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Engineering Core/hollow Shell Nanomaterials to Load Herbicide Active Ingredient for Controlled Release

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ABSTRACT

In rainfed agriculture, besides moisture stress, weed infestation is considered to be one of the most important limiting factors of crop production. Lack of moisture at the appropriate time restricts the use of herbicides existing, necessitates the need for new formulations to release the active ingredient in a controlled manner based on the soil moisture status. Manganese carbonate (MnCO_3) core-shell nanoparticles were synthesised using wet chemical methods for loading herbicide active ingredients. Manganese and calcium carbonate were originally selected for core material synthesis. Since, CaCO_3 results in the formation crystals against spherical structure; MnCO_3 was selected for the study. The core material was coated with suitable water soluble polymers such as sodium Poly Styrene Sulfonate (PSS) and Poly Allylamine Hydrochloride (PAH) Layer by Layer (LbL) method to obtain a water-soluble (for rainfed systems) core-shell particles. By using suitable solvents MnCO_3 core materials were etched out and results in the formation of hollow-shell particles. The hollow-shell particles were loaded with herbicide pendimethalin using passive method. Thus, synthesised core-shell, hollow-shell and loaded particles were characterized using TEM, FE-SEM, EDX, XRD, particle size analyzer, TGA and GC for size determination and loading efficiency.

Key words: Nanomaterial, core-shell, hollow-shell, polymer, nanoherbicide, controlled release

INTRODUCTION

Agricultural production in the rainfed areas depends on various factors which interact either to enhance output or to limit production. Among the factors limiting the production, weed ranks top. Under rainfed condition water is the most important resource which decides the success or failure of the crop. Presence of weeds with well-developed root system and more efficient in extracting moisture, become threat to crop production in the rainfed areas.

Employing manual or mechanical control of weeds besides adding additional cost it takes time to cover larger area by that time damage might have reached unrecoverable stage (Ghanizadeh *et al.*, 2010). Alternatively chemical methods integrated with cultural practices proved to be the best option in rainfed areas (Awan *et al.*, 2002). However, lack of sufficient moisture, limits the choice of chemical weed management. Due to lack of moisture in the top layer of the soil, the weed seeds present, unable to germinate during initial growth stages of crop, subsequent rain

favour the germination of weeds which will become too late to choose a herbicide. Further, herbicide application with insufficient soil moisture may lead to loss due to photolysis and volatilization. Still, we are unable to predict the rainfall precociously; herbicides cannot be applied in advance anticipating rainfall. With this background a research project has been formulated to develop a nano-encapsulated herbicide, programmed to release as soon as the rain begins during the course of crop growth. The results of project are abridged hereunder.

The field of nanotechnology opens up novel applications in agriculture (Speiser, 2008; Lai *et al.*, 2006). Replacement of conventional agrochemical formulations by controlled-release systems not only helps to avoid treatment with excess amounts of active substances, but also offers, besides ecologic and economic advantages, the most suitable technical solution to a specific situation. To improve the existing weed management techniques development of nanodevices, a smart delivery systems and nanocarriers for controlled release was developed to avoid wastage and better utilization of applied herbicides. Strong evidences encourage the use of nanoparticles to protect the encapsulated active ingredients from degradation and control delivery to a specific situation (Florence *et al.*, 1995; Takeuchi *et al.*, 2001). Several methods are employed to prepare polymeric encapsulation of active ingredients based on the needs and application (Reis *et al.*, 2006). Further the polymeric nanocapsules have been widely considered as drug delivery system to control the drug discharge from microparticles (Beck *et al.*, 2007; Couvreur *et al.*, 2002). With this background in order to achieve the smart release it has been programmed to synthesis core, core-shell and hollow-shell nanomaterials for loading herbicides. The details for fabrication of core, encapsulating the core with water soluble polymers (layer by layer) to make as hollow-shell particles and loading of herbicides were discussed in this study.

MATERIALS AND METHODS

Materials: Pendimethalin, Sodium poly (styrene sulfonate) (PSS, Mw ~70 kDa) and poly (allylamine hydrochloride) (PAH, Mw ~70 kDa) were purchased from Alfa Aesar. All other chemicals were of analytical grade and were used as received without any further purification. The water used in all the experiments was double-distilled.

Methods

Synthesis of MnCO_3 core and fabrication hollow-shell: Equal volume of 0.33 M ammonium bicarbonate (NH_4HCO_3) was mixed with equal volume of 0.33 M manganese sulphate monohydrate solution (MnSO_4). Equal volume of 0.5% ethanol solution was added and the resulting solution was vigorously stirred for 10 min. It was then left undisturbed for 10 min and incubated for 1 h in water bath at 50°C (Tong and Gao, 2007). The solution was centrifuged to settle down the particles completely and washed thrice with distilled water followed by centrifuging after each wash. Then the particle was isolated by filtering with filter paper Whatman No. 41.

MnCO_3 core-shell was prepared by adding Layer by Layer (LbL) adsorption of opposite charge polyelectrolyte onto the MnCO_3 nanosphere templates. The polyelectrolyte solution was prepared by taking each 20 mg of Poly (allylamine hydrochloride) and Sodium poly (styrene sulfonate) in 20 mL of 0.5 M NaCl at neutral reaction, respectively. Twenty milliliter of each polyelectrolyte solution was added to 40 mg of dry MnCO_3 microparticles and the suspension was gently stirred in magnetic stirrer for 15 min. Then the suspension was centrifuged at 1000 rpm for 15 min and rinsed for three times with 0.1 N NaCl to remove the unbounded particles. The same procedure was repeated with oppositely charged polyelectrolyte (Tong and Gao, 2007). After defined number of

polyelectrolyte layer reached, MnCO_3 core was removed by adding 0.1 M HCl and stirred for 15 min. Then the suspension was centrifuged and rinsed with distilled water for several times to remove unreacted HCL and kept for overnight drying (Antipov *et al.*, 2003; Zhu and McShane, 2005). The size and shape of core, core-shell and hollow-shell was monitored using particle size analyser, Transmission Electron Microscope (TEM), Field Emission Scanning Electron Microscope (FE-SEM).

Preparation of encapsulated herbicide: Took 20 mg of hollow-shell and added 25 mL of 20 ppm pendimethalin (1:1) and stirred it for 15 min in magnetic stirrer. Then the suspension was allowed to dry overnight. After drying, the encapsulated particles were washed with methanol to remove excess herbicide adhered outside the hollow-shell and again dried over night. Dried particles were collected and stored in a vial. The nanomaterials were subjected to SEM and EDAX examination at every step.

Sample extraction for GC analysis: The encapsulated nanoparticles were analysed for encapsulation efficiency. The pendimethalin (10 ppm in 10 mg) extracted sample was transferred to separating funnel and 30 mL of 5% aqueous NaCl was added. The content was partitioned with n-hexane passed through anhydrous sodium sulphate. Hexane layer was concentrated on rotary vacuum evaporator at 60°C to approximately 2 mL and quantified by gas chromatograph.

RESULTS AND DISCUSSION

The fabricated core, core-shell, hollow-shell and encapsulated herbicide nanoparticles were analysed using various instruments and the results are discussed here under.

MnCO_3 core material: MnCO_3 microparticle templates were prepared by mixing MnSO_4 and NH_4HCO_3 solutions. Ethanol was added to decrease the dielectric constant of the system and the solubility of the inorganic salts. This reaction results in the formation of modified porous surfaced MnCO_3 (Tong and Gao, 2007). The unique porous spherical superstructure can be seen from the FE-SEM image (Fig. 1) of MnCO_3 core particles. Similarly X- ray diffraction pattern of core particle (Fig. 4) confirms the formation of manganese carbonate spherical particles three characteristic peaks at an angle of (2 θ) 24°, 32° and 52° which matches with the standard pattern

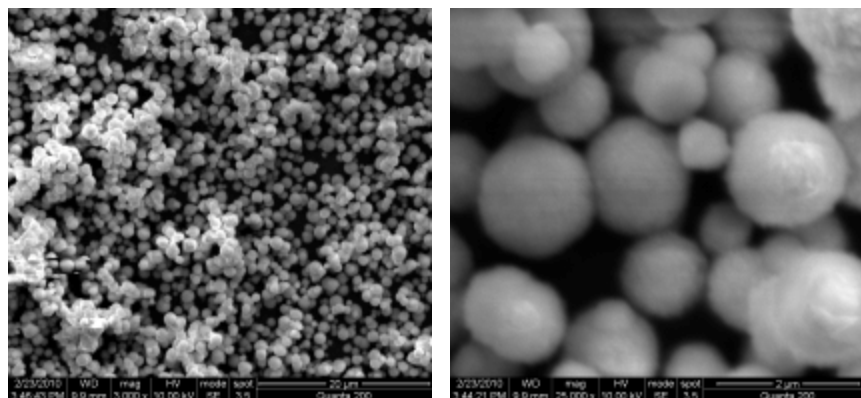


Fig. 1: FE-SEM image of MnCO_3 core particle

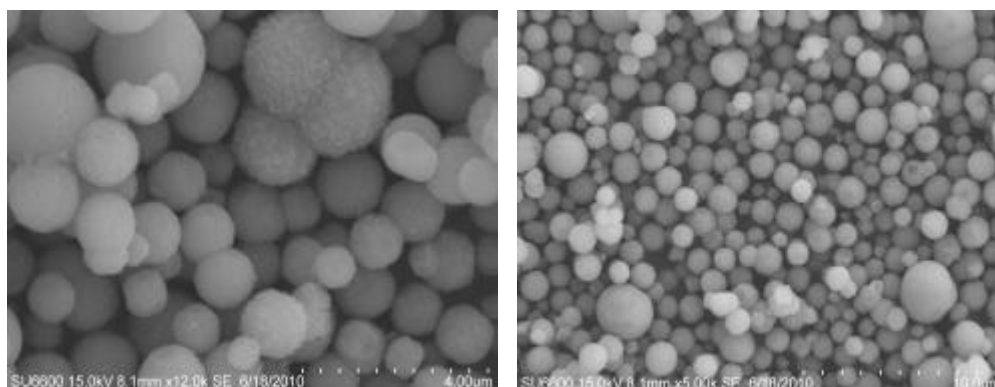


Fig. 2: FE-SEM image of core-shell particles

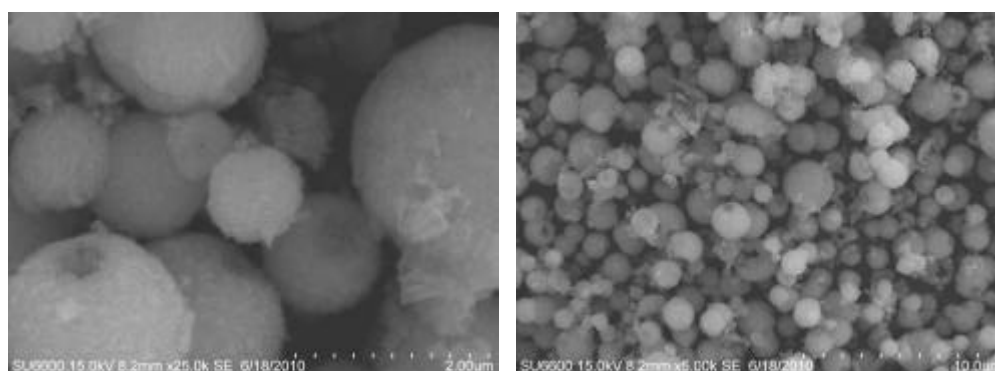


Fig. 3: FE-SEM of hollow-shell particles

of rhodochrosite (Tong and Gao, 2007). Also the particle size distribution and cumulative results of the MnCO_3 core particle from particle size analyser confirms it further (Fig. 6). The size of the core was seen to be 154.9 and 443.7 nm of diameter as 50 and 90% distribution which also confirms the formation of nanoparticles in spherical dimension.

MnCO_3 core-shell materials: Single bilayer of the polyelectrolytes (PAH and PSS) were coated onto the MnCO_3 core by Layer-by-Layer assembly method. The LbL assembly is the most widely used method to produce uniform spherical sized particles. Thus produced core-shell particles can be seen in the FE-SEM image (Fig. 2). The porous natured surface of MnCO_3 allows small molecules of PSS and PAH to penetrate inside through the pores during consecutive polyelectrolyte treatments and results in the formation of polyelectrolyte complex which remains stable after the core dissolution (Volodkin *et al.*, 2004). This open up the way to fabricate hollow microsphere with controlled wall thickness. The X- ray diffraction pattern of core-shell (MnCO_3 +single bilayer of polymers) (Fig. 4), shows the characteristic peaks at 2θ equal to 24, 32 and 52° which were seen in the MnCO_3 core too and also matches with the literature (Hamada *et al.*, 1987). At $2\theta = 32^\circ$, we can find a maximum intensity peak due to the polymers interface and this intense peak was not

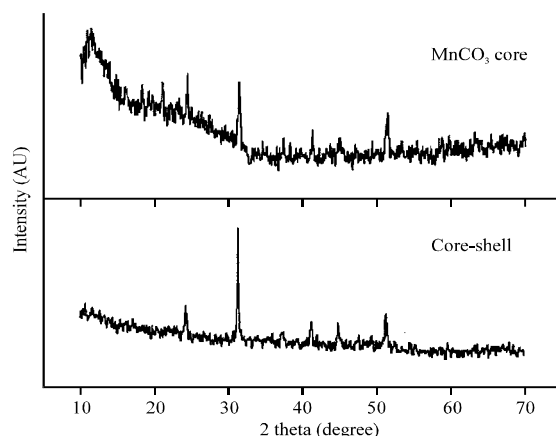


Fig. 4: XRD diffractogram of MnCO_3 core and core-shell particles

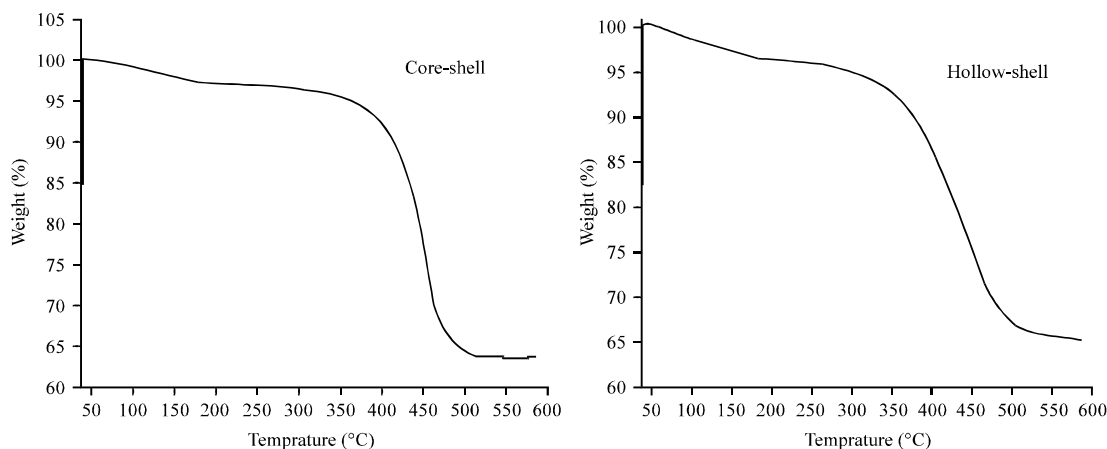


Fig. 5: TGA thermogram of core-shell and hollow-shell particles

found in core which confirms the coating of single bilayer of polyelectrolyte PAH and PSS onto manganese carbonate core particles. The TGA Thermogram of core-shell (Fig. 5) shows a single stage decomposition at 350°C which corresponds to the polymers coated. Also it was observed a high stable steady transition from the decomposition points and this confirms the formation of single bilayer of polyelectrolytes onto the core. From the particle size analyser we can clearly see the differences in the particle size distribution data of core and core-shell. The particle size distribution of the MnCO_3 core and core-shell were 126.4 and 250.5 nm, respectively. The size of core shell (Fig. 7) was 230 nm which clearly indicates the polyelectrolyte coatings of two linear polymers such as PAH and PSS onto the core.

Hollow-shell: Thus formed core-shell particles were made into hollow-shell using chemical etchants. After the incubation of core-shell particles in 0.1 M HCl for 10 min (Antipov *et al.*, 2003; Zhu and McShane, 2005), etching of MnCO_3 core occurs and results in the formation of hollow-shells which can be seen in the FE-SEM image (Fig. 3). The dissolution of core occurs due to the presence of the porous structures on the exterior surface which aids in the process. The acid

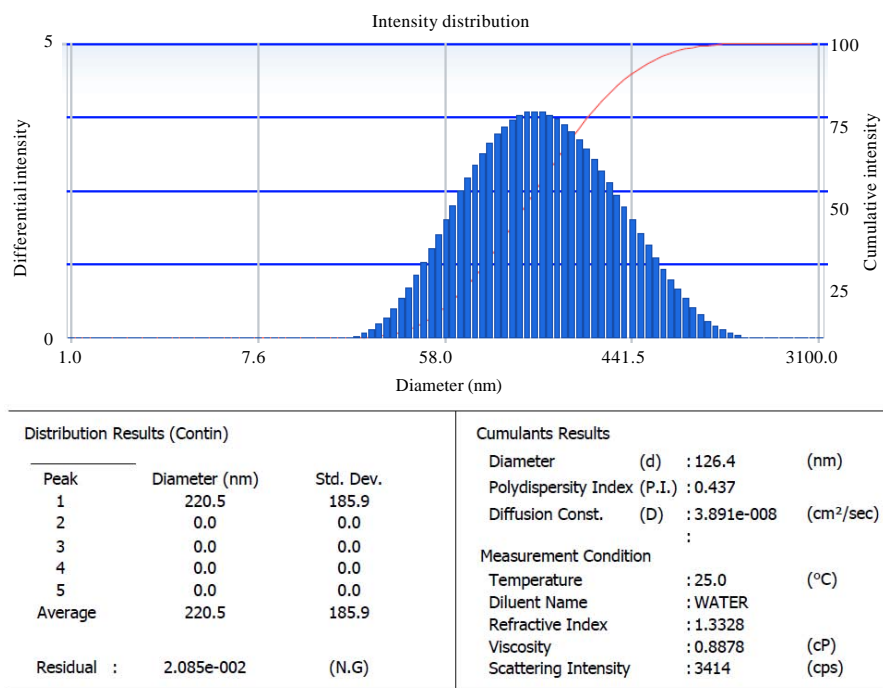


Fig. 6: Particle size analyzer intensity distribution and cumulative results of MnCO₃ core

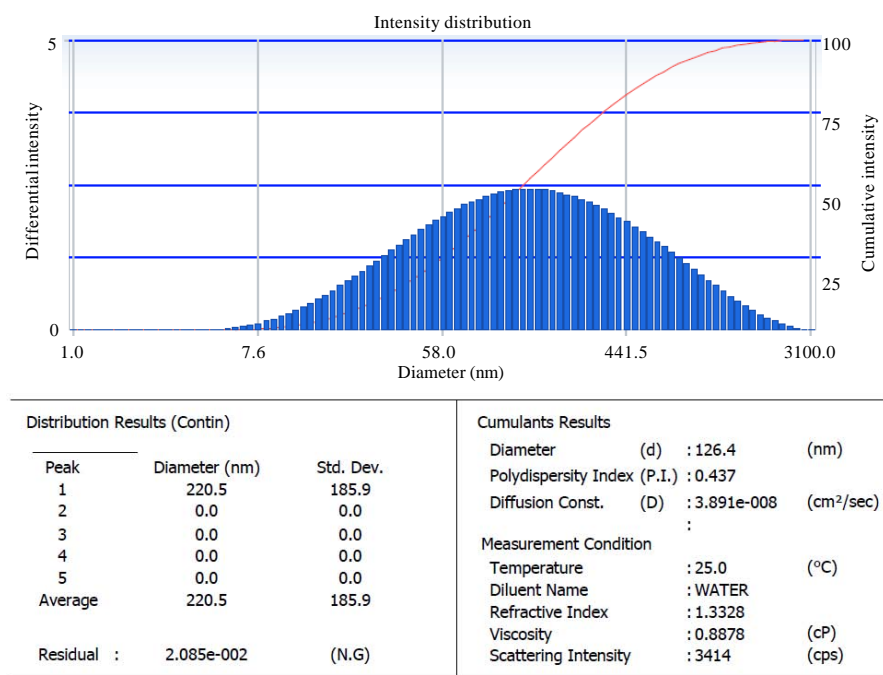


Fig. 7: Particle size analyzer intensity distribution and cumulative result of core-shell (MnCO₃+single bilayer of polymer)

concentration plays a major role in core dissolution process which dissolves the MnCO_3 particles and results in the change of particle size. This phenomenon is not dependant on the etching agent it is based on the core material used. Here the core material acts as a sacrificial template to obtain the polyelectrolyte capsule used for control release of any active ingredient. The FE-SEM image (Fig. 3) illustrates the fabrication of hollow shell. The size of the hollow shell was similar to the size of the template which can be clearly seen from the electron microscopic image. The hollow-shell was permeable to the external stimuli like pH, salt, temperature and organic solvent with tuneable physico-chemical properties (Kim and Choi, 2007). The formation of hollow-shell was also confirmed from the Thermo Gravimetric Analysis. From the Fig. 5, we can find a difference in the decomposition point and the onset point of weight percentage difference had shifted from 95 to 90% for core-shell and hollow-shell respectively. Also we find a slight decreased pattern in transition temperature of hollow-shell to 300°C instead of 350°C which confirms the formation of hollow-shell particles.

Encapsulation of herbicide: The active ingredient of pendimethalin herbicide was encapsulated into the hollow-shell by passive loading method. The pendimethalin was dissolved in the solvent and encapsulated into the hollow-shell by simple mixing and solvent evaporation process. Since the solvent treatment has effect on the microcapsules permeability due to the dielectric constant a small change in the structure occurs which is very small compared to the hollow formation. The hollow-shells then turned into closed one by the permeability of the polymer in the solvent which can be seen from the FE-SEM image (Fig. 8). To remove the sticking herbicide on the surface of the microspheres a suitable organic solvent was selected. Care has been taken for solvent selection as it should not dissolve the outer polymer layer. Also this solvent treatment increases the encapsulation efficiency of the herbicide due to the reaction between solvent and polyelectrolytes (Fig. 9) shows the encapsulated herbicide inside the hollow sphere which is partially closed due to electrostatic force of attraction. The SEM-EDX image (Fig. 10) shows the Carbon and Oxygen peak which represents the presence of active ingredient inside the shell and the trace amount of Mn peak was also seen in the EDX. The solvent washed encapsulated shell was washed again with water to know the releasing of encapsulated pre-emergence herbicide by dissolving water soluble polymer layer coated around the active ingredient. It was evident from the image of SEM and SEM-EDX

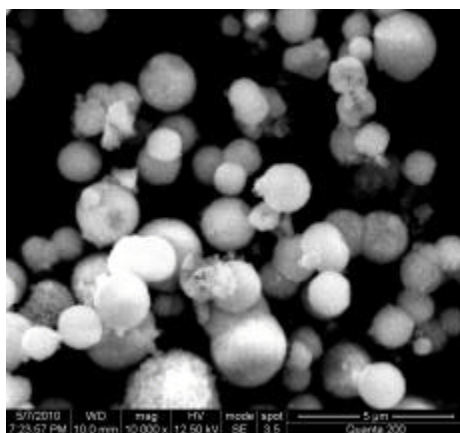


Fig. 8: FE-SEM image of encapsulated of pendimethalin active ingredient into hollow-shell

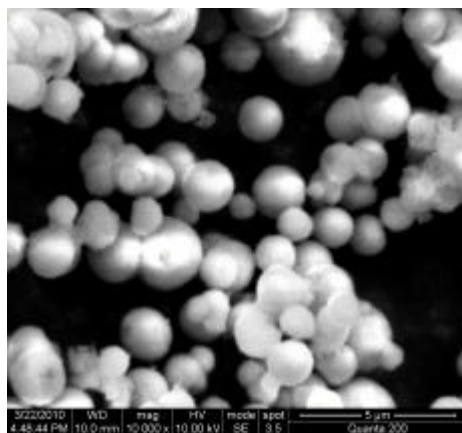


Fig. 9: FE-SEM image of encapsulated of pendimethalin active ingredient into hollow-shell after solvent wash

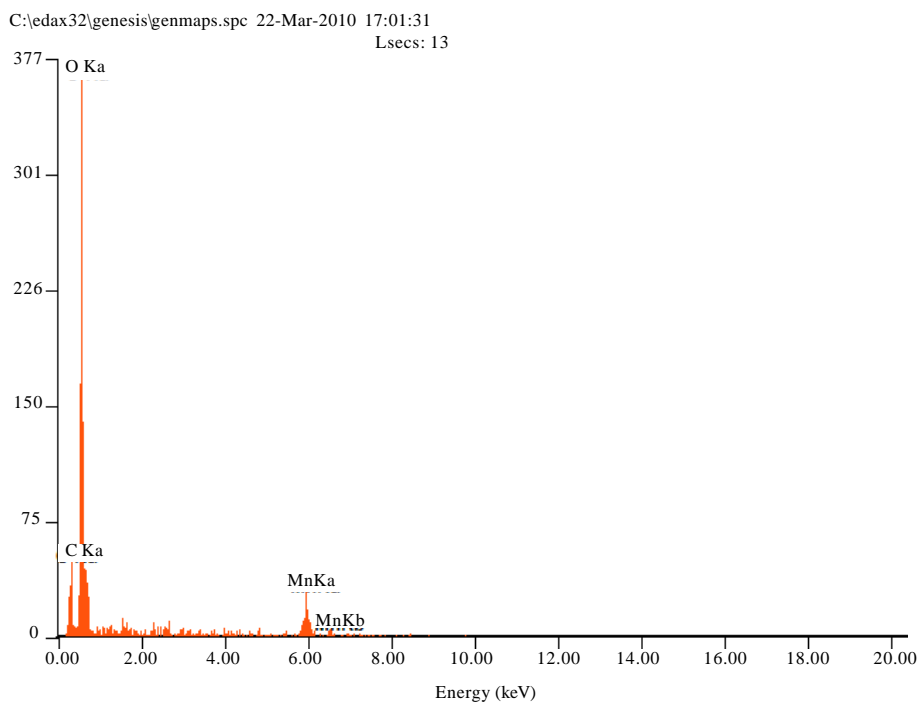


Fig. 10: EDX of Pendimethalin loaded hollow-shell

(Fig. 11, 12) that the polymers were dissolved when water was added and the herbicides encapsulated were released. This method looks similar with “Controlled release formulation of an herbicide, 2, 4-dichlorophenoxyacetate (2,4-D)” developed in the inorganic Zn-Al layered double hydroxide (ZAL) matrix, hosting an active agent by self-assembly technique Hussein *et al.* (2005). After loading with pendimethalin, small deviation in the peak intensity could be observed from the X-ray diffractogram of halo shell loaded with pendimethalin before wash (HSP1). Also the size

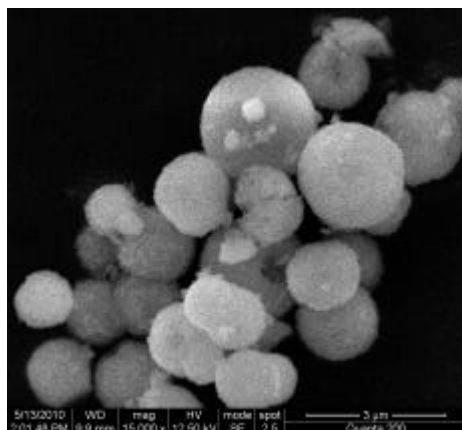


Fig. 11: FE-SEM image of Encapsulated of pendimethalin active ingredient into hollow-shell after water wash

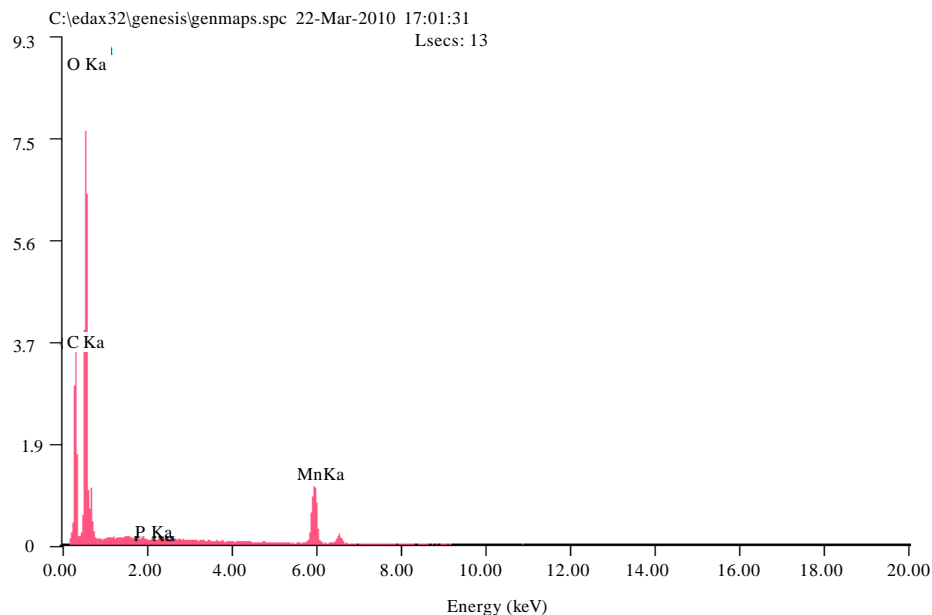


Fig. 12: EDX of Pendimethalin loaded hollow-shell after water wash

reduction in peak broadening was observed at the peaks 32 and 52° (Fig. 13). In the X-ray diffractogram of halo shell loaded with pendimethalin after methanol wash (HSP2) we can find the intensity of peaks had reduced at 24, 42, 44 and 52°. This was occurred due to the organic solvent wash which paved the way for the entry of herbicides into the shell rather than sticking onto the surface. This clearly pictures that after loading with herbicide it is necessary to give a solvent wash for uniform distribution of herbicide into the hollow-shell. This change confirms the active ingredients had been loaded into the hollow-shell. The 100% peak at 32° and other

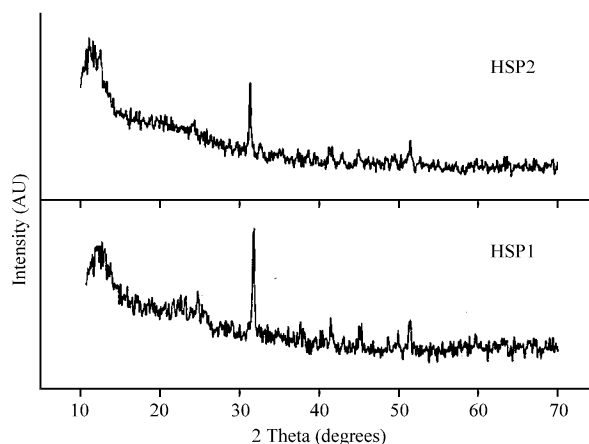


Fig. 13: XRD image of pendimethalin loaded halo-shell before (HSP1) and after methanol wash (HSP2)

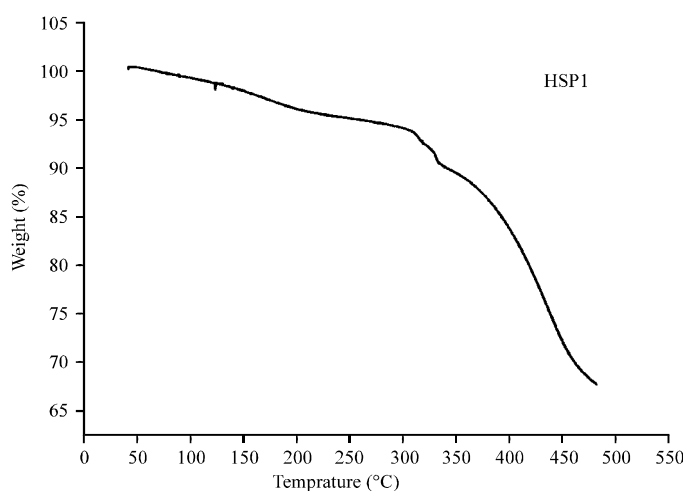


Fig. 14: TGA thermogram of pendimethalin loaded (HSP1)

characteristic peaks at 24, 42, 44 and 52° are seen clearly in the diffractogram. Thus from X-ray diffraction pattern we can qualitatively confirm the formation of core-shell, hollow shell and herbicide loaded hollow-shells. However the 100% peak at 32° remains the same which is the characteristic peak for core. This confirms the herbicides had loaded into the core thus diminishing the effect of MnCO_3 core-shell.

The TGA thermogram of pendimethalin loaded sample (Fig. 14) shows two stages decomposition at 180 and 320° and shows a steady decomposition pattern. Whereas for methanol washed sample (Fig. 15) shows a neat decomposition pattern. The decomposition points were at 180 and 290° which are different from the previous graph which clearly indicates the loading pattern of herbicides.

GC analysis-Encapsulation efficiency: The extracted pendimethalin sample was analyzed in the Gas (GC) chromatography to know the herbicide loading efficiency of the hollow-shell

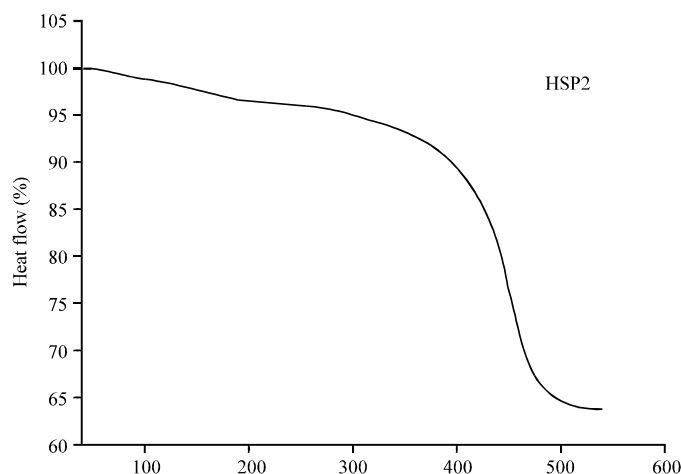


Fig. 15: TGA thermogram of pendimethalin loaded (HSP 2)

Table 1: GC analysis - Encapsulation efficiency of pendimethalin in the hollow-shell

S. No	Treatment	Encapsulation (ppm) Pendimethalin	
		HSP 1	HSP 2
1	10 mg of hollow-shell+10 ppm of active ingredient	0.486	0.461

(Table 1). In HSP1 without surface wash showed the highest herbicides loading of 0.486 ppm. In the solvent washed sample (HSP2) herbicide particles attached on the surface of hollow-shell were removed lead to reduction in the loading efficiency of herbicide (0.461 ppm) within the hollow-shell.

CONCLUSION

In this experiment, manganese carbonate (MnCO_3) core was synthesized using chemical method and characterised using XRD, FE-SEM, TGA which demonstrates that the particles were in homogeneity. Thus, formed MnCO_3 core particles were made into core-shell by coating with selected water soluble polymers such as sodium poly (styrene sulfonate) (PSS) and poly (allylamine hydrochloride) (PAH) using Layer by Layer method (LbL). Then they were made into hybrid inorganic/organic hollow spheres using dilute hydrochloric acid. Thus formed hollow-shell particles were used for loading the selected pre-emergence herbicide pendimethalin programmed to release smartly based upon the requirements. In addition the fabricated porous hollow-shell material can also be useful for loading other active ingredients such as drugs, chemicals, fertilizers etc for conditional release.

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