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Selective extraction of triazine herbicides based on a combination of membrane assisted solvent extraction and molecularly imprinted solid phase extraction

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ABSTRACT

A selective extraction technique based on the combination of membrane assisted solvent extraction and molecularly imprinted solid phase extraction for triazine herbicides in food samples was developed. Simazine, atrazine, prometon, terbumeton, terbuthylazine and prometryn were extracted from aqueous food samples into a hydrophobic polypropylene membrane bag containing 1000 μL of toluene as the acceptor phase along with 100 mg of MIP particles. In the acceptor phase, the compounds were re-extracted onto MIP particles. The extraction technique was optimised for the type of organic acceptor solvent, amount of molecularly imprinted polymers particles in the organic acceptor phase, extraction time and addition of salt. Toluene as the acceptor phase was found to give higher triazine binding onto MIP particles compared to hexane and cyclohexane. Extraction time of 120 min and 100 mg of MIP were found to be optimum parameters. Addition of salt increased the extraction efficiency for more polar triazines. The selectivity of the technique was demonstrated by extracting spiked cow pea and corn extracts where clean chromatograms were obtained compared to only membrane assisted solvent extraction or only molecularly imprinted solid phase extraction. The study revealed that this combination may be a simple way of selectively extracting compounds in complex samples.

1. Introduction

Sample preparation and clean up steps are of paramount importance prior to analysis of triazines in plant materials because of the many interferences found in such samples. One such technique for sample preparation is liquid–liquid extraction (LLE) which is now less popular because of its drawbacks of being time consuming, not easy to automate and consuming large quantities of organic solvents [1]. Other alternative sample preparation techniques for aqueous samples are solid-phase extraction (SPE) [2], solid-phase microextraction (SPME) [3], stir bar sorptive extraction [4] and membrane extraction [5–7].

Membrane based extraction [5–7] techniques and techniques using selective sorbents such as molecularly imprinted polymers (MIPs) [8,9] in solid-phase extraction are attractive for plant samples for a number of reasons. In membrane extraction, because the membrane is nonpolar, any polar or charged matrices are prevented from diffusing to the acceptor side. Further, neutral macromolecules have slow mass transfer across membrane and in somes cases, depending on the pore size, may be excluded altogether. Membrane based extractions also use very little organic solvents and are in most cases cheap and simple to use.

MIPs are known to be much more selective than other SPE media since analyte extraction is based on the size, shape and structure [8,9]. A number of researchers have therefore used MIP based sorbents for selective extraction of organic compounds from various complex samples giving desired selectivity [9–12]. In some cases, the use of MIP sorbents [13] alone may not give the desired selectivity for plant materials because of the complexity of such samples. Thus, a two step extraction approach was reported by Cacho et al. [13], in which the plant materials were first extracted on the non-imprinted polymer (NISPE) followed by on a molecularly imprinted polymer sorbent (MISPE).

A combination of supported liquid membrane extraction (SLM) and molecularly imprinted polymers has been reported by Mhaka et al. [14], in trying to increase the selectivity in extracting plant materials. In the work of Mhaka et al. [14], MIPs were incorporated as part of the acceptor phase that contained toluene as a solvent. The solvent was also impregnated in the pores of a hydrophobic flat sheet membrane that separated the acceptor and donor phases. The combination resulted into good selectivity compared to SLM extraction alone or SLM-NIP combination. However, no comparison

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between the MISPE and SLM-MIP combination was made. Further, the set-up was designed in such a way that only the bottom part of the membrane was in contact with the stirred sample. This limited the mass transfer of the compounds from the sample into the acceptor side. After extraction, the separation of the MIP particles from the rest of organic solvent was performed by passing the entire extract into a 0.1 μm syringe filter where the particles were retained. This was followed by subsequent washing and elution of the trapped analytes. This procedure of separating the MIP particles from the bulk acceptor solvent was found to be not very efficient.

In order to minimise some of the problems experienced in SLM-MIP combination, instead a combination of membrane assisted solvent extraction (MASE) and molecularly imprinted polymers is proposed. The MASE technique [7,15–19] which involves a dense polypropylene bag is ideal for incorporating MIP particles as part of the acceptor solution. Since it is in the form of a bag, the mass transfer of analytes from the sample is not limited to only one direction. Instead of passing MIP particles and bulk acceptor solvent through a 0.1 μm syringe filter in order to separate the two as reported by Mhaka et al. [14], a convenient way using empty cartridges and solid phase extraction unit was used. The MASE–MISPE combination was tested by extracting cowpea and baby corn plant materials.

2. Experimental

2.1. Chemicals

Simazine, atrazine, prometon, terbumeton, terbuthylazine and prometryn were purchased from Sigma–Aldrich (Darmstadt, Germany). Organic solvents were also from Sigma–Aldrich. All other chemicals used were of analytical grade.

2.2. HPLC of triazines

The HPLC system used was from Hewlett Packard (model 1050, Palo Alto, CA, USA). It incorporated an autosampler set to an injection volume of 5 μL and a UV detector set to 230 nm for detection of triazines. Agilent Chemstation software was used for acquiring of the data. A C_{18} Hypersil column (2.1 mm × 150 mm, 5 μm) from Supelco (Darmstadt, Germany) was used. The mobile phase was composed of 30% acetonitrile and 70% water pumped with a flow rate of 0.2 mL min^{-1}. A stock solution of each triazine was prepared in acetonitrile at a concentration of 1 g L^{-1}. From this a working stock solution consisting of 300 mg L^{-1} of each triazine as mixture was prepared. External calibration was made with samples of 100, 250, 500 and 2000 μg L^{-1} of each triazine mixture.

2.3. MIPs, membrane bags and other accessories

MIP particles for triazines were supplied by MIP Technologies AB (Lund, Sweden) along with NIP particles (both are part of the ExploraSep™ screening library, the MIP is designated A31 and the NIP is designated A32). Empty 3 mL cartridges with frits 10 μm, were from Sorbent AB (Frölunda, Sweden). Membrane bags and their accessories were supplied by Gerstel (Mülheim, Germany).

2.4. MASE preparation

The MASE device has been described in previous publications [15–19]. The membrane extraction cell consisted of a 20 mL headspace vial filled with 18 mL of aqueous sample. The membrane bag (4 cm long, 0.03 mm wall thickness, 6 mm internal diameter) was attached to a metal funnel and fixed with a PTFE ring. The material of the membrane bag is dense polypropylene. Before extraction, the membrane bags with their metal cylinders were preconditioned in 40 mL cyclohexane by shaking overnight at 160 rpm. After 3 h, the cyclohexane was replaced with fresh cyclohexane. The extraction cell caps were preconditioned by shaking them in cyclohexane for 3 h. Thereafter, both the caps and membrane bags were dried at room temperature. The membrane bags were dried by putting them upside down through clean Pasteur pipettes. The same glass tubes were used to make sure that the membrane bags were filled with cyclohexane during the preconditioning stage. When cyclohexane was not used as the organic acceptor solvent in the extraction, the dried membrane bags were soaked in the respective solvent for about 2 h and dried again before extraction. The assembled membrane bag was tested for any leakages at the joints by pipetting inside 1000 μL of extraction solvent. The membrane bag with organic solvent was placed inside the extraction cell containing the aqueous sample and stirrer (MASE only) or MIP particles added into the organic acceptor solvent (MASE–MISPE combination). Any organic solvent on the outside of the bag was wiped with a tissue before putting the bag in the sample.

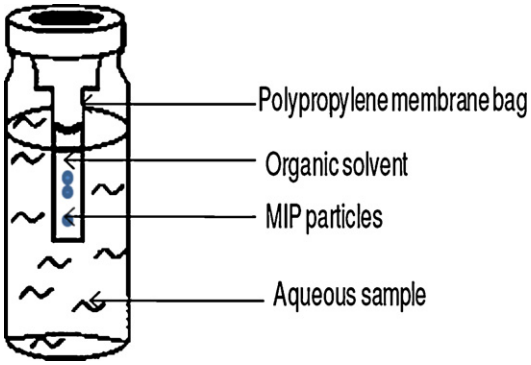

Fig. 1. Schematic representation of a MASE-MIP combination.

2.5. Extraction procedure with MASE–MISPE technique

100 mg of MIP particles was placed inside the membrane bag filling about two thirds of the volume followed by 1000 μL of toluene. The membrane bag was then compressed with clean gloves so as to mix the organic solvent with MIP particles. The membrane bag was placed in the extraction cell containing 18 mL of aqueous sample saturated with sodium chloride and stirrer (Fig. 1). The extraction proceeded for 120 min. At least three parallel extractions were performed simultaneously.

After extraction, the acceptor content was transferred into a 3 mL empty cartridge with a filter at the bottom and mounted onto solid phase extraction unit. As the bags are quite stiff, they can easily be handled manually and easily be rinsed with fresh 1000 μL toluene for quantitative transfer of MIP particles onto the cartridge. The outside of the membrane bag was rinsed with deionised water to remove any salts while wet. Toluene was separated from MIP particles by opening the SPE valve slowly and allowing it to flow out by gravity at about 0.5 mL min^{-1}. The membrane bag was then washed with 1000 μL of dichloromethane which was also passed into the cartridge containing the MIP particles. Thereafter, a full vacuum was applied for 2 min. The trapped analytes were eluted with 3× 1000 μL fractions of methanol. The first two portions of methanol were also used to rinse the membrane bag for any remaining MIP particles and then transferred into the cartridge. Between elutions, methanol portions were allowed to pass completely through the cartridge. The combined extracts were either analysed directly or reduced to almost dryness with gentle stream of nitrogen and then made up with 500 μL of methanol.

The used membrane bags, still attached to the metal funnels were then soaked into about 40 mL of acetonitrile or in combi-

nation of acetonitrile and water (50:50) (i.e. after extraction an aqueous sample saturated with sodium chloride) for about 3 h. They were then dried and left soaked in about 40 mL of cyclohexane overnight. In between, old cyclohexane was exchanged for a fresh one as described during preconditioning. The membrane bags were re-used during the entire optimisation process since pure water samples were extracted.

In the MASE procedure, the same steps above were followed except that the acceptor contained only toluene. A comparison was also performed where the acceptor solvent after MASE were passed through a cartridge packed with 100 mg of MIP particles. The cartridge was first conditioned with 1500 μL of dichloromethane and without allowing the cartridge to dry; the organic acceptor was passed through in the same way as described above. The membrane bag was rinsed with fresh 1000 μL toluene and passed through the cartridge. The same procedure for washing and elution was followed as described already.

2.6. MISPE of triazines

The MISPE extraction procedure was adapted from the SupelMIP™ SPE Triazine 10 instruction sheet downloaded from the Sigma–Aldrich website (http://www.sigmaaldrich.com). In brief, the empty cartridges with filters at the bottom were filled with 75 mg of MIP particles. Another filter was placed with help of a glass rod on top of the MIP particles to hold them in place. MIP particles were conditioned with 1000 μL of methanol followed by 1000 μL of ultra pure water and 1000 μL of 25 mM ammonium phosphate ($NH_4H_2PO_4$) buffer at pH 3. Conditioning was done without allowing the cartridge to dry and then 18 mL of aqueous sample was passed at about 0.5 mL min^{-1}. The washing step was performed by passing 1000 μL of 0.1 M HCl followed by 1000 μL of ultra pure water. A gentle vacuum was applied between each step followed by a full vacuum for 20 min to remove any residual moisture from the cartridge. 1500 μL of dichloromethane was then applied as a washing step. A full vacuum was applied for 2 min to remove residual solvent. Elution was performed with 3× 1000 μL methanol at about 0.2 mL min^{-1}. A gentle vacuum was applied between each fraction. The selective cavities of the MIP contain weak ion exchange moieties that can interact with the various triazine candidates. Extraction efficiency (E) was calculated as described previously [5,20] and was used to measure the performance of the MASE–MISPE combination, MASE only and MISPE only.

2.7. MASE–MISPE technique extraction of food samples

Maize baby corn (River Kwai Brand, Thailand) and cow peas (Everest Enterprise LTD, Kenya) were randomly bought from a local supermarket in Lund, Sweden. Both samples were crushed and homogenised with pestle and mortar. To 60 g of each wet sample, 120 mL of acetonitrile was added. The mixture was shaken at room temperature for 24 h at 160 rpm. Thereafter, it was filtered with through a Munktell filter paper (No. 3). The filtrate was left to evaporate to dryness at room temperature. Thereafter, 120 mL of deionised water was added and this was shaken for 3 h at 160 rpm to completely dissolve any matrix residues. The extracts were filtered once more with a Munktell filter paper (No. 3) followed by 0.45 μm Whatman membrane filter. The filtrate was made up to 300 mL of deionised water. pH of the extracts was measured at about 5.5. The extracts were diluted three times with deionised water before extraction. The extracts could also be processed without any of this dilution but it could mean that membrane bags will require more washing before being re-used. Samples for extraction with MASE and MASE–MISPE were saturated with sodium chloride while those for MISPE were extracted without any further processing. In the extraction of cow peas extract with MISPE, a washing volume of 5000 μL dichloromethane was compared to that of 1500 μL as recommended.

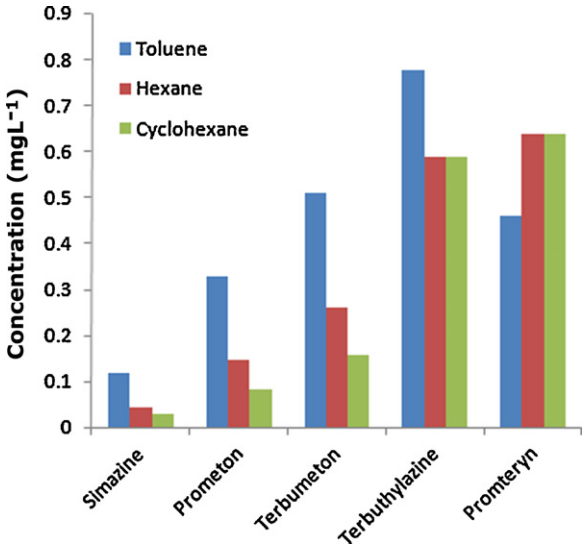

Fig. 2. Variation of the organic acceptor solvent with 50 mg MIP and 60 min extraction time. 0.6 mg L^{-1} each mixture of triazines extracted in the MASE-MIP combination.

Blank and spiked samples were extracted for each of the three techniques. The spiking concentration in the samples was 50 μg L^{-1} of each of the triazine mixture except for the cowpea extract where 200 μg L^{-1} was used. The extraction procedures for the three techniques is as described already except in the MASE and MASE–MISPE combination where the membrane bags were rinsed on the outside with deionised immediately after extraction to remove matrix components on the surface while wet.

3. Results and discussion

3.1. Optimization of extraction conditions

3.1.1. Variation of the type of organic acceptor solvent

Three solvents were screened as possible acceptor solvents. A good solvent should give high partitioning coefficient of the analytes from the membrane bag into the bulk of the acceptor solution, but also not interfere with binding of the analytes onto the MIP particles. The results shown in Fig. 2 indicate that more triazines were extracted with toluene as solvent. The partition coefficients of the triazines from water into three solvents were also measured using LLE followed by HPLC analysis of equilibrium concentration. The variation in partition coefficients of the analytes was found to be consistent with results shown in Fig. 2. Van Pinxteren et al. [7], optimised the acceptor solvent in MASE for related compounds. Good extraction efficiency was achieved with solvents such as toluene, xylene, butyl acetate, ethyl acetate, chloroform, and diisopropyl ether. The results in Fig. 2 are therefore consistent with this finding. Further, in a previous study where LM-MIP combination was optimised for triazines [14], toluene was also found to give better extraction efficiency compared to hexane or combination of hexane with ethyl acetate. The effects of solvents on the selectivity and binding of triazines unto MIP have been studied in detail by Pap et al. [21]. The acceptor phase solvent should not disrupt the specific interaction between the triazines and MIP particles. Solvents with both low dielectric constants and hydrogen bonding capabilities are preferred.

Generally speaking, the present approach makes use of the fact that MIPs operate best under organic conditions. Typically, in a

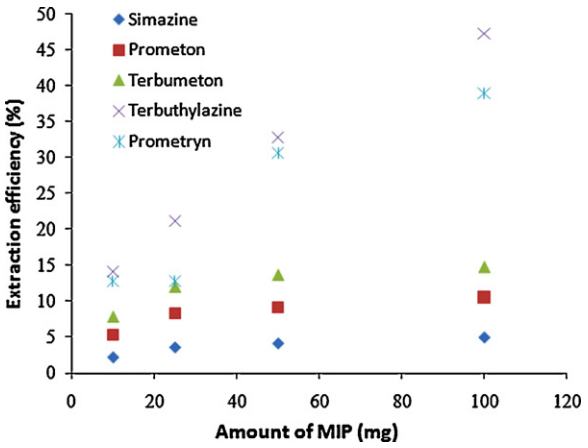

Fig. 3. Variation of the amount of MIP in the acceptor phase with toluene as organic solvent. 60 min extraction time and 0.6 mg L^{-1} each mixture of triazines extracted in the MASE-MIP combination.

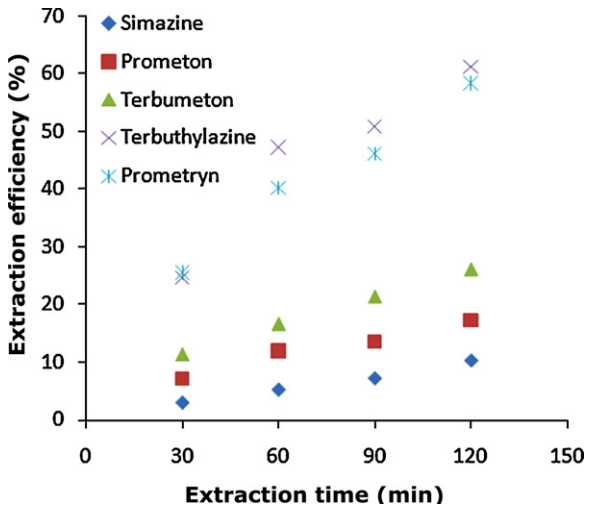

Fig. 4. Variation of the extraction time with toluene as organic solvent and 100 mg of MIP. 60 min extraction time and 0.6 mg L^{-1} each mixture of triazines extracted in the MASE-MIP combination.

MISPE procedure, the SPE column containing the MIP is conditioned and then the aqueous sample is loaded. In order to fully exploit the selective power of the MIP, a solvent switch step is conducted. This means that the aqueous residuals in the SPE column are dried by e.g. 10 min of vacuum at this stage. Subsequent to that an organic solvent is used to change the previously aqueous environment to an organic environment. Also there, solvents with a low dielectric constant and lack of protic properties are preferred. In this solvent switch step, the organic solvent removes any hydrophobically bound compounds and at the same time, maximises the selective polar interactions between analyte and the functional groups in the binding site. The MASE approach in combination with the resin, elegantly utilises this requirement of MIPs. In other words, the solvent switch is being taken care of the by present approach.

3.1.2. Variation of the amount of MIPs in the acceptor phase

Fig. 3 shows the results of the optimisation of the amount of MIP particles in the acceptor phase of the membrane bag. As expected, an increase in amount of MIP particles lead to an increase in bound triazine. This is also consistent with what Mhaka et al. [14], and Nemulenzi et al. [22], observed in the optimisation of MIP amounts in LM-MIP combination. However, in this case, the increase deviates from linearity except for terbuthylazine and prometryn which are the most non-polar triazines. The departure from linearity in Fig. 3 could be due to slow mass transfer of the analytes in diffusing through the membrane into the bulk of the acceptor solution. The concentration of the donor solution remaining after each extraction in Fig. 3 was measured and revealed that diffusion of the analytes from the sample into the membrane bag was not the rate limiting step but diffusion through the bag into the organic acceptor solvent. At high MIP amounts, not much gain in extraction efficiency was therefore realised except for more non-polar compounds.

3.1.3. Influence of extraction time on the extraction efficiency

The extraction efficiency was found to increase with extraction time (Fig. 4). This is expected because both diffusion of the analyte through the membrane bag into the bulk of the acceptor solution and the subsequent binding onto MIP particles are time dependant. After 2 h of extraction, there was no sign of the extraction efficiency reaching a plateau. In the LM-MIP combination reported by Mhaka et al. [14], the extraction efficiency showed signs of reaching a plateau after 60 min of extraction. This indicates that the diffusion of analytes in dense membranes is much slower compared to liquid membrane. This is expected since diffusion coefficients of analytes are higher in liquids compared to solids. The membrane bag however is much more stable compared to a liquid membrane. It is also easily re-usable. Hauser et al. [15] have varied the extraction time in MASE with similar or related compounds. An increase in the extraction time resulted in higher extraction yield of all compounds from 10 to 30 min. Further increase did not result in changes in extraction yield. The extraction temperature in this case was 55 °C. This suggests that the MASE–MISPE combination in future could be operated at a higher temperature than room temperature to reduce the extraction time.

3.1.4. Influence of the addition of salt to the sample

The addition of salt enhanced the mass transfer in the extraction process (Fig. 5). These results are consistent with other MASE optimisations where salt was added [7,18]. When salt is added, water molecules solvate around ions [7] leading to poor solubility of polar compounds. This enhances the dissolution of these compounds in the membrane bag. Since the diffusion of analytes into the membrane bag to the acceptor solvent was the rate limiting step, it does suggest that the addition of salt speeds up this process. For more non-polar compounds, the addition of salt slightly reduced the amount extracted. This could be due to increased movement to the water sample surface for these non-polar compounds also referred to as “oil effect” [19]. All further experiments were thus performed with sample saturated with salt.

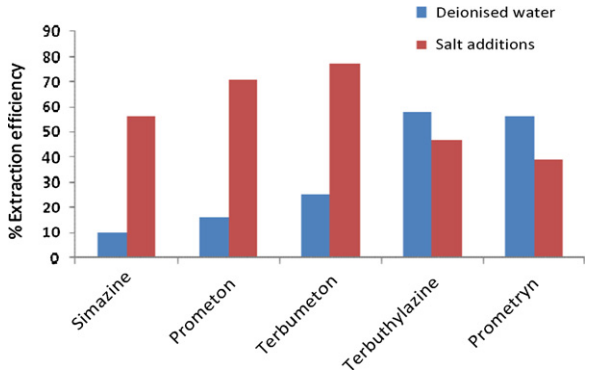

Fig. 5. Comparison of extraction efficiency with and without salt addition in the aqueous sample in MASE-MIP technique. 120 min extraction time, 100 mg of MIP particles and 1000 μL of toluene as acceptor phase.

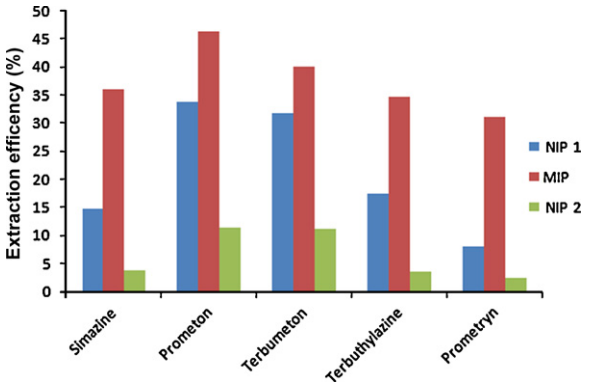

Fig. 6. Comparison of NIP and MIP extraction with toluene as organic solvent and 100 mg of particles. 60 min extraction time and 0.6 mg L^{-1} each mixture of triazines extracted in the MASE-MIP combination. NIP 1 and NIP 2 are after washing of 1 and 3 mL dichloromethane before elution with methanol, respectively.

3.1.5. Comparison of the NIP and MIP on the extraction efficiency

It is always important to compare the performance of the MIP particles to those of the NIP. This gives an idea whether the MIP particles are well prepared and selective for the target compounds. Fig. 6 shows such a comparison between MASE–NISPE and MASE–MISPE. The results indicate that the MIP particles were superior in binding the compounds compared to the NIP particles. This is consistent with similar comparisons where MIP particles are used as sorbents. In Fig. 6, NIP 2 means that the polymer was additionally washed before elution of the compounds as compared to NIP1 or MIP. Since the concentration was further reduced, it suggests that most of the compounds are trapped on the NIP particles through non-specific interaction. In LM-MIP combination by Mhaka et al. [14], the selectivity between the NIP and MIP was compared. The LM-NIP was found to be less selective compared to the LM-MIP supporting the idea of the non-specific interaction.

3.1.6. Precision and LODs

Fig. 7 compares the relative standard deviation obtained for extraction of triazines spiked at 50 μg L^{-1} each in cowpea and baby corn extracts. MISPE gave the lowest %RSD values with ranges of about 5–16 for baby corn extract. For MASE, RSD values ranged about 4–20% in cowpea extraction and between 2 and 14% in baby corn. For MASE–MISPE combination, the %RSD values ranged from 7 to 20 in cow pea extraction and 2–9 in baby corn. In general, cowpea extracts were therefore found to give high relative standard deviation regardless of the technique. This can be attributed to intense matrix components found in such samples. Considering the type of samples, the %RSD values despite being on the high side are acceptable. Zuin et al. [19], is reported to have developed a MASE method for extraction of pesticides and benzo[a]pyrene

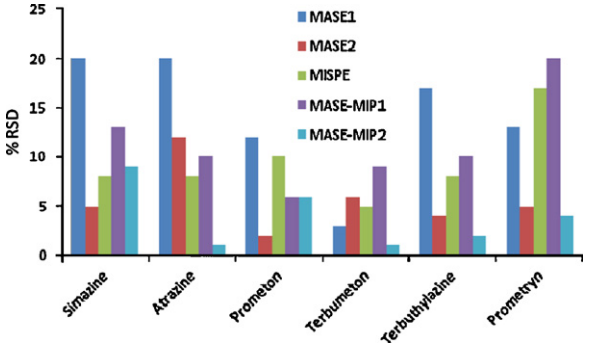

Fig. 7. Comparison of %RSD values in the extraction of cow pea extracts (MASE 1 and MASE-MIP 1) and baby corn extracts (MASE 2, MASE-MIP 2 and MISPE).

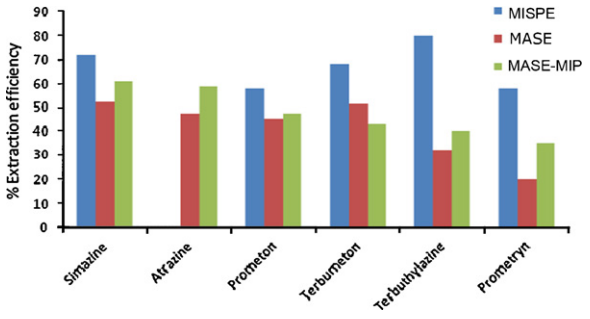

Fig. 8. Comparison of extraction efficiency of the three extraction techniques in the extraction of cow pea extract. Conditions: 120 min extraction time for both MASE and MASE-MIP, 100 mg of MIP particles and 1000 L of toluene as acceptor phase for MASE-MIP. 50 g L^{-1} mixture of each triazine was extracted. Note that membrane bags were re-used for 10 times and could account for slightly lower %*E* values in MASE and MASE-MIP combination.

residues in Brazilian sugarcane juice. The RSD values of the developed method ranged from 3.5 to 17.1%. The values reported here in the MASE–MISPE combination are not very different from those reported in MASE method only by Zuin et al. [19]. The limit of detection for MASE and MASE–MISPE was very similar except for prometon where it was twice higher in the MASE method. This was due to an interfering peak. The mean limit of detection (μg kg^{-1}) was found to be about 3.3 for MASE, 1.3 for MISPE and 3.0 for MASE–MISPE combination. MISPE of baby corn extracts had generally lower limit of detection compared to other methods due to higher extraction efficiency.

3.1.7. MASE–MISPE technique extraction of food samples

The extraction efficiency obtained during extraction of cowpea and baby corn plant materials using the three methods is shown in Figs. 8 and 9, respectively. The extraction efficiency was superior in MISPE compared to MASE or MASE–MISPE combination. This difference was more pronounced in the extraction of baby corn extracts which seemed to have less matrix components compared to the cowpea. In extraction of baby corn plant materials, old membrane bags that have been re-used about 10 times were used. This may have contributed to lower extraction efficiency in both MASE and MASE–MISPE combination shown in Fig. 9. The extraction efficiency obtained in MASE–MISPE combination was limited by analytes diffusing into the bulk acceptor solution which is why the extraction efficiency in both MASE and MASE–MISPE combination were similarly low. A comparison was also made where MASE was performed and extracts passed through cartridges packed by MIP particles as sorbent. The extraction efficiency obtained was similar to the MASE–MISPE combination. The obtained extraction

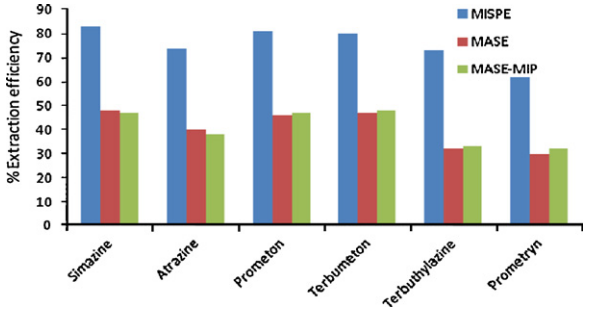

Fig. 9. Comparison of extraction efficiency of the three extraction techniques in the extraction of baby corn extract. Conditions: 120 min extraction time for both MASE and MASE-MIP, 100 mg of MIP particles and 1000 μL of toluene as acceptor phase for MASE-MIP. 50 μg L^{-1} mixture of each triazine was extracted. Note: membrane bags were re-used for 10 times and could account for slightly lower %*E* values in MASE and MASE-MISPE combination.

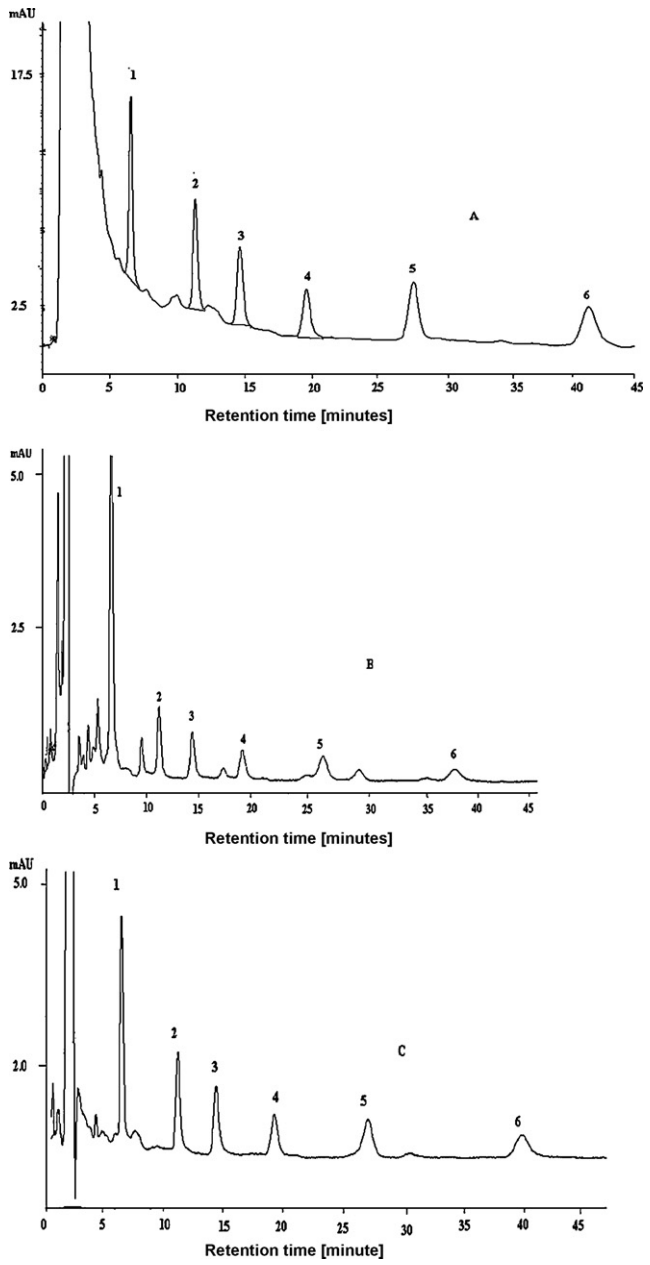

Fig. 10. Chromatograms obtained after extraction of baby corn extract spiked with 50 μg L^{-1} mixture of each triazine with MISPE (a), MASE (b) and MASE-MIP (c) combination. 100 mg of MIP with toluene as acceptor solution with 120 min extraction time was used. 1 = simazine, 2 = atrazine, 3 = prometon, 4 = terbumeton, 5 = terbuthylazine, 6 = prometryn.

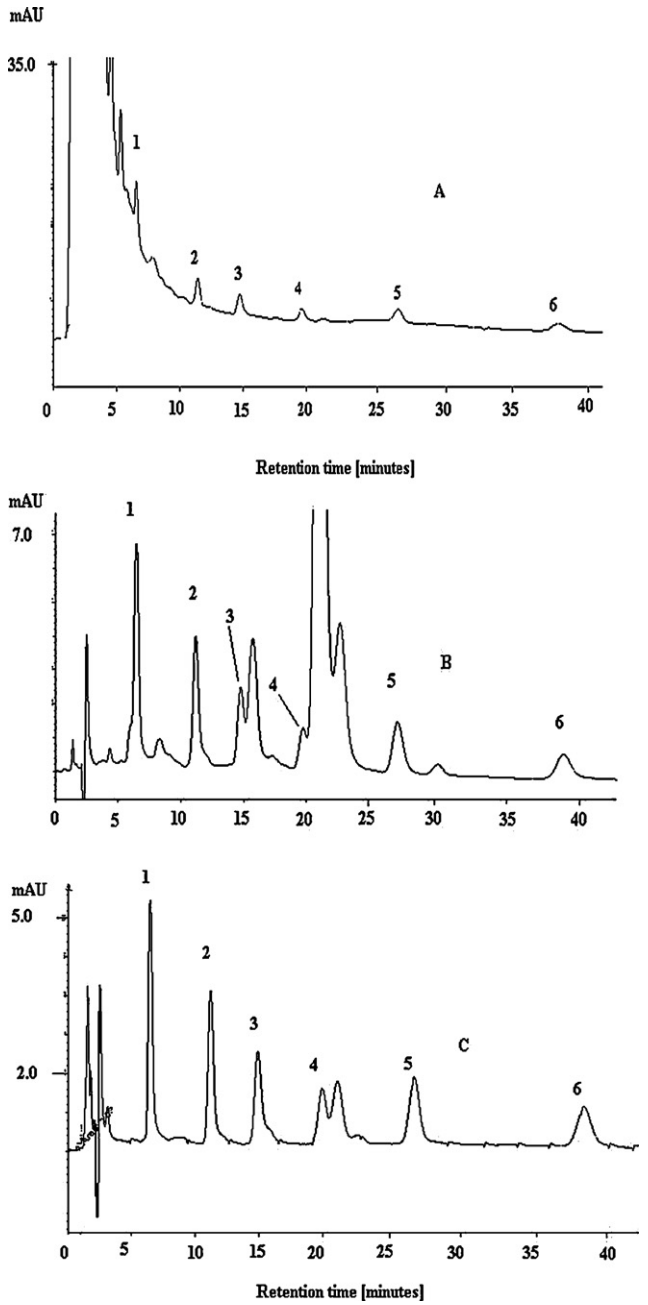

Fig. 11. Chromatograms obtained after extraction of cow pea extract spiked with 50 μg L^{-1} mixture of each triazine with MISPE (a), MASE (b) and MASE-MIP combination (c). Other conditions are as in Fig. 10.

efficiency for the triazines in MASE and MASE–MISPE combination are comparable to what has been reported by Van Pinxteren et al. [7], in MASE.

The selectivity of the MASE–MISPE technique was demonstrated by extracting spiked food samples, and analyzing the final extracts by HPLC. The chromatograms are shown in Figs. 10 and 11 for baby corn and cowpea extracts, respectively. The MASE–MISPE combination gave superior selectivity in both cases. This was more pronounced in cowpea extracts where the matrix components were intense. The huge peak coming out with the solvent front was eliminated and very little other unwanted peaks were seen in the rest of the chromatograms. The MASE also removed the huge peak coming out with the solvent front which is expected but failed to remove other unwanted small peaks in the rest of the chromatograms. The resulting selectivity in MASE–MISPE combination is not surprising. This is because the combination prevents many interfering compounds reaching the MIP particles. It can be seen as "a prevention is better than cure approach" in sample preparation. In MISPE approach, the matrix components are allowed to come into contact and bind with sorbent which are later washed out before elution. The washing step may not remove all these matrix components. In such a case, the proposed approach may offer the solution. Since few other unwanted components bind onto MIP particles in the proposed combination, it allows the sorbent to easily be regenerated and re-used much easier. Cacho et al. [13], reported to have developed a two step extraction method in the extraction of triazines from potato, corn and peas extracts. A one step MISPE did not give very clean extracts. Thus a two step non-imprinted polymer (NP) and molecularly imprinted polymer solid-phase extraction (MISPE) was developed [13].

4. Conclusion

The potential of the MASE and MISPE combination has been demonstrated for extraction of plant extracts. The combination has great potential in extraction of complex samples because of its high selectivity. In a combination, most marix components are prevented from binding onto the MIP particles because they have to cross the membrane barrier. This "prevention is better than cure approach" may be an alternative for extraction of very complex samples.

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