

## Recent advances in biosensors for micro- and nanoplastics detection

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### Abstract

Microplastics (MPs) pollution has become a major environmental issue due to their widespread presence and potential harmful effects on both aquatic and terrestrial ecosystems. Reliable and efficient detection methods are crucial for identifying MPs in environmental samples, which helps in understanding their sources, distribution, and impacts. This review covers various biosensors (recognition elements: aptamer, antibody, whole cell, and estrogen) used to detect MPs. It discusses the strengths and limitations of each biosensor, as well as recent innovations. By comparing different biosensors and their applications across various sample types, this review aims to provide a foundation to guide future research and monitoring efforts.

**Keywords:** Biosensors; Nano- and microplastic; Aptamer; Antibody; Bacteria

### 1. Introduction

The increasing population contributes to heightened environmental pollution, posing a significant challenge for society. Humans cause various types of contamination, including smoke, fertilisers, and synthetic substances. Plastics are extensively used in daily life today due to their superior qualities—such as durability, lightness, stability, and insulation—and their affordability. It is well known that we produce large quantities of plastic waste every day, which endangers the environment in many ways. Global plastic production was estimated to reach 390.7 million metric tonnes in 2021, with an annual growth rate of about four percent [1]. Microplastics (MPs) and nanoplastics (NPs) are tiny particles derived from synthetic polymers like nylon, polyethylene, polyester, Teflon, and epoxy. Plastics are classified by size into MPs (>25 mm), mesoplastics (5–25 mm), smaller MPs (0.1–5 mm), and NPs (1 nm–1000 nm) [2]. The most common MPs in the environment are made from polymers, including high-density polyethylene (HDPE), low-density polyethylene (LDPE), polystyrene (PS), polypropylene (PP), polyethylene terephthalate (PET), polyvinyl chloride (PVC), and polyamide (PA; also called nylon) [3]. Besides these traditional polymers, other MPs originate from biodegradable (natural) bioplastics derived from renewable biomass sources such as corn starch, sugarcane, and microbes. These include poly(lactic acid) (PLA), polyhydroxyalkanoates (PHA), poly(3-hydroxybutyrate) (PHB), poly(3-hydroxyvalerate) (PHV), and poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV), which are used on an industrial scale. MPs created from styrene-butadiene rubber (SBR) account for 40–60% of synthetic polymers and are known as tyre wear particles (TWP) [4]. Synthetic polymers, including polyester (PES), alkyds, vinyl and acryl (co)polymers, epoxy resin, and urethane resins, are also used in paints and surface coatings. These polymers act as binders and film formers, significantly enhancing important paint characteristics such as drying time, gloss, durability, toughness, resistance to abrasion, flexibility, and adhesion [5]. Although their small size makes them difficult to detect, their contribution to pollution is substantial. Microplastics mainly originate from sources such as microbeads in shampoos, body scrubs, and toothpaste, as well as glitter and synthetic materials like polyester, nylon, and acrylic. Additionally, microplastics are generated from wear on car tyres, brake pads, and larger plastic waste. After use, approximately 79% of manufactured plastics end up in the environment, either through deliberate or accidental disposal [6]. Consequently, plastics break down into smaller fragments. Microplastics have been found in every part of the world, indicating their widespread presence, including within living organisms.

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The pollution of water bodies such as oceans, rivers, and lakes with microplastics poses serious consequences. Studies have shown that microplastics are present on the ocean floor and inside living organisms, including fish and zooplankton [7-9]. Marine animals, such as fish, seabirds, and marine mammals, face significant risks as they can mistake microplastics for food or accidentally ingest them while feeding. This can result in severe consequences like internal injuries, malnutrition, and even death, disrupting the fragile balance of marine ecosystems. The potential threats to human health come from consuming marine-derived food products or relying on these water systems [10]. Other ways humans may be exposed to microplastics include ingesting packaged foods, inhaling airborne particles, and skin contact [11]. While research in this field continues, genuine concerns remain about the entry of microplastics into the human body through contaminated food and water. Microplastics are not limited to marine life; they have also been found in human placentas, leading to the term 'cyborg babies,' describing infants composed partly of biological and inorganic materials rather than solely human cells [12]. Furthermore, microplastics have been detected in newborns, potentially affecting fetal immune development [13].

The use of plastic mulch creates a source and sink of microplastics (MPs), which negatively impact soil organisms, including those within the soil profile and rhizospheric microorganisms [14,15]. In terrestrial ecosystems, MPs influence the entire system, affecting communities, populations, individual organisms, and their enzyme activity [16]. Furthermore, MPs hinder seed germination and restrict the growth of both root and shoot structures in plants [14]. Microplastics can either adhere to root surfaces or be absorbed by roots, fruits, and vegetables, leading to their accumulation within these plant parts [17]. The presence of MPs, especially those associated with heavy metals, has a significantly harmful effect on crop yields and their production traits [18]. MPs have become a major environmental concern, causing severe damage to ecosystems and posing potential risks to wildlife and human health [2]. Due to their slow breakdown process, MPs are regarded as hazardous environmental pollutants [19, 20].

The scientific evidence regarding the impact of MPs is both compelling and concerning. Their widespread presence in the environment demands immediate efforts to detect them in ecological samples and a collective initiative to reduce their impact. Identifying MPs is crucial for understanding their environmental and health implications, informing regulatory policies, advancing scientific research, and raising public awareness. Currently, techniques such as microscopy, thermal analysis, and spectroscopy are primarily used to identify microplastics. However, most detection methods require some form of sample pretreatment; especially for submicron and nanometer-sized microplastics, many techniques face limitations that hinder the detection of the tiniest traces [21-23]. Although more advanced technologies are available, their high costs, lengthy processing times, and low recovery rates do not support the need for real-time, in situ detection of microplastics in aquatic environments.

Biosensors are devices designed to analyze and measure biological materials. The primary objective of a biosensor is to provide rapid, precise, real-time, and reliable information about the analyte being tested. Moreover, biosensors can be highly selective for a particular analyte, enabling accurate detection that is generally unaffected by other substances in the sample [24-28]. They have become promising alternatives by offering specificity, lower detection limits, and potential for real-time monitoring in complex matrices. These systems incorporate biorecognition elements, such as enzymes, antibodies, or cells, in combination with a transducer that converts biochemical interactions into measurable signals (optical, electrochemical, or thermal).

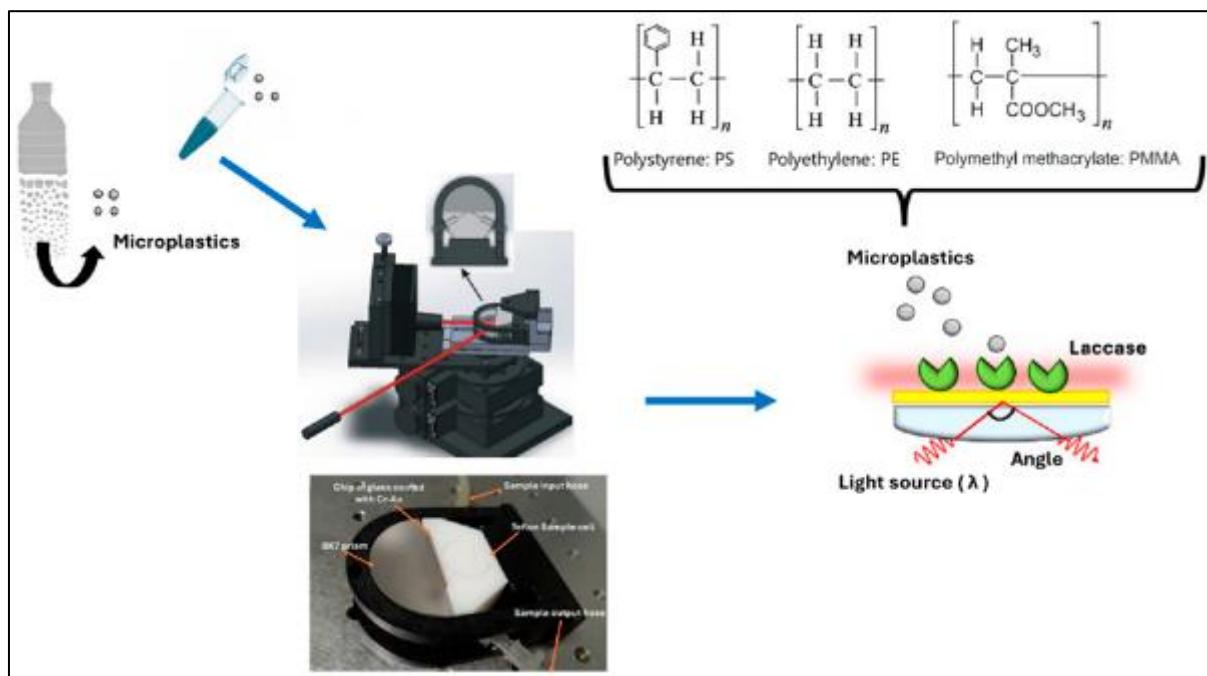
This review article examines the recent advancements in the application of biosensors for detecting micro- and nanoplastics. These biosensors are classified and examined according to the recognition element used (aptamer, antibody, enzyme, or cell). Furthermore, a comparison of the detection limits of the different biosensors is provided. Lastly, the challenges associated with biosensors for micro- and nanoplastics are addressed, and potential directions for future research on the use of these sensors for microplastics detection in marine environments are suggested, highlighting the advantages of their portable, cost-effective, and real-time monitoring capabilities.

## 2. Classification of biosensors for micro- and nanoplastics detection

### 2.1. Enzyme-Based Biosensors

Enzymes capable of breaking down or binding to polymers serve as promising recognition elements. Among bioreceptors, laccase enzymes are notable for their ability to identify and decompose plastics. A study presented a new enzymatic biosensor built on the Surface Plasmon Resonance (SPR) platform for sensitive detection of MPs and NPs, using laccase as the recognition component [29]. Standard plastic particles, including polystyrene (PS, 0.1  $\mu\text{m}$ ), polymethyl methacrylate (PMMA, 1.0  $\mu\text{m}$  and 100  $\mu\text{m}$ ), and polyethylene (PE, 34–50  $\mu\text{m}$ ), were examined using SPR angular interrogation and a fixed-angle approach. The angular method demonstrated a clear correlation between the resonance angle, particle size, and refractive index. In contrast, the fixed-angle technique, combined with immobilized

laccase, facilitated specific detection through enzyme-substrate interactions. Detection limits ranged from  $7.5 \times 10^{-4}$   $\mu\text{g/mL}$  (PE, 34–50  $\mu\text{m}$ ) to 253.2  $\mu\text{g/mL}$  (PMMA, 1  $\mu\text{m}$ ), with notable differences depending on polymer type and enzymatic affinity. Testing the biosensor on real rainwater samples from Tula and Molango in Mexico verified its functionality. However, performance varied with matrix composition, showing inhibition in samples with high manganese ( $\text{Mn}^{2+}$ ), chromium ( $\text{Cr}^{2+}$ ), and zinc ( $\text{Zn}^{3+}$ ). Despite these challenges, the sensor achieved a 113% recovery rate in Tula rainwater, indicating promise for easy *in situ* environmental monitoring. This research demonstrated the potential of laccase-based SPR biosensors to enhance microplastic detection and highlights the importance of considering matrix effects in practical applications.



**Figure 1** Schematic diagram of laccase-SPR biosensor. Reproduced from reference [29] with permission from MDPI (Copyright 2019)

## 2.2. Peptide-Based Biosensors

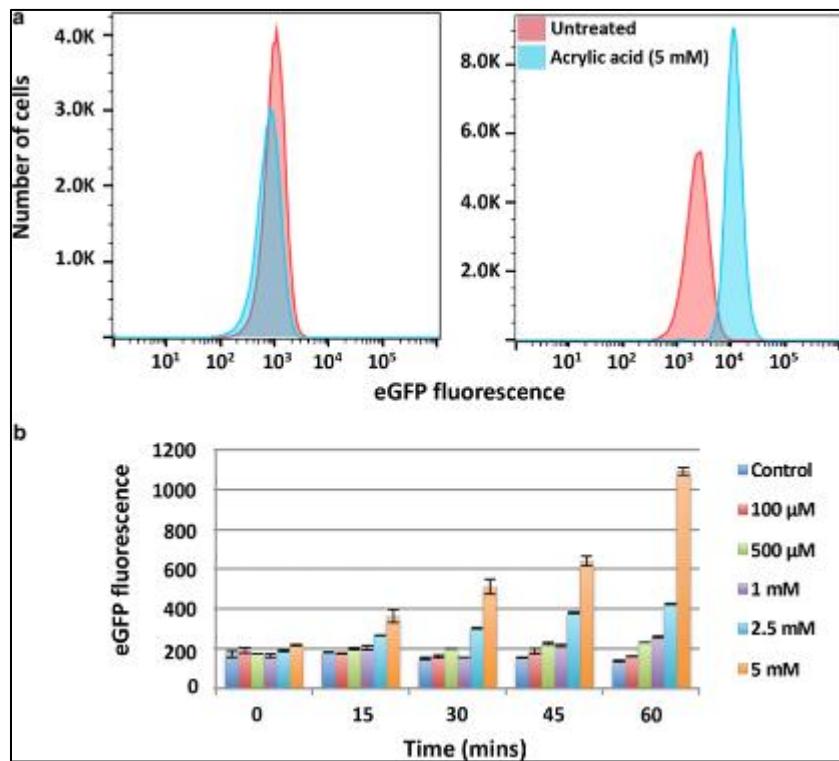
Peptides are polymers of amino acids linked by peptide bonds and are building blocks of proteins. In the past decade, polymer matrices have been designed to target specific peptide sequences using molecular imprinting technologies known as “plastic antibodies”. Recently, these plastic-binding peptides have been used to detect MNPs. Oh et al. demonstrated a localized surface plasmon resonance (LSPR) system for PS MPs and their fragmented debris [30]. Compared to the conventional SPR, a stronger plasmon is generated in LSPR by confining the incident light within the metal nanoparticles that are smaller than the wavelength [31]. The detection system consisted of a gold-layered sensor chip and 5 nm gold nanoparticles. Both gold components were functionalized with PS-binding peptide, HWGMWSY, sandwiching the PS MPs and their fragmented debris when they were present. The MNP binding to the LSPR sensor surface induced the wavelength shift detected by UV-vis spectroscopy. The LOD was 1  $\mu\text{g/mL}$ . Fluorescent dyes can be added to peptides to enable binding measurement. Rübsam et al. created eGFP-peptide fusion proteins that bind to plastic for PP and PS, with a microplate reader used to quantify fluorescence intensity [32]. Similarly, Woo et al. detected PS and PP MPs by attaching fluorescein isothiocyanate (FITC) to peptides. They also assessed the binding efficiency of various peptides to each plastic type using fluorescence microscopy to identify the most effective peptide sequence [33].

Currently, only a limited number of studies have been published on detecting MNPs using peptides. Additional research is necessary to discover a broader range of peptide sequences capable of specifically identifying MNPs. Although these efforts are primarily trial-and-error, recent progress in machine learning can greatly decrease the amount of effort required before conducting these experiments.

## 2.3. Cell (bacteria)-Based Biosensors

Cell-based sensors have a long history. Numerous cell-based approaches or techniques have been developed for detecting a wide range of targets, including biotoxins [34], biomarkers [36], and environmental pollutants [35]. Since

the target needs to enter the cell to interact with the promoter, the “lights on” cells indicate both the bioavailability and the amount of the substance. Raghavan et al. developed a whole-cell biosensor to detect acrylic acid, the breakdown product of polyacrylic acid (PAA) MNPs [37]. The process began with transcriptome analysis, which identified genes in *Escherichia coli* BL21 that were specifically upregulated in response to acrylic acid. Live cell biosensors were created by transforming *E. coli* BL21 with plasmids containing these genes and the eGFP gene. When exposed to acrylic acid, the responsive gene increased expression, leading to eGFP fluorescence, which was quantified via fluorescence microscopy. The *PyhCN*-eGFP *E. coli* strain showed the best performance among various strains with different promoters. The *yhCN* gene encodes an 87-amino acid polypeptide located in the periplasm, making its changes easier to detect due to its proximity to the cell surface. The detection limit was 500  $\mu$ M acrylic acid, though statistical significance was not discussed. Notably, the sensor demonstrated high specificity, as it produced no significant signal when exposed to other chemicals, such as acrylamide, lactic acid, or 3-hydroxypropionic acid. For faster and more sensitive detection, a different reporter, such as *Photinus pyralis* firefly luciferase (lucFF), could be used [38]. Another study designed a recombinant whole-cell sensor, *E. coli* pBAV1K-ACU-lucFF, using the lucFF reporter [39]. Bioluminescence in response to acrylic acid was measured with a microplate reader for samples treated with acrylic acid compared to controls. This biosensor can detect acrylic acid monomers at concentrations as low as 1 mM in sterile water and environmental samples, such as lake water. The strain was also incubated with polymer PAA for hours to test its ability to sense degradation products. Additionally, this method enabled high-throughput detection using a 96-well plate. The cells exhibited a dose-dependent response within a narrow range from 1  $\mu$ M to 1 mM. However, at higher concentrations of acrylic acid, toxicity reduced the response. Wang et al. investigated the impact of PE microplastics on the electrochemical and microbiological properties of exoelectrogenic biofilms in microbial fuel cells (MFCs) and microbial electrolysis cells (MECs) [40]. When the PE microplastics concentration increased from 0 to 75 mg/L in MECs, there was a noticeable decrease in maximum current density (from 1.99 to 0.74 A/m) and the abundance of electroactive bacteria (EAB) within the biofilm. In contrast, in MFCs, the current output remained largely unaffected, and EAB levels slightly rose at 25 mg/L of microplastics. Additionally, PE microplastics reduced biofilm viability in both systems, resulting in increased electrode resistance. The microbial community diversity and microplastics-associated operational taxonomic units also declined with higher microplastic levels. Moreover, genes related to electron transfer (such as *pilA* and *mtrC*) and cytochrome c concentrations diminished after microplastic exposure. This research provided initial insights into the effects of PE microplastics on exoelectrogenic biofilms, highlighting potential genetic mechanisms and establishing a foundation for developing effective water treatment technologies.



**Figure 2** a) Control *E. coli* (left) or stably transduced *E. coli* (*PyhCN*-eGFP) (right) were treated with acrylic acid (5 mM) overnight and analysed by FACS. b) Stably transduced AA reporter cells treated with indicated concentrations of AA and fluorescence measured over time. Reproduced from reference [37] with permission from BMC (Copyright 2019)

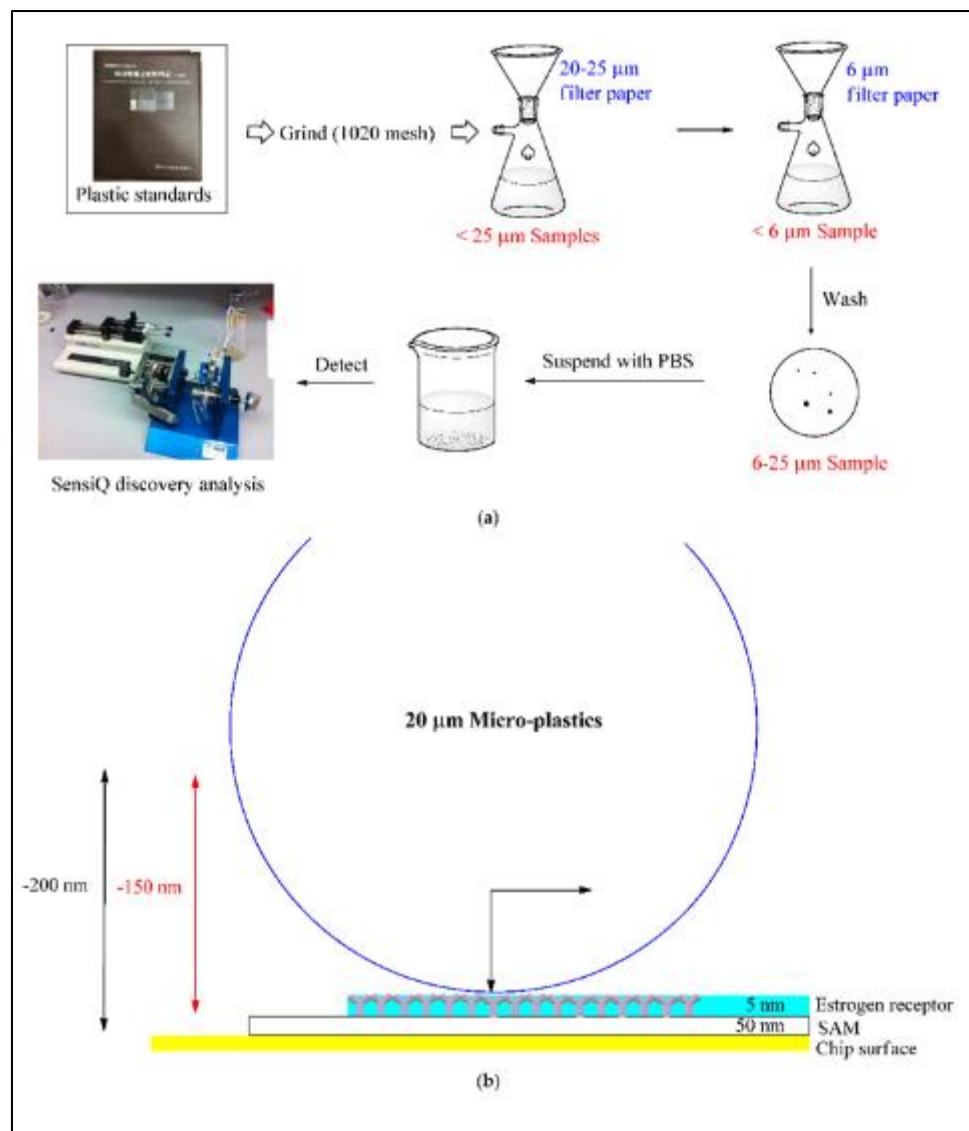
Unfortunately, these whole-cell biosensors are not ideal, as they are still in the laboratory stage, requiring bulky instruments such as fluorescence or microplate readers to measure the sensing signal. They have not been applied for detecting MNPs in field analysis. Many optical reader devices have recently been developed for portable and handheld platforms, often incorporating microfluidic devices for pre-treating and processing samples. As such, we may be able to demonstrate the whole-cell MNP biosensor in portable and handheld platforms in the future.

#### 2.4. Antibody and estrogen-Based Biosensors

Antibody-based immunoassay platforms, including Enzyme-Linked Immunosorbent Assay (ELISA), biosensors, and lateral flow assays, are well-established and widely employed for rapid detection of a wide range of targets, from microbes and proteins to small compounds. These immunoassays are valued for their high specificity and sensitivity, ease of standardization for both qualitative and quantitative results, and cost-effectiveness [43-45]. A study conjugated polystyrene (PS) nanoparticles, a common type of MNPs, to different carrier proteins for rabbit immunizations. The resulting PS-specific antibodies were strongly produced after immunization, with up to a 128-fold increase in IgG titer, as confirmed by multiple-tier immunoassays using various PS products. Notably, these antibodies did not react with polypropylene, a different plastic type. Additionally, a PS antibody-based enzyme-linked immunosorbent assay (ELISA) was used to demonstrate that this method can quantitatively analyze PS particles in various environmental matrices. Overall, this research showed, for the first time, the potential to generate plastic-specific antibodies, laying the groundwork for developing innovative antibody-based immunoassays for future risk assessment and management of MNPs, particularly at the nanoscale [46].

The estrogen receptor (ER) belongs to the nuclear hormone receptor superfamily, which are ligand-activated transcription factors [42]. A nano-plasmonic biosensor was developed by utilizing an Estrogen Receptor (ER) recognition element grafted onto a polymer-based gold nanograting (GNG) plasmonic platform [42]. The ER-GNG biosensor required only small sample volumes (2  $\mu$ L), enabled rapid detection (3 min), and could determine nanoplastics in simulated seawater with a linear dynamic range of 1-100 ng/mL, covering the expected environmental loads. The nanostructured grating (GNG) provided significant performance improvements, extending the measurement range across five orders of magnitude, thanks to both the SPR and localized SPR phenomena occurring at the GNG chip. Finally, the ER-GNG biosensor was tested on real seawater samples collected from the Naples area, with results (~30 ng/mL) verified by a conventional method (filtration and evaporation), confirming that the ER-GNG sensor offers a straightforward and highly sensitive approach for direct in-field nanoplastic monitoring. Another study investigated the movement mode and low-concentration detection of microplastics using surface plasmon resonance (SPR). Initially, 20-micrometer microplastics were prepared through grinding and filtering, and their movement was examined. The characteristics were then analyzed via SPR. Chromatographic analysis revealed that the surface charge of microplastics primarily affected elution time, with estrogen receptors (ERs) playing a supporting role. A variation in micro-plastics was observed in the SPR sensorgram, suggesting that the micro-plastics move in a rolling mode on the ERs. Analysis showed that the response of low particle numbers of microplastics on SPR was linearly related to the response unit (RU). When ERs were immobilized on the biosensor, the binding strength of microplastics to ERs under ultra-low background conditions corresponded to the dissociation rate constants: PS (0.05 nM) > PVC (0.09 nM) > PE (0.14 nM). An ELISA-like magnetic bead experiment confirmed the specificity between ERs and microplastics [41]. Therefore, using SPR, a higher binding affinity and longer retention time of PS were observed, indicating its biological over-occupation via ERs. Future potential applications include identifying microplastics in natural samples, biomarking, real-time detection in specific environments or regions, and assessing impacts on human health.

The main challenge facing antibody and estrogen-based biosensors for plastics is their high specificity for individual chemicals. Detecting various plastic types requires identifying a broad range of antibodies, enzymes, and receptors. Additionally, morphological differences in MNPs can influence the sensitivity and specificity of protein-based plastic sensors. Notably, the number of research studies on protein-based plastic sensors remains very limited.



**Figure 3** Schematic presentation of the experimental procedure. (a) General procedure of detection of micro-plastics based on SPR biosensor. (b) Schematic presentation of micro-plastic detection using ERs immobilized on COOH5 SPR sensor. Here, 200 nm (double arrow) refers to the detectable range of SPR; 150 nm (double arrow) refers to the detectable range of micro-plastics after subtracting the SAM and estrogen receptors immobilized on the chip; the arrow inside the particle indicates the driving force of gravity and the flow rate of the micro-plastics in the microfluid.

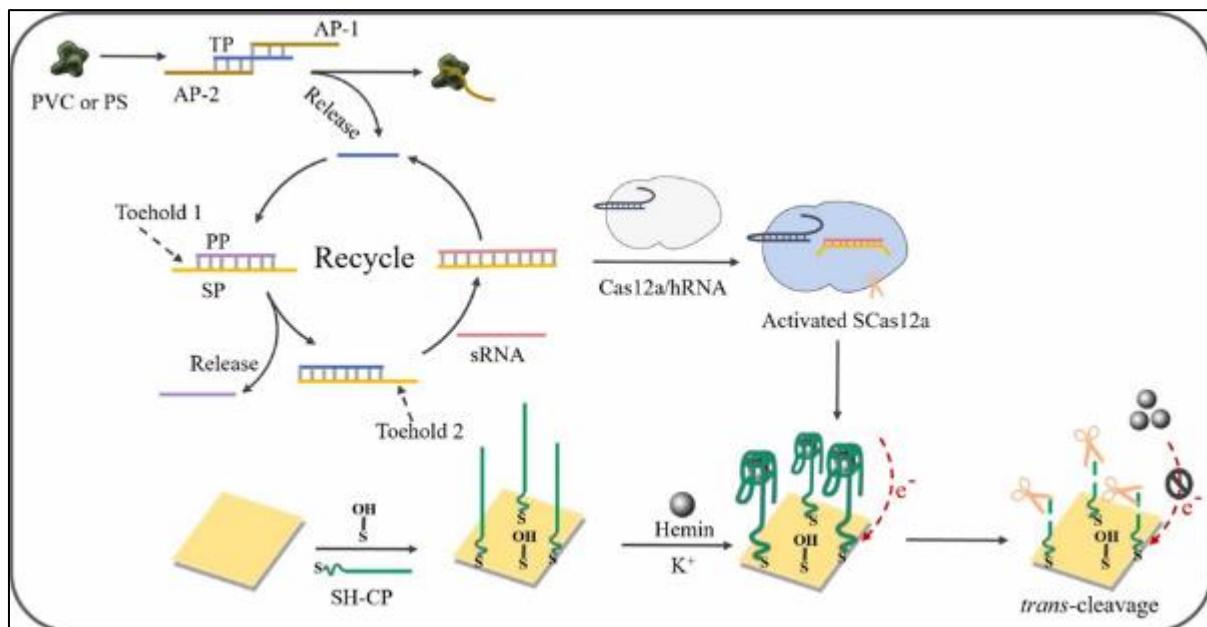
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## 2.5. Aptamer-based Biosensors

Aptamers are single-stranded DNA or RNA molecules that change their three-dimensional shape when they bind to specific target molecules, allowing for high affinity and selectivity [47, 48]. DNA aptamers are useful for detecting a wide variety of biomedical and environmental substances [49-51]. Aptasensors offer several advantages over other sensor types. First, an aptamer can act as a receptor that binds to a specific target or interacts with multiple similar molecules [52, 53]. Second, different aptamers targeting various molecules can be integrated into a single sensor to identify multiple compounds at once [54, 60]. Moreover, aptamers can be used with various transducers, including electrochemical, optical, and colorimetric methods, without being restricted to only one signaling platform [55-58]. A new label-free electrochemical aptasensor, called CRISPR-MP, was developed for detecting MPs using split gRNA with CRISPR/Cas12a-mediated cascade strand displacement (CSDR) and aptamers. When MPs (PVC/PS) are present, the split gRNA activated CSDR, leading to the formation of multiple heteroduplexes. These heteroduplexes then activated the DNase activity of SCas12a upon binding to Cas12a/handle RNA complexes. The active SCas12a subsequently cleaved the G4/hemin complex on the gold electrode, resulting in a shift in current signal. This strategy eliminated the need for redundant probes, lowering detection costs. The system can detect as low as 37 ng/mL of PVC and 45 ng/mL of PS. The

CRISPR-MP system features two key innovations: 1) a dual-signal amplification method driven by SCas12a's sRNA to enhance CSDR, and 2) the novel integration of the CRISPR system with aptamers for MP detection [59].

Currently, only a few studies have been published on detecting MNPs with aptamers. More research is needed to identify a wider variety of aptamer sequences that can specifically target MNPs.



**Figure 4** Schematic illustration of CRISPR-MP system for MP detection. Reproduced from reference [59] with permission from Elsevier (Copyright 2025)

### 3. Research gaps and future directions

Many sensors can detect micrometer-sized plastics, but accurately measuring particles smaller than 1  $\mu\text{m}$  in complex matrices remains challenging. Techniques like O-PTIR 25 and nanofabricated SERS substrates 14 are advancing, yet their availability is limited. Future efforts should focus on scaling these technologies. There is also an urgent need for sensors that can selectively identify MPs among abundant natural particles such as clay, silica, and diatoms. This challenge has received limited systematic study. Future research could explore multiplexed readouts, such as combining fluorescence intensity with lifetime (FLIM) or polarization, to enhance signal differentiation. Currently, most sensors are proof-of-concept prototypes tested only in laboratories, with few undergoing long-term field validation (beyond 6 months) or inter-laboratory comparisons. Developing reliable microfluidic chips that integrate sample preparation, capture, and detection is a key future priority 30. Additionally, the library of biorecognition elements for plastics remains limited; while progress has been made with antibodies for PS and aptamers for PVC/PS, there is significant potential to discover and engineer new plastic-binding molecules, such as synthetic peptides used in the LSPR sensor 10, to target the most common polymers. Finally, the absence of certified reference materials, especially for nanoparticles and environmentally aged plastics, presents a major obstacle for method validation and data comparability across studies.

### 4. Conclusion

A recent shift in microplastic analysis has transitioned from traditional laboratory methods to innovative, rapid, and portable optical biosensors. These biosensors excel in sensitivity and selectivity, with detection limits in the low  $\text{ng L}^{-1}$  range—crucial for evaluating ecological and health impacts. By delivering fast and precise information on microplastic distribution and levels, these sensors will play a pivotal role in the global efforts to understand, manage, and mitigate plastic pollution. I hope this article provides readers with an updated view of the latest advancements in micro- and nanoplastics biosensor detection systems, enhances their understanding of the subject, and inspires further innovations in portable nanoplastics biosensor technologies.

## Compliance with ethical standards

### Disclosure of conflict of interest

No conflict of interest to be disclosed.

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