biosensors were proposed and developed in the last few years as well, with remarkable

applicability to environmental monitoring. Table 3 summarizes the results of several such investigations reported in the literature.

**Table 3.** Examples of microbial biosensors used for environmental monitoring.

| Analyte | Microorganism | Immobilization Method | Transducer | Target | LOD | Reference |
|---|---|---|---|---|---|---|
| $As^{3+}$ | Genetically engineered *S. oneidensis* | Biofilm formation | Electrochemical | Environmental monitoring | 40 µM | [73] |
| $Cu^{2+}$, $Cd^{2+}$, $Ni^{2+}$, $Pb^{2+}$ | *Saccharomyces cerevisiae* S288C | Physical adsorption on BND-chitosan hydrogell polymer on GCE | Amperometric | Wastewater | - | [74] |
| $As^{3+}$, $Cd^{2+}$, $Pb^{2+}$, $Zn^{2+}$ | *E. coli* | Microbial culture in microfluidic device | Fluorescent | Water | - | [75] |
| $Pb^{2+}$ | *E. Coli* DH5α | Microbial culture in a microfluidic device | Fluorescent | Environmental monitoring | | [76] |
| $Cd^{2+}$, $Cu^{2+}$, $Zn^{2+}$ | *Bacillus megaterium* VR1 | Entrapment in sol-gel matrix | Fluorescent | Soil | $1.42 \times 10^{-4}$, $3.16 \times 10^{-4}$, $2.42 \times 10^{-4}$ | [14] |
| $Cu^{2+}$ | *S. Cerevisiae* | Entrapment in alginate beads | Colorimetric | Water | 1 µM | [77] |
| Paraoxon, parathion, methyl-parathion | Genetically engineered *Escherihia coli* | Biofilm on GCE modified with OMCs | Amperometric | Environmmental monitoring | 9 nM, 10 nM, 15 nMz | [78] |
| Atrazine (herbicide) | *Anabaena variabilis* | Entrapment in alginate | Amperometric | Environmmental monitoring | 0.07 µM | [79] |
| Diuron (herbicide) | *Chlamydomonas reinhardtii* | $Ti/TiO_2$ ultramicroelectrodes in algal suspension | Chronoamperometric | Water | 0.2 µM | [80] |
| Simazine (herbicide) | *Dictyosphaerium chlorelloides* Dc1M | Adsorption on porous silicone disks | Luminescent | Drinking water | 40.8 µg $L^{-1}$ | [81] |

LOD—limit of detection; BND—boron-doped nanocrystalline diamond; GCE—glassy carbon electrode; OMCs—ordered mesopore carbons.

### *2.3. Antibody-Based Biosensors*

Antibodies or immunoglobulins are a large class of glycoproteins produced by specialized cells as part of the immune system to detect harmful substances (antigens), such as microorganisms and chemicals. The antibodies can recognize and bind antigens, leading to stable antibody–antigen complexes [82–84]. Depending on how they are harvested, antibodies can be monoclonal or polyclonal. Monoclonal antibodies are laboratory-produced by hybridoma selection, whereas polyclonal antibodies are complex mixtures of antibodies isolated after animal immunization [85].

Antibody-based biosensors, also called immunosensors, are compact devices that detect and quantify, using a transducer, the specific interaction between immunoglobulins and antigens. Depending on the transducing mechanism, immunosensors are classified as electrochemical (amperometric, potentiometric, and impedimetric), colorimetric, optical, and microgravimetric. They can also be classified as labelled or nonlabelled sensors [17,86–88]. The labelling consists of attaching a sensitively detectable marker to the targeted analyte or the bioreceptor. During the analysis, the tag's activity is measured. These tags may can be various sorts of compounds, including enzymes, fluorescent dyes, electroactive compounds, and nanoparticles [89]. Nonlabelled immunosensors are designed so that the

antigen–antibody complex can be directly determined by estimating the physical changes produced by its development [51].

Immunosensors possess the advantages of better selectivity and sensitivity than classical analytical methods. At the same time, the evolution of immunoreactions on the detector's surface can be observed in real-time [83,90]. However, the limitations in using antibody-based biosensors must also be considered, such as pH and temperature sensitivity, considerable time consumption, and the need for developing specialized reagents for each compound [91].

Several applications of the antibody-based biosensors within environmental monitoring are summarized in Table 4.

**Table 4.** Examples of immunosensors used for environmental monitoring.

| Analyte | Transducer | Electrode/Sensing Material | Target | LOD | Linearity | Reference |
|---|---|---|---|---|---|---|
| Chlorpyrifos | Impedimetric | Chip modified with gold nanoparticles | - | 0.5 ng $mL^{-1}$ | 0.5–500 ng/ml | [92] |
| TBBPA-DHEE and TBBPA-MHEE | Impedimetric | Silica nanoparticles | Aquatic environments | 0.08 ng $mL^{-1}$ | 0.21–111.31 ng/mL | [93] |
| Atrazine | Electrochemical | SWCNT | Seawater, riverine water | 0.01 ng $mL^{-1}$ | - | [94] |
| Microcystin-LR | Impedimetric | Gold electrodes with MoS2 andgold nanorods | Water | 5 ng $L^{-1}$ | 0.01–20 $gL^{-1}$ | [95] |
| Okadaic acid Domoic acid | Optical (SPR) | Gold electrode with carboxymethylated surface | Seawater | 0.36 ng $mL^{-1}$ 1.66 ng $mL^{-1}$ | - | [96] |
| Okadaic acid | Impedimetric | Graphene | Seawater | 0.05 ng $mL^{-1}$ | - | [97] |
| *Legionella pneumophila* | Optical (SPR) | Gold substrate | Water | 103 CFU $mL^{-1}$ | - | [98] |

Abbreviations: TBBPA-DHEE—tetrabromobisphenol A bis(2-hydroxyethyl) ether; TBBPA-MHEE—tetrabromobisphenol A mono(hydroxyethyl) ether; SWCNT—single-walled carbon nanotubes; SPR—surface plasmon resonance; microcystin—LR-microcystin-leucinearginine.

### *2.4. DNA/Aptamer-Based Biosensors*

#### 2.4.1. Aptamer-Based Biosensors

Aptamers or "chemical antibodies" [99] are artificial, single-stranded oligonucleotide (DNA (deoxyribonucleic acid) or RNA (ribonucleic acid) sequences (15–80 base pairs in length) that can bind to specific target molecules [100]. The range of aptamer targets is extensive, from small molecules (peptides, proteins, carbohydrates, metal ions) to cells, viruses, and bacteria [101–103].

Aptamers can be selected in vitro through a process called SELEX (systematic evolution of ligands by exponential enrichment) [104–106]. The SELEX procedure starts with preparing an extensive library of oligonucleotides with different sequences, with which the target molecules are incubated for some time. After incubation, unbounded molecules are separated, and the target-bound oligonucleotides are eluted by heating or washing. The bound aptamer molecules are amplified by the polymerase chain reaction (PCR) to create the input for the following selection rounds. The entire process uses 5–15 cycles of selection and amplification [107–109].

In comparison with antibodies, aptamers have some specific advantages, such as higher stability in various environmental conditions (temperature, pH), lower cost, the ability to regenerate, and the possibility of being chemically synthesized or modified in accordance with target molecules [89,102,108].

In the last few years, several biosensors (colorimetric, fluorescent, electrochemical, and SERS—surface enhanced Raman spectroscopy) have been designed to detect environmental pollutants, using aptamers as the bioreceptors. Furthermore, the synthesis of new nanomaterials showed their significant potential for the development of innovative aptasensors. The latter are sustained by their strong biocompatibility with aptamers [102,106].

Table 5 summarizes recent studies on aptasensors developed for the detection of pollutants.

**Table 5.** Examples of aptamer-based biosensors used for environmental monitoring.

| Analyte | Detection Method | Target | LOD | Linearity | Reference |
|---|---|---|---|---|---|
| $Ag^{+}$ | SERS based on Au@Ag core–shell nanoparticles | Tap water, river water | $50 \times 10^{-12}$ mg $L^{-1}$ | 0.1–100 nM | [110] |
| $As^{3+}$ | Colorimetric with GNPs | Wastewater | 0.0006 mg $L^{-1}$ | 1–400 range/ppm | [111] |
| $As^{3+}$ | Colorimetric with AuNPs | Soil | 1.97 ppm | - | [112] |
| $Cd^{2+}$ | Fluorescence with use of SYBR green I as signal reporter | Tap water, river water | $3 \times 10^{-9}$ mg $L^{-1}$ | 1.12–224.82 μg $L^{-1}$ | [113] |
| $Hg^{2+}$ | SERS based on dual recycling | Water environment | 0.11 fM | 0.2–125 fM | [114] |
| $Hg^{2+}$ | SERS based on $SiO_2$@Au core/shell nanoparticles | Lake water | $10 \times 10^{-9}$ mg $L^{-1}$ | - | [115] |
| $Pb^{2+}$ | Electrochemical (Impedance), G-rich aptamer/MWCNTs/GNPs | Water | $4.3 \times 10^{-15}$ M | $5.0 \times 10^{-11}$–$1.0 \times 10^{-14}$ M | [116] |
| $Pb^{2+}$ | Fluorescence based on gold nanoflowers | Tap water | 0.285 nM | 0.01–850 nM | [117] |
| $Pb^{2+}$ | Colorimetric with use of silver staining | Soil | $5.0 \times 10^{-7}$ mg $L^{-1}$ | - | [118] |
| Acetampirid | Chemiluminescence with use of AuNPs | Wastewater Soil | $62 \times 10^{-12}$ mg $L^{-1}$<br>$1.0 \times 10^{-9}$ mg $L^{-1}$ | - | [119] |
| Malathion | Colorimetric based on AuNPs and cationic polymer | Lake water | $6 \times 10^{-14}$ mg $L^{-1}$ | 0.5–1000 pM | [120] |
| Omethoate | Fluorescence based on S-GQD | - | 1 ppb | 0–200 ppm | [121] |
| Organophosphorus pesticides | Fluorescence with poly(T) CuNPs | Lake water | 0.22 nM | 0–200 nM | [122] |
| Tetracycline | Photoelectrochemical based on CdTe-BiOBr heterojunction | Soil | 9.25 pM | 10–1500 pM | [123] |

Abbreviations: GNPs—gold nanoparticules; G—guanine; SERS—surface-enhanced Raman scattering; CuNPs—copper nanoparticles; S-GQD—sulphur-doped graphene quantum dot, SYBR—N′,N′-dimethyl-N-[4-[(E)-(3-methyl-1,3-benzothiazol-2-ylidene)methyl]-1-phenylquinolin-1-ium-2-yl]-N-propylpropane-1,3-diamine; G-rich—guanine-rich; MWCNTs— carboxylic acid group functionalized multiwalled carbon nanotubes (MWNTs-COOH).

#### 2.4.2. DNA-Based Biosensors

DNA-based biosensors use nucleic acids (single-stranded DNA, ss-DNA) as recognition elements. Their working principle is based on two mechanisms: (i) the hybridization process between the target DNA and its complementary strand immobilized on a sensing area through the spontaneous hydrogen bonding between adenine–thymine and cytosine–guanine pairs [49,124]; (ii) the alteration of the ss-DNA structure by the target analyte's molecules [125]. These mechanisms induce various physicochemical changes that lead to

the generation of a specific signal that can be converted into a measurable response by an appropriate transducer, usually optical or electrochemical [126].

A significant stage in the design of DNA-based biosensors is the immobilization procedure of the nucleic acid fragments on the electrode surface. Regardless of the method used (adsorption, covalent bonding, or avidin–biotin interaction), the immobilization must preserve the activity of these fragments—that is, ensure their stability and accessibility to the target molecules [127].

Due to their multiple advantages, such as specificity, sensitivity, biocompatibility, and cost-effectivity, DNA-based biosensors are used in several fields, including disease prognosis, clinical diagnosis, food control, and environmental screening [126,128].

Several studies have illustrated the ability of DNA-based biosensors to detect traces of heavy metals in the environment [125,128–130]. In this case, the working principle is based on the affinity of some heavy metal ions toward forming stable duplex structures together with certain DNA bases. Mercury ion ($Hg^{2+}$) selectively binds thymine (T) bases and creates a thermal stable T-$Hg^{2+}$–T duplex [131]. Similarly, silver ions ($Ag^{+}$) selectively interact with two cytosine (C) bases and form C–$Ag^{+}$–C base pairs, which stabilize the DNA duplex [49,125]. Therefore, in the presence of some metal ions, thymine-rich or cytosine-rich single-stranded DNA can form stable structures by which metals can be detected with adequate transducers [125].

Some of the recent DNA-based biosensors' applications are presented in Table 6.

**Table 6.** Examples of DNA-based biosensors used for environmental monitoring.

| Analyte | Transducer | Target | LOD | Linearity | Reference |
|---|---|---|---|---|---|
| $Hg^{2+}$ | Electrochemical | Tap water, river water | 0.05 nM | 0.1–200 nM | [132] |
| $Pb^{2+}$ | Fluorescent | Aqueous systems | 5 nM | 0–50 nM | [133] |
| $Pb^{2+}$ | Fluorescent | Lake water | 0.6 nM | 2–10 nM | [134] |
| Organophosphorus pesticides | Fluorescent | Lake water | 0.018 μg $L^{-1}$ | 2–10 μg/L | [134] |
| Cyanazine | Impedimetric | Water | 0.8 nM | 4.0 nM–70 μM | [135] |
| Pirazon | Impedimetric | Water | $1 \times 10^{-10}$ M | $5 \times 10^{-9}$–$5 \times 10^{-5}$ M | [136] |
| *Legionella pneumophila* | Optical (SPRi) | Water | 104 CFU $mL^{-1}$ | - | [137] |
| *Vibrio cholerae* | Impedimetric | - | $7.41 \times 10^{-30}$ mol $L^{-1}$ | $10^{-8}$–$10^{-14}$ mol $L^{-1}$ | [138] |
| *Escherichia coli* | Amperometric | Soil | 100 cells/g soil | - | [139] |
| *Bacillus thuringiensis* | Impedimetric | - | $0.997 \times 10^{-12}$ M | 1 pM–1 μM | [140] |
| *Ostreopsis* cf. *ovata* | Colorimetric | Plankton, bentonite | 9 pg/μL | - | [141] |

Abbreviations: SPR—surface plasmon resonance imaging; CFU—colony-forming units.

## 3. Biomimetic Sensors

Although the terminology may seem new, the basis of biomimetics was laid years ago. Its principle is finding solutions that mimic a natural system's mechanisms, especially regarding the structure of an organism or its specific interactions with the environment. The created products can be performant and adequately adapted to real environments [142].

Biomimetic sensors were first constructed while considering the basic principles of the related enzymatic biosensors. The intention was to maintain high sensibility, selectivity, sensitivity, and easy operation, while simultaneously decreasing some of the disadvantages. The limitations that need to be overcome mainly relate to each enzyme's specific features, such as inactivation issues, or high costs because of the purification and standardization pro-

cesses. In such contexts, the research was conducted toward finding sustainable solutions for creating imitative systems. Some of the developed models are based on metal complexes, molecularly imprinted polymers, nanozymes, synzymes, and nanochannels [143].

In the last few years, the domain of biomimetic sensors has registered significant progress. Initially, biomimetic sensors were constructed using uni- or bi-dimensional structures (Figure 5). Then tridimensional assemblies were widely used, and the results indicated improved performances, sometimes exceeding the natural models' performances [143]. Finding the proper ligand for the targeted analyte is the first step in designing precise tools. The peptide selection used in the recognition systems is important for the sensor's affinity [144]. Computer modelling [145] and simulation are two stages that improve the performances of these devices.

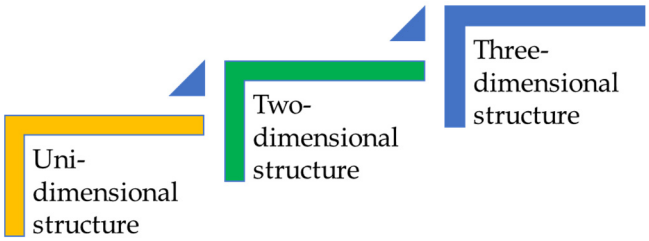


**Figure 5.** Structures used for the construction of biomimetic sensors.

The domain of biomimetic sensors used for environmental pollutants detection is currently developing. Research has opened multiple promising directions for the construction of such sensors: modified nanoparticles [146–148], metal chalcogenides nanocrystals built on various microorganisms [149], valorization of classical imprinted electrodes [150], and nanozymes for phenol removal [151].

Some examples of sensors created based on mimetic principles with applications in environmental monitoring are summarized in Table 7.

**Table 7.** Examples of biomimetic sensors used for environmental monitoring.

| Analyte | Mimetic Structure | Transducer | Target | Sensibility (LOD) | Linearity | Reference |
|---|---|---|---|---|---|---|
| | | | Heavy metals | | | |
| $Cu^{2+}$, $Cr^{3+}$, $Fe^{3+}$, $Pb^{2+}$, $Fe^{2+}$, $Cd^{2+}$, $Cr^{6+}$, $Co^{2+}$, $Zn^{2+}$, $Ag^{+}$, $Al^{3+}$ | Enzyme immobilization Metal phosphates-acetylcholinesterase nanoflowers | Colorimetric | Water | $Cu^{2+}$—0.81 µM, $Cr^{3+}$—0.75 µM $Al^{3+}$—1.06 µM | 2.5–500 µM. | [152] |
| $Pb^{2+}$ | Gold nanoparticles with glutathione linker | UV–vis spectroscopic | Water | 47.6 nM (9.9 ppb) | 2–14 mM | [153] |
| $Hg^{2+}$ | Cysteine-decorated ferromagnetic particle (Cys-$Fe_3O_4$) | Colorimetric | River water | 5.9 pM. | 0.02–90 nM | [154] |
| | | | Chemicals | | | |
| Methyl green | Magnetic molecularly imprinted polymer | Square-wave adsorptive anodic stripping voltammetry | River waterIndustrial wastewater | $1.0 \times 10^{-8}$ mol $L^{-1}$ | $9.9 \times 10^{-8}$–$1.8 \times 10^{-6}$ mol $L^{-1}$ | [145] |
| Acetylcholinesterase inhibitors | Microchannel 1-phenyl-1,2,3-butanetrione 2-oxime (PBO)-based microsensor | Potentiometric | Surface waters used for municipal drinking water supplies | LD50, LC50 | 2–1360 mg $kg^{-1}$ | [155] |
| Acetone gas | Zeolitic imidazolate framework-90 polyhedron crystals | quartz crystal microbalance | Air | Lower than 20 ppb | - | [156] |

**Table 7.** *Cont.*

| Analyte | Mimetic Structure | Transducer | Target | Sensibility (LOD) | Linearity | Reference |
|---|---|---|---|---|---|---|
| Nitrite ions | Oxo-bridged dinuclear manganese-phenanthroline complex immobilized into an ion-exchange Polymeric film deposited on glassy carbon electrode | Cyclic voltammetry | Environmental samples | $6.50 \times 10^{-6}$ mol $L^{-1}$ | $2.49 \times 10^{-6}$–$9.90 \times 10^{-6}$ mol $L^{-1}$ | [157] |
| Catechol | Metal-organic frameworks | | Water | 33 nmol $L^{-1}$ | - | [158] |
| Urea | Embedding urease and bovine hemoglobin in metal-organic frameworks through biomimetic mineralization | Colorimetric | Sewage | 0.02 mM | 0.08–20.00 mM | [159] |
| | | | Pesticides | | | |
| Diurone | Carbon paste electrode modified with the nickel(II) 1,4,8,11,15,18,22,25-octabutoxy-29*H*,31*H*-phthalocyanine complex | Cyclic voltammetry and amperometry | River water, soil | $6.14 \times 10^{-6}$ mol $L^{-1}$, | $9.9 \times 10^{-6}$ and $1.5 \times 10^{-4}$ mol $L^{-1}$ | [160] |
| Organophosphorus pesticides | Employing a functionalized polyacrylamide, polyhydroxamicalkanoate | Amperometric | Water supply | 0.26 µmol $L^{-1}$ | - | [161] |
| Carbamate | Gold nanoclusters-anchored $MnO_2$ (AuNCs-$MnO_2$) nanocomposite | Fluorimetric/Colorimetric | Soil, water | 0.125 µg $L^{-1}$. | - | [162] |
| Paraoxon | $Cu_3(PO_4)_2 \cdot 3H_2O$, AChE and ChO -based lab-on paper platform | Cyclic voltammetry and Colorimetric | Tap and river water | 6 fg $mL^{-1}$ | - | [163] |
| | | | Toxins | | | |
| Bacterial toxins | Microcystins inserted into a polymeric matrix | Potentiometric | Water | below the guideline value establishedby WHO | $7.24 \times 10^{-10}$–$1.28 \times 10^{-9}$ M | [150] |

Abbreviations: LOD—limit of detection; LD50—lethal dose (50%); LC50—lethal concentration (50%); WHO—World Health Organization; Cys—cysteine.

## 4. Future Perspectives

Another approach of biosensors regards the possibility of simultaneous detection of multiple pollutants. Several investigations have been successfully conducted to that end. Raymundo-Pereira et al. [164] evidenced the possibility of using carbon screen-printed electrodes for parallel identification of estradiol, paracetamol, and hydroquinone in tap water. Their findings could have an important application in wastewater analysis. Good prospects for use in water quality analysis were also provided by a luminescent sensor derived from a stable europium(III) metal–organic framework. It was tested for antibiotic identification [165]. The interest in using biosensors for water contaminant detection was also fostered by Martins et al. [166]. They identified sulfamethoxazole and trimethoprim from water samples.

The first steps toward making a biosensor with two detection mechanisms were made by Belaidi et al. [167]. Their electrochemical and optical detection biosensor, based on different algae responses, showed promising perspectives for simultaneous pesticide identification in water samples. These findings also provoked the design of a mimetic biosensor capable of detecting multiple pollutants.

The biosensors constructed for environmental quality monitoring will continue to be improved by using novel nanocomposites and nanomaterials, and new functionalization

methods, but the necessity for in situ and real-time monitoring of pollutants will lead to the development of new sensing systems and even their coupling with aircraft systems [168].

With the current need for cheap, sensitive, fast, and reliable devices for environmental monitoring, the main challenge remains the gap between the results of academic research and the implementation of these biosensors as marketable products.

## 5. Conclusions

This review aimed to show that the need for fast, reliable, and stable devices for the detection of environmental pollutants can be satisfied by biosensors. However, these should answer the demands of sensitivity and selectivity when used in complex and unpredictable environmental samples with changeable compositions.

Independent of the sensing element or transducer, when developing biosensors for environmental pollutants detection, it is important to consider the possibility of continuous use, which would require fast renewal of the biological activity during the detection cycles; portability; cost; and last but not least, the possibility of automatization and integration into professional devices. In most investigations, the performance of the biosensor is assessed based on standardized laboratory samples.

The biological sensing elements—enzymes, aptamers, DNA, antibodies, and microorganisms—might face challenges in terms of stability, possible interference, and optimal working conditions, but these still have the advantage of being open to improvements in terms of specificity and selectivity.

As a result of scientific research in recent years, biomimetic sensors are characterized by better kinetic performances than enzyme-based biosensors. Still, specificity and selectivity remain their main shortcomings.

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