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# Recent developments on magnetic molecular imprinted polymers (MMIPs) for sensing, capturing, and monitoring pharmaceutical and agricultural pollutants

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## **Abstract**

Among the many classes of pollutants, bioactive contaminants are causing significant concern include pesticides, herbicides, and other pharmaceutical active compounds (PhACs). These chemicals have been linked directly to contamination of fresh water and food sources, threatening water and food security. Contamination from antibiotics, one class of PhACs, is also considered to link strongly to the global problem of antimicrobial resistance (AMR). Pollution from these contaminants have hit developing countries harder due to less stringent legislation on waste discharge from agricultural industries. Current sampling and monitoring methods (e.g. LC-MS) are expensive, time consuming, immobile and requiring skilled users. Recent development of devices fitted with molecular imprinted polymers (MIPs) have been found to be particularly attractive due to their low cost, stability, high selectivity and mobility. MIPs can also be used to extract these organic pollutants at low concentrations. The latest development of magnetic MIPs (MMIPs) further facilitates the separation and recovery of these absorbents by using an external magnetic field after the target molecules have been bound. Thus avoiding laborious centrifugation and filtration during separation and recovery. The purpose of this perspective is to summarize the recent development of MMIPs in the past 10 years. We will focus on their applications in food industries and the agricultural sector. Several potential developments in combined analytical techniques using MMIPs will also be discussed. As pollution to our environment is now a focal point in human life, new analytical techniques based on MMIPs will be of great interest to the science communities.

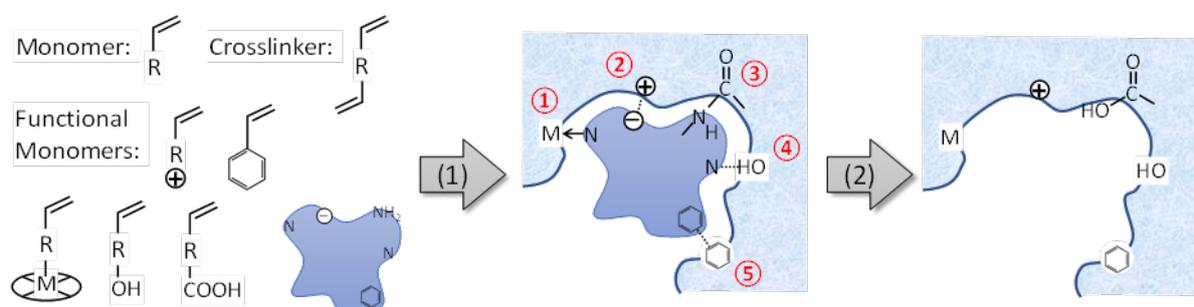
## INTRODUCTION

The ever-expanding global population has put extreme pressure on both food and water supplies. To meet the ever increasing food demand, science and technology (particularly in agriculture) have provide many solutions to maintain a strong supply of food, by using drugs, growth hormones, pesticides, and fungicides. However, excessive use of these pharmaceutical active compounds, or PhACs, has caused pollution problems to the environment, particularly in developing countries. PhACs uncontrollably can enter the natural water systems via agricultural activities, contaminating food stocks and threatening fresh water supplies [1]. Furthermore, some PhACs, such as antibiotics, have led to the development of antimicrobial resistance (AMR) in some bacteria [2]. These multidrug resistance (MDR) bacteria become one major concern in WHO [3]. Moreover, these pollutants also directly contribute to the contamination of food and fresh water for human consumption, directly threatening food and water security.

To tackle this problem efficiently and effectively sophisticated monitoring of PhACs is urgently required, but the challenge is the detection of ultralow concentration of PhACs (usually in ppb range) in food and water sources. Common methods to detect antibiotics and other PhACs include mass spectrophotometry [4], liquid chromatography [5], solid phase extraction (SPE) [6], capillary electrophoresis [7] and electrochemical techniques [8]. These methods have notable disadvantages as they are time-consuming, expensive, highly complex and require experienced users. Sophisticated automation systems are also necessary for continuous monitoring while the mobility of instrumentation needs to be enhanced. These would also aid in monitoring and sampling of water sources in remote areas. As a result, there is an urgent need for new detection methods and systems with the requirements of being: highly sensitive, specific, fast response time, reproducible, cost effective, reliable, portable and easy to implement by users.

## DEVELOPMENT OF MOLECULARLY IMPRINTED POLYMERS (MIPS)

In the past few decades, molecular imprinting technique with precise molecular recognition has been used in various scientific fields, namely in the analysis of organics within the environment and biomedical diagnoses. Molecularly imprinted polymers (MIPs) have shown advantages including their rapid and highly selective adsorption on target analytes [9]. MIPs are synthesized from polymerization of specific functional monomers, an initiator and a stabilizer in the presence of molecules of a targeted template. The functional monomers have to be chosen with consideration on the covalent or non-covalent interactions with the targeted template (see Figure 1). After polymerization, the template molecule is removed from the polymer matrix with a suitable desorption agent to generate printed cavities that have a complementary size, shape, three-dimensional structure and binding sites with high affinity for a specific analyte, usually the template molecule itself [10]. For monitoring organic compounds from complex matrices i.e. natural water resources, wastewater, food samples and human fluids, the high selectivity of MIPs is a clear advantage over conventional chromatography techniques. MIPs also have advantages over antibodies and natural receptors as they exhibit higher chemical and mechanical stability, as well as long shelf life. The preparation of MIPs is also relatively simple and of low cost.



**Figure 1.** Schematic to illustrate the formation of a MIP with binding sites. The five types of interactions that are used in MIP binding are highlighted as (1) co-ordination bonding, (2) opposite charge, (3) covalent bonding, (4) hydrogen bonding and (5) hydrophobic interaction.

Recently, MIP nanoparticles have gained increasing interest as they have superior properties compared to conventional bulk MIPs such as cryogel, monolith, microparticles [11] and membranes [12]. These superior properties of MIP nanoparticles include high surface area, fast binding kinetics, high binding amounts, improved stability, good accessibility and easy handling during experiments [13, 14]. One obstacle for using MIP nanoparticles lies on the recovery of these valuable materials. Separation using filtration causes a significant loss of materials since nanoparticles can easily be trapped in filters (e.g. filter papers). Some nanoparticles required filters with fine pores ( $< 100$  nm), which is not widely available and can be easily blocked causing slow filtration. Centrifugation can be adapted but it is a laborious method.

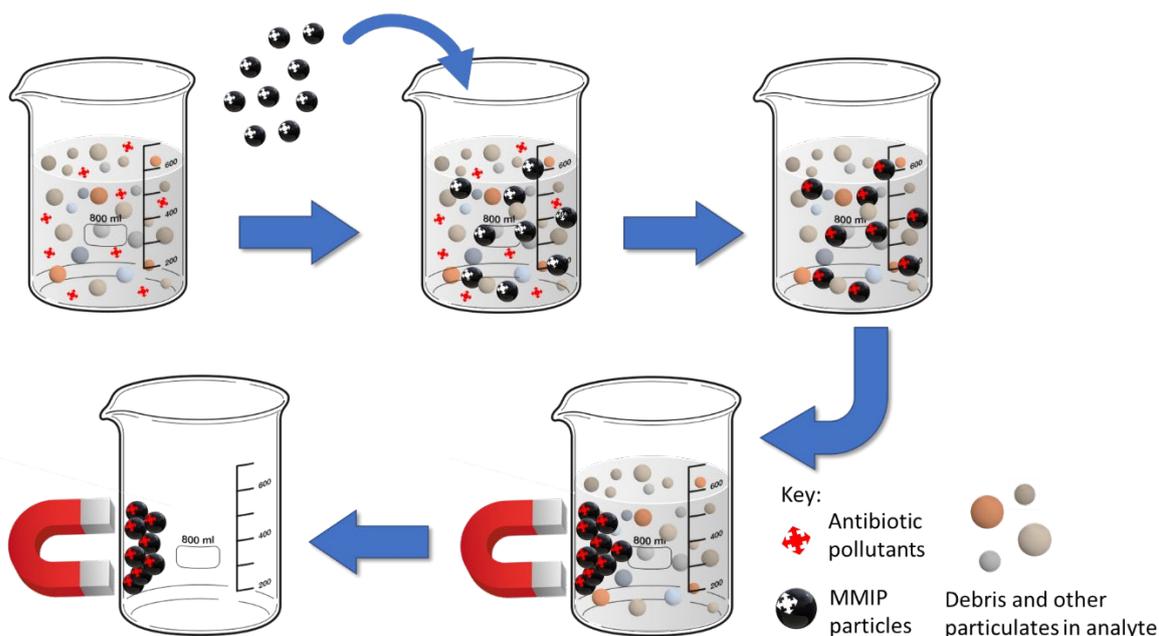
### **MAGNETIC MOLECULAR IMPRINTED POLYMER (MMIP)**

Separation and recovery of materials using a magnetic field have been used in biomolecular separation for decades [15-17] and this method is particularly advantageous when the amount of material is too small to recover through filtration or centrifugation. The combination of magnetic separation with molecular recognition ability of MIPs can be a power tool in analytical sciences, especially for analysis of pollutants at low concentrations in complex analytes.

A magnetic MIP (MMIP), can be prepared by embedding magnetic nanoparticles during the MIP synthesis. There are several different polymerization routes for the preparation of MMIPs but in general the principles are similar. The essential ingredients for preparing an MIP were discussed in the previous section. For MMIPs an extra magnetic component is required. This magnetic component can either be embedded inside the MIP structure or act as a platform for coating the imprinted polymer on surface. The main aim is to ensure the final MMIP materials can respond to an external magnetic field through the

magnetic component firmly attached to the MIP. Iron oxide based magnetic materials (e.g.  $\text{Fe}_3\text{O}_4$  or  $\gamma\text{-Fe}_2\text{O}_3$ ) are the most widely used for this application. [18,19] Surface functionalization is usually required for creating strong binding to the polymer but unfunctionalized iron oxide have also been used [20]. Commonly used functional surfaces include amine and vinyl groups [21-23]. For instance, vinyl groups on iron oxide surface can be co-polymerized with vinyl monomers such as methacrylic acid, which is amongst the most popular monomers for MIP synthesis [24, 25]. Other materials such as magnetic carbon nanotubes (MCNT) offer electrical conductivity properties and have also been used as a platform for coating MIP polymers [26, 27].

Applications of MMIPs so far have been limited to use in laboratory. In practice, the advantages of using MMIP over conventional non-magnetic MIP stand out in the capture of target molecules from complex analyte samples. Since these samples can have large amounts unwanted solid debris and particulates, e.g. soil samples, river water and sewage. If a non-magnetic MIP is used, it becomes difficult to recover the MIP materials once it has captured the target molecules or pollutants, leading to difficulties in the follow-on analysis on them. Using MMIPs, the captured pollutants or targeted molecules can be isolated magnetically from these complex analytes, as shown in Figure 2. Another scenario for needing MMIP is if non-magnetic MIP cannot be made in large enough quantities for other means of separation/recovery, including filtration or centrifugation. For instance, the template molecules are simply too expensive for use in a meaningful quantity, MMIP may be a possible solution to minimize the loss of MIP materials. Here we introduce some examples in three areas of applications that present particular challenges to non-magnetic methods due to the complexity of the analytes.



**Figure 2.** Illustration of the principle in separating/recovering MMIPs from a complex matrix with solid debris/particulates using a magnetic field.

### **MMIP for extraction and monitoring for pharmaceutical pollutants (PhACs) in sewage**

Although there are many MMIP examples used in targeted applications for the extraction of PhACs from water sources. Many studies in the early stages were carried out in relatively clean samples (e.g. river water, tap water or sea water) spiked with pollutants [28-30]. Studies using MMIPs in sewage samples are few and far between. This highlights the challenge in working with real complex analytes. Qin et al. [31] prepared chitosan-based MMIP materials for the detection and removal of sulfonamides (a class of antibiotics) in water and sewage samples. It was found that  $112 \text{ ng.L}^{-1}$  of sulfamethoxazole was in the sewage sample using MMIPs while only  $64 \text{ ng.L}^{-1}$  of sulfamethoxazole was found in the sample using a C-18 extraction column. This suggests that the MMIP was a more efficient extractor for sulfamethoxazole. Zuo et al. have also demonstrated the synthesis of MMIP for extracting dimethyl phthalate from soil samples [32].

### **MMIP for monitoring pesticides and herbicides in soil samples**

As the food demand increases in response to the growing population in the world, increase use of pesticides and herbicides to ensure food productivity is inevitable. Despite advancements in modern technologies in agriculture, the application of active chemicals is used in a uncontrolled fashion and in high quantities. The method of spraying crops is widely practised around the world. However, this practice causes huge environmental problems; while achieving to destroy their targets (insects or weeds), it promotes pesticide resistance [33], food contamination [34] and further environmental impacts. Furthermore, the residual pesticides and herbicides remain in soil as contaminants and then leach and runoff to contaminate ground and surface water [35]. As a consequence, the concentration of these active chemicals can be different in each environmental compartment (soil, ground or surface water). It is important to monitor them in low concentrations to avoid environmental problems and to detect the levels of contamination in agricultural areas.

To tackle this, soil samples needs to be closely monitored but extracting the target contaminants from soil is not easy due to the soil's complex composition. Several examples in the literature have demonstrated the possibility of using MMIP for extracting these agricultural contaminants from soil. One early example of research in this area was reported by Zhang et al. who demonstrated the use of MMIP 100 – 200  $\mu\text{m}$  particles (prepared using microwave heating) for extracting triazine herbicides from soil, grains, and vegetable samples [36]. Analysis using HPLC-MS showed extraction from spiked samples using MMIP was over 70% for all triazines after 30 minutes. With a similar principle, Miao et al. demonstrated the preparation of MMIP microspheres of 1-2  $\mu\text{m}$  for extracting a herbicide bensulfuron-methyl (BSM), from soil, rice and rice water samples [37]. The binding capacity (Q) was found to be around 38  $\text{mg.g}^{-1}$  but the non-imprinted sample also recorded a high 19  $\text{mg.g}^{-1}$  at maximum, giving an imprinted factor (IF) of *ca.* 2. A selectivity test was also carried out against other sulfurons, showing a selectivity factor (SF) from 1.8 to 10. Nonetheless, the

MMIP microspheres were able to extract  $\geq 90\%$  of bensulfuron-methyl from spiked soil samples which was around a 30% improvement from C-18 extraction cartridges. Xu et al. reported the synthesis of MMIP particles (0.5 – 1  $\mu\text{m}$ ) was prepared using silica-coated  $\text{Fe}_3\text{O}_4$  as the magnetic component for the extraction of methyl parathion from a soil sample [38]. The MMIP sample showed a high IF of 12 but the binding capacity of the target methyl parathion was relatively low (at  $Q = 9.1 \text{ mg}\cdot\text{g}^{-1}$ ). The spiked soil samples showed a recovery of  $> 80\%$  of methyl parathion was recorded. These examples show that MMIPs can be used for extracting agricultural contaminants from soil with an improved efficiency than C-18 extraction columns.

### MMIP for binding contaminants in food samples

Another area of interest is extraction and the monitoring of contaminants in food samples. Although food samples are also of high complexity, more research in this area have been seen in the literature than that on sewage or soil. This is possible due to food safety is in a higher priority in the public. Nonetheless, challenges in selective binding using MMIPs remain. Use of MMIPs have been seen for the extraction of a broad spectrum of contaminants (from antibiotics to pesticides) from a wide range of food samples (meat, dairy products, grains, and vegetables). Table 1 summarizes several examples available in the literatures.

Table 1. Examples of MMIP used for extraction of contaminants from food and drink samples.

Test samples	Contaminants/ MMIP templates	Magnetic component	Monomers/ crosslinkers	Binding capacity ( $Q$ in mg/g)	Ref
<i>Meat products</i>					
Pork	Sulfamethazine	$\gamma\text{-Fe}_2\text{O}_3@\text{ZnO}$	MAA; EGDMA	653	39
Fish, pork, shrimp	Erythromycin	$\text{Fe}_3\text{O}_4@\text{SiO}_2\text{-MPS}$	MAA; EGDMA	94.1	40
Fish	Erythromycin	MMWCNTs	MAA; EGDMA	22.8	41
Fish, pork	E1, E2, E3, EE2	$\text{Fe}_3\text{O}_4@\text{ZIF-8}$	ABPA; ABPA; APS	ND	42

<i>Fruits, crops and vegetables</i>					
Rice	Chlorpyrifos	Fe <sub>3</sub> O <sub>4</sub> @SiO <sub>2</sub>	MMA	171	43
Orange	Carbendazim, thiabendazole	Fe <sub>3</sub> O <sub>4</sub> @SiO <sub>2</sub>	MAA; EGDMA	1.75	44
Fruits	β-Cyfluthrin, cyhalothrin, cyphenothrin, permethrin	MCNT@SiO <sub>2</sub> -MPS	MAA; EGDMA;	225	45
Tomato	Triazines	Fe <sub>3</sub> O <sub>4</sub> /PEG	MAA; TRIM, DVB	ND	46
<i>Drinks</i>					
Honey	λ-Cyhalothrin	Fe <sub>3</sub> O <sub>4</sub> @SiO <sub>2</sub> -MPS	AM, DVB	ND	47
Strawberry juice, milk	Triazines	Fe <sub>3</sub> O <sub>4</sub> /PEG	MAA; TRIM, DVB	ND	46
Tea	Dicofol, pyridaben	Fe <sub>3</sub> O <sub>4</sub> @OA	MAA; EGDMA; DDT	ND	48
Milk	Enrofloxacin	Fe <sub>3</sub> O <sub>4</sub> @POSS	MAA; POSS	67.3	49
Milk	β-Lactam	Fe <sub>3</sub> O <sub>4</sub> @OA	MAA; EGDMA; PENV	20.2-49	50
Milk	Bisphenol A	Fe <sub>3</sub> O <sub>4</sub> @MAPS	VP; EGDMA	390	51

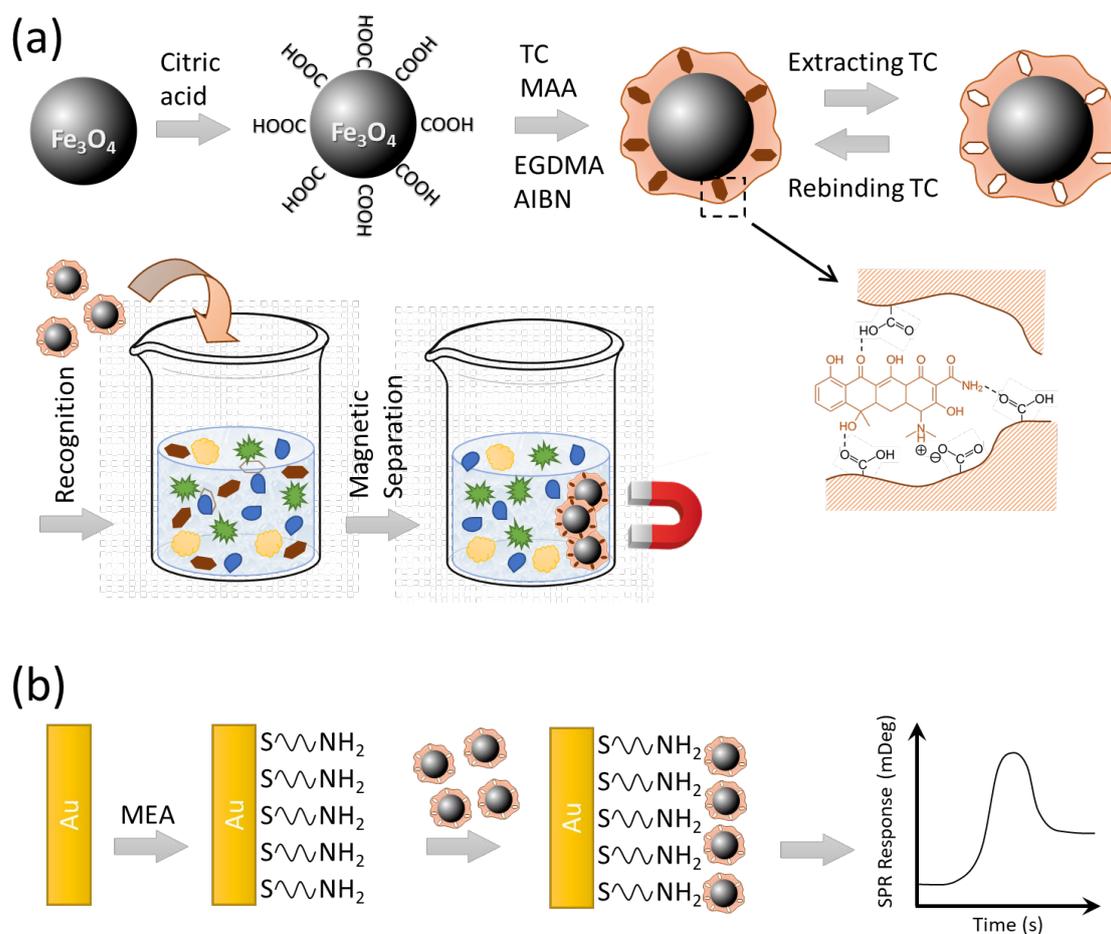
Latest developments in this area include some responsive MMIP constructs. For example, a light responsive MMIP was prepared for extracting dimethoate from olive oil [52]. Unlike conventional MIP, the release of the dimethoate template from MMIP can be triggered by light. Li et al had developed a flat 2D MMIP on boron nitride (BN) for extracting flavonoids from Ginkgo biloba leaf extracts [53]. Enhanced recoveries for all three tested flavonoids (>93%) were recorded compared with conventional MIPs. Wang et al. reported the preparation of a hollow MMIP construct for detecting triazines from crops including corn, wheat, and soybean samples [54]. Unlike most MMIPs, where the magnetic component tends to be embedded inside or coated with the MIP polymer, this construct immobilized magnetic Fe<sub>3</sub>O<sub>4</sub> nanoparticles by growing on the dialcohol groups on the nanoparticle surface. In addition to new design on the MMIPs, Zhang et al. demonstrated an internal extractive electrospray ionization mass spectrometry (iEESI-MS) system for quantifying fluoroquinolones in milk samples [55]. This system by-passed the releasing the target

molecules stages, which can be time consuming and reducing the accuracy of analysis if releasing is less than 100%.

### **COMBINED MMIP TECHNIQUES FOR ANALYSIS**

As discussed previously, the advantages of using MIPs include high loading capacities and enhanced selectivity allowing for improved sample recovery. The added magnetic property of MMIPs allows for an easier handling of samples. This has been demonstrated in the quantification of target organic molecules at low concentration while the eluent is analyzed by various common instruments [56-58]. In recent years, the effect of emerging pharmaceutical pollutants has been an increasing cause for concern from evidence linking them to many global issues such as AMR, destruction of ecosystems, and threatening water and food security. Using tetracycline (TC) as an example, TC is a broad-spectrum antibiotic and widely used in agriculture due to its low cost. However, use of TC is mostly mismanaged; it is added to the animal feed as a supplement to reduce bacterial infections, regardless the health conditions of the animal. This widespread use of TC has resulted in trace amount of residues in animal-derived food. These residues eventually transfer to humans via the food chain and are harmful to humans' health [59]. The European Union (EU) and US Food and Drug Administration (FDA) have established maximum TC residue limits in various food items [60]. Monitoring of TC residues in food has become essential. There are numerous methods to measure the level of TC residues in food, but in many current methods (e.g. solid-phase dispersion [61] or magnetic MIP extraction [62, 63], followed by LC-MS/MS) complex extraction and separation steps are required before measuring the TC contents. Gao et al. [64] highlighted the advantages of using MMIPs for the detection of pharmaceutical pollutants using a MMIP for TC detection in milk. They combined the MMIP with the surface plasmon resonance (SPR) to achieve a very low detection limit of 1.0 pg/mL. In their study, methacrylic acid (MAA) was chosen as the

functional monomer, ethylene glycol dimethylacrylate (EGDMA) as the cross linker and citric acid modified  $\text{Fe}_3\text{O}_4$  nanoparticles were used as the magnetic component. The MMIPs were able to extract TCs from milk samples and allowed separation and recovery using a strong magnet. The TC-bound MMIPs were flowed over the mercaptoethylamine (MEA) modified SPR chip. The SPR response is due to the interaction between amino groups of the chip surface and hydroxyl groups or carbonyl groups of the MMIPs. Figure 3 illustrates the whole working process using this MMIP-SPR combined detection method. Indeed, such a simple but delicate combined method allows the detection of many organic molecule contaminants in a complex matrix with a low detection limit, rivalling high-end analytical techniques including HPLC-MS. A similar study using combined MMIP-SPR technique has also demonstrated in the detection of pesticide residues [65].



**Figure 3.** Schematic diagram illustrating TC detection by (a) MMIP extraction from water sample, and (b) MMIP-SPR measurement.

Magnetic MIPs have also been incorporated with electrochemical sensing methods, mostly as a coating material on a sensing electrode. Since this MMIP coating needs to be conductive, many research studies had chosen magnetic carbon nanotubes (or multi-walled carbon nanotubes MWCN) or magnetic graphene oxide (GO) as the magnetic component. For example, MMIPs has been used for the capture and preconcentration of sulfonamides and then deposited onto the electrode surface for electrochemical detection. [66] The detection limit was found to be extremely low at  $2.8 \times 10^{-4}$  ppb (S/N = 3). However, in this work, the MMIP was not directly incorporated onto the electrode when the measurement was carried out. Another example had demonstrated a similar method for the detection of  $17\beta$ -estradiol (E2) hormones and showed a low detection limit (0.82 nM) with a good selectivity against other steroid hormones [67].

In addition to SPR and electrochemical detecting methods, MIP techniques can also be incorporated with other analytical techniques for the detection of organic pollutants, such as resonant mass sensors, which mainly consists of Quartz Crystal Microbalance (QCM) and microcantilever sensor systems. They provide an advantage of label-free analysis and allow pico-gram mass resolutions. Combining resonant mass sensors with MIPs becomes one of the key developments in sensing and monitoring pharmaceutical pollutants in water sources. In one study, a QCM sensor incorporated with a poly(2-hydroxyethyl methacrylate-N-methacryloyl-L-tyrosine methylester) MIP was used for the real-time detection of  $17\beta$ -estradiol [68]. This QCM-MIP sensor exhibited a high specificity and sensitivity in a wide range of concentration (3.67 pM–3.67 nM) with the detection and quantification limits as low as 613 fM and 2.04 pM respectively. This sensor also showed an improved selectivity of almost 2.5 times when the detection was carried out against similar molecules (stigmaterol and

cholesterol). This sensor represents a promising, cost-friendly alternative for quantification and monitoring of estradiol pollutants in ground water. In another study, an MIP microsphere-modified QCM sensor (prepared via a spin coating method) has been demonstrated to detect endosulfan from drinking water and milk samples [69]. The sensor showed a good selectivity against similar molecules and specificity for the recognition of endosulfan, with a good linear correlation over 10–40 ng/mL and a detection limit of 5.59 ng/mL. Moreover, the chips could be reused for six times and have a shelf life of 6 months.

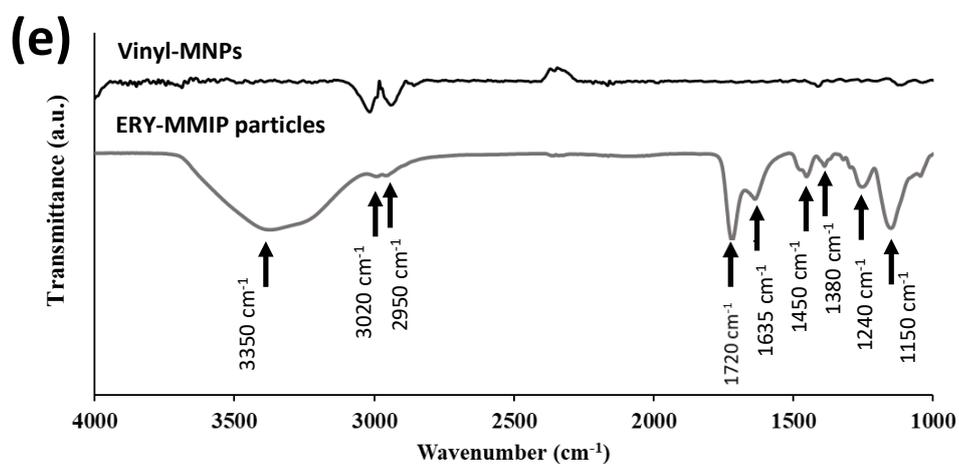
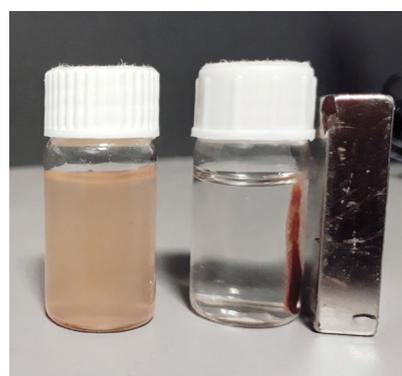
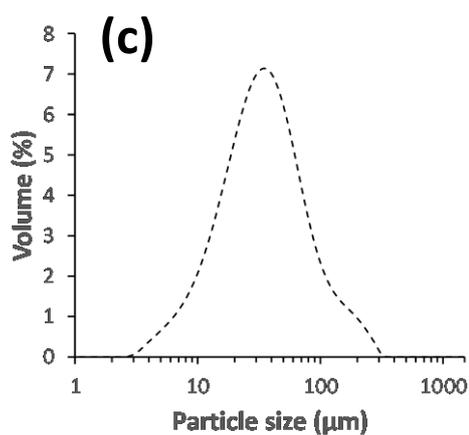
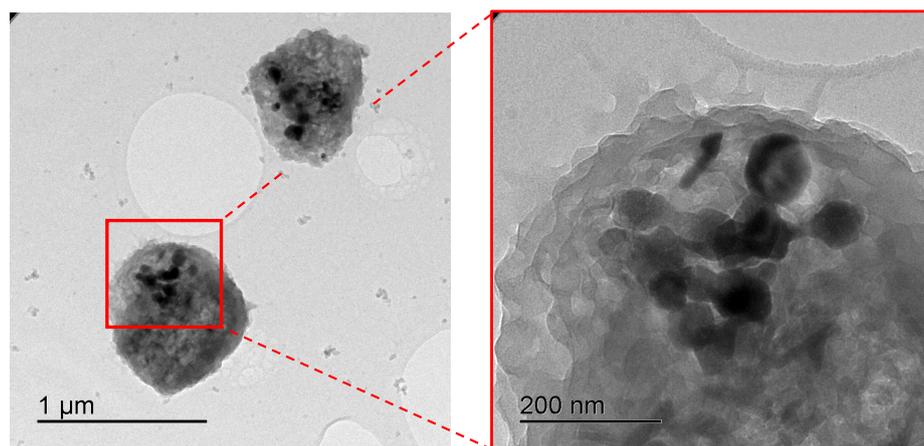
Micromechanical sensor is another technique that senses an accumulating mass through a MIP layer on cantilevers. Once the target molecule is in contact with the MIP cavity sites it produces a frequency change of the cantilever. This technique has been demonstrated recently in the detection of antibiotic ciprofloxacin (CPX) in water sources [70]. In this study, highly specific CPX imprinted nanoparticles (160 nm) were chemically bound onto the cantilever surface via the carbodiimide (e.g. EDC) and N-hydroxysuccinimide (NHS) crosslinking route. This sensing system showed a linearity for the concentration range of 1.5–150.9  $\mu\text{M}$  and a sensitivity of 2.6 Hz/pg. It also showed a high selectivity against another antibiotic with a similar structure, enrofloxacin (ENF). In a separate study, the particle size of MIP nanoparticles was found to have a positive effect on the sensitivity [71]. A similar system incorporated with erythromycin (ERY) imprinted polymeric nanoparticles of 22 nm were found to have a higher sensitivity of 1.58 Hz/pg at the range of concentration from 0.68 to 67.94  $\mu\text{M}$ . Furthermore, the adsorption of target molecule ERY was found to follow the Freundlich model. The combination of imprinting technology with resonant mass sensor is simple and comparable to conventional techniques in terms of selectivity, sensitivity and reusability. However, use of magnetic MIPs has not been reported although their magnetic property may offer additional advantage in adhesion onto the cantilever. This can be the next stage of development in this area.

## MMIP PARTICLES FOR SELECTIVE BINDING OF ANTIBIOTICS: A CASE STUDY

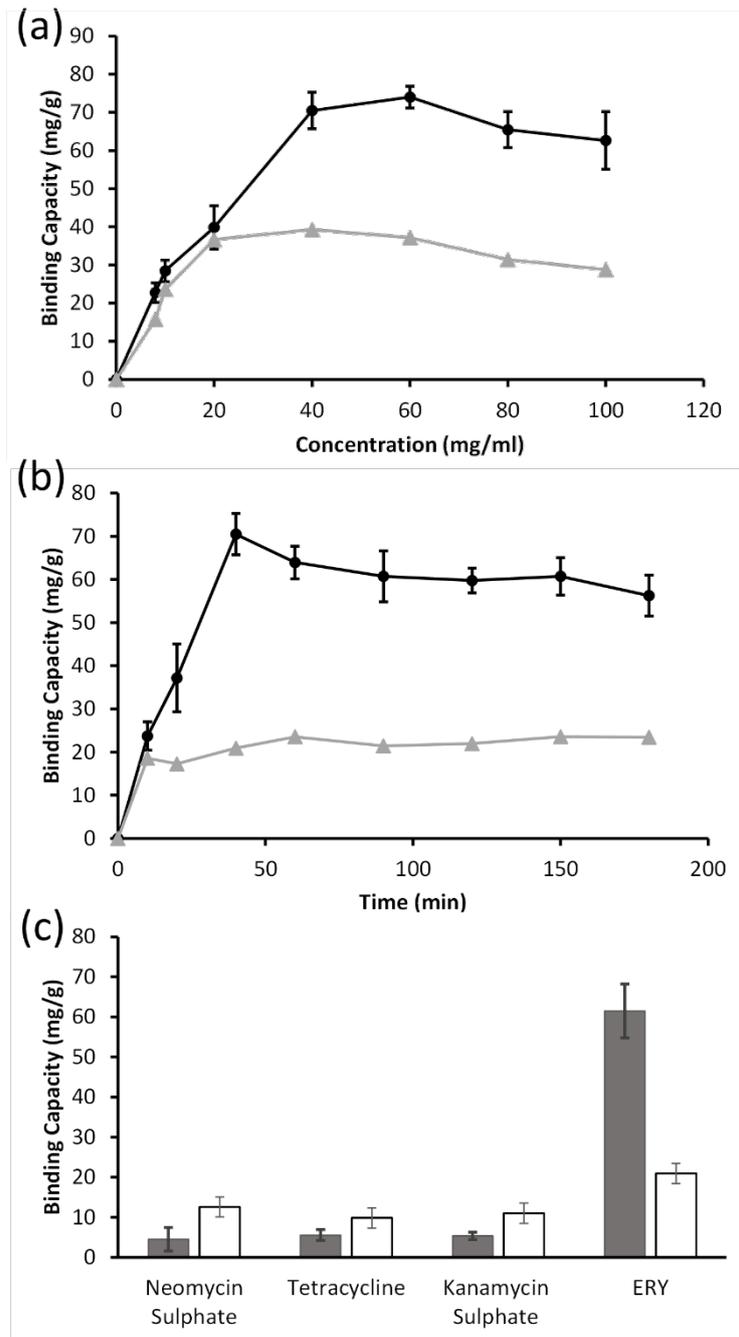
Here, we demonstrate the preparation of a MMIP particle sample using erythromycin as the antibiotic template. Erythromycin is a widely prescribed antibiotics, which is also on the WHO's Essential Medicines List (EML) [72]. Therefore, it can be found in municipal wastewater streams [73]. A system capable to selectively bind erythromycin in a complex medium would be essential for its monitoring in wastewater. In brief the erythromycin-specific MMIP particles (ERY-MMIP) were prepared using MAA as the monomer and EGDMA as the crosslinker via a two-phase microemulsion polymerization route (see Supplementary Information S1). Vinyl-functionalized Fe<sub>3</sub>O<sub>4</sub> nanoparticles (25 nm) were used as the magnetic component. A hand-held homogenizer was used to form the particles of ca. 1 μm. It is worth to note that the power and speed of the homogenization is critical to the formation of the particles, which was isolated using a neodymium-iron-boron NdFeB magnet.

TEM analysis revealed that the MMIP particles formed had a diameter around 1 – 2 μm with magnetic nanoparticles embedded inside the polymer particles (Figure 4a-c). However, the dynamic light scattering (DLS) results showed a hydrodynamic size much larger than the particle size measured from our TEM study (Figure 3d). Although there are many reports in the literature showing DLS data (hydrodynamic size) for magnetic particles, the results can be questionable [74]. This is partly due to the inaccuracy for using DLS technique in measuring the hydrodynamic size of dark materials/particles. Moreover, we cannot rule out the effect of aggregation of particles. Figure 3e showed the FITR spectra of the particles, highlighting the PMAA coating. To test the selective binding properties of ERY-MMIP particles, three experiments were carried out. Figure 5a shows the binding capacity of ERY-MMIP particles against the concentration of erythromycin, with a plateau at

around 70 mg of erythromycin per g of ERY-MMIP is shown at around 40 – 60 mg/mL. In a kinetic study shown in Figure 4b, the binding of erythromycin achieved the maximum after 40 mins. The selectivity study shown in Figure 4c highlighted the ERY-MMIP particles with a high selectivity towards erythromycin over other three antibiotics. A high selectivity factor (SF) of > 11.1 was recorded for all three competitors while the non-imprinted MNIP showed an SF around 2. These figures are comparable with the non-magnetic MIP for other pharmaceutical pollutants reported in the literature [75, 76]. The same technology should be applicable to prepare MMIP particles for many other organic pollutants, suitable for their capture and monitoring.



**Figure 4.** Characterization of ERY-MMIP particles. (a) TEM images of ERY-MMIP particles and (b) zoom-in TEM image of an ERY-MMIP particle to depict the embedded iron oxide magnetic nanoparticle (MNP) component, shown as darker particles. (c) Hydrodynamic size distribution analyzed of ERY-MMIP using DLS. (d) A image depicted the separation of ERY-MMIP particles before and after magnetic separation. (e) FTIR spectra of ERY-MMIP compared to vinyl-MNPs.



**Figure 5.** Binding capacities of ERY-MMIP particles. (a) Binding capacity of ERY-MMIP against various concentration of ERY after 40 minutes; (b) binding capacity of ERY-MMIP over time at 40 mg/mL of ERY; the grey line represents the binding from non-imprinted MNIP particle; (c) selectivity binding of ERY-MMIP on other antibiotics (grey bars) compared with non-imprinted particles (white bars).

## FURTHER CONSIDERATIONS

Although MMIP enhances recycling and reuse of MIPs and prolongs their shelf life, eventually the spent MMIPs need to be disposed once the binding capacity has diminished. Problems with most MIPs reported in the literature are that they are synthesized with non-biodegradable polymers (PMMA, PS etc). They are also crosslinked to maintain a certain degree of rigidity to allow for shape recognition. However, crosslinked polymers are even harder to degrade than non-crosslinked polymers. The small size of MIP micro- or nano-particles could cause further issues upon disposal, leading to possible leakage to wastewater and eventually to the environment. Due to the small size of MIP particles, conventional water treatment plants may not be able to completely remove them from sewage, although the amount is small compared to most other pollutants.

One possible solution is to use biodegradable polymer instead. A recent example showed that an MIP nanoparticle system based on poly(lactide-co-glycolide) (PLGA) was prepared using biotin as the template [77]. This MIP nanoparticle sample was tested for binding biotin and biotinized BSA. A very high binding imprinting factor of 21.1 was recorded but no selectivity test was carried out. More importantly, a degradation test in PBS at 37°C showed a 54% mass loss after 30 days. Although the degradation is slow, the test conditions were mild and could be sped up using an enzyme. In another example in this area, a poly( $\epsilon$ -caprolactone) or PCL based MIP was prepared using theophylline as a template, showing a 6.1  $\mu\text{mol/g}$  (1.1 mg/g) [78]. This MIP was tested against caffeine for selectivity, with a selectivity factor of 2.55. More importantly, the MIP was submitted for degradation in 1M NaOH and 90% degradation was recorded in 24 h. However, a much slower (10% over 30 days) was recorded in neutral PBS. Since the polymer component of MMIP in most studies is still PMAA based non-degradable polymer, combining magnetic and (bio)degradable properties has still not been seen in MIPs. This could be a new direction for MMIP development.

Another possible direction of development could be the multi-target capability of MMIPs, such as selective binding and detection for more than one chemical/contaminant. This is due to the ever increase in the complexity in formulation of additives used in food industries. Currently, this can be achieved by a mixture of a number of (M)MIPs for binding of multiple targets but those so called “chimera (M)MIPs” could help to simplify the system. However, applications of MMIP are still limited to laboratory scale research because the preparation includes homogenizing the reaction mixtures. Scaling up this step is still an obstacle. Therefore, use of MMIP for removing contaminant is likely to be associated with a high operational cost. This issue is less prominent if the use of MMIP is of a small amount, such as in assembling sensors, or the targeted molecules to be recovered or isolated are of high values (e.g. essential drugs or noble metals). We anticipate a rapid expansion in the application of MMIPs in a broad range of environmental applications.

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