Journal of Hazardous Materials 177 (2010) 290–299

Contents lists available at ScienceDirect

Journal of Hazardous Materials

journal homepage: www.elsevier.com/locate/jhazmat

# Development of a new controlled pesticide delivery system based on neem leaf powder

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A R T I C L E I N F O

*Article history:*
Received 2 July 2009
Received in revised form
30 November 2009
Accepted 6 December 2009
Available online 29 December 2009

*Keywords:*
Bio-pesticide
Controlled release
Fungicide

A B S T R A C T

In order to minimize the agro-environmental pollution and health hazards caused by pesticides, in the present study, the neem leaf powder "(NLP)", a bio-pesticide, has been exploited to develop the pesticide delivery devices. The presence of neem in the formulations along with the pesticide may enhance the potential of these systems due to its inherent pesticidal activity. We have prepared the NLP and alginate based beads by using $CaCl_2$ as crosslinker. To study the effect of composition of the beads on the release dynamics of fungicide (thiram), beads were prepared by varying the amount of NLP and crosslinker. The beads formed were characterized with Fourier transform infrared spectroscopy (FTIR), scanning electron micrograph (SEM), energy dispersion analysis by X-rays (EDAX), thermogravimetric analysis and swelling study. Formulation characteristics such as entrapment efficiency, bead size, percentage equilibrium swelling of the beads and diffusion mechanism for thiram release have been evaluated. Maximum (78.33 ± 2.89)% swelling has occurred in the beads prepared with 1.5% NLP, 2.5% alginate and 0.1 M crosslinker solution. In most of the formulations the values for the diffusion exponent '*n*' have been obtained >1 and hence the release of fungicides occurred through Case II diffusion mechanism.

## 1. Introduction

Use of pesticides in India began in 1948. Currently, there is large number of pesticides registered for use and their production has increased enormously. Rampant use of these chemicals has given rise to several short-term and long-term adverse effects of these chemicals to human health such as immune-suppression, hormone disruption, diminished intelligence, reproductive abnormalities, and cancer. Both, polymer based controlled release formulations and bio-pesticides (i.e. pesticides obtained from natural plant sources such as neem extracts) are some of the future strategies for minimizing risks from pesticides [1]. Hence their detail discussion will be praiseworthy.

The neem, *Azadirachta indica*, has been known as a bio-pesticide against various insects [2,3] and most of its parts contain azadirachtin, which has most insecticidal activity among other limonoids, or tetranortriterpenoids present in neem trees [4]. Its properties such as toxicity, repellence, feeding deterrence, and insect growth regulator activity contribute mainly toward insecticidal activity [3]. Nowadays neem-based insecticides with azadirachtin as an active ingredient are also available commercially. Neem leaf powder (NLP) with other neem-based commercially available insecticides (Agroneem, Neemix and Ecozin) has been evaluated for its insecticidal effects [5]. Neem products are also known to show synergistic effect when used with other pesticides [6,7]. The concentration of azadirachtin content in neem leaves has been reported as 0.0244% (w/w), i.e. about 250 μg of azadirachtin per gram of neem leaves [8].

The polymer based controlled release formulations of pesticides are superior to conventional formulations [9–17] in extending activity [14], reducing leaching, evaporation and degradation [15,16] and decreasing dermal toxicity [17]. Natural polymers are preferred to synthetic polymers to prepare controlled release devices since these are relatively cheaper, biodegradable and left no polymer residues in the atmosphere. Alginate, a naturally occurring polysaccharide, after ionotropic gelation with metal ions has been used as controlled release formulation of pesticides [18–19].

In this sense, the usefulness of calcium-alginates as matrix material for controlled release formulations has been investigated with various herbicides such as monolinuron, desmetryn, chloridazon, atrazine, simazine, and chloroxuron as active ingredients. Release rate from these formulations showed sufficient retardation of herbicide release [18]. The addition of some additives (linseed oil, lignin, clays) in calcium alginate formulations has further modified the release rate of pesticides from the formulations [19–23].

In view of the above, we have developed a novel formulation by combining both the strategies. We have prepared the controlled release formulation by using bio-pesticides, i.e. neem leaf powder. This formulation may work with double action, i.e. inherent

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doi:10.1016/j.jhazmat.2009.12.031

**Table 1**
Reaction parameters for the synthesis of NLP-alginate beads.

| Formulation code | Neem leaf powder (NLP) (% w/v) | Alginate (% w/v) | Thiram (mg) | Crosslinker concentration (M) |
|---|---|---|---|---|
| $NA_1$ | 0 | 2.5 | 30 | 0.1 |
| $NA_2$ | 0.5 | 2.5 | 30 | 0.1 |
| $NA_3$ | 1.0 | 2.5 | 30 | 0.1 |
| $NA_4$ | 1.5 | 2.5 | 30 | 0.1 |
| $NA_5$ | 2.0 | 2.5 | 30 | 0.1 |
| $NA_6$ | 1.5 | 2.5 | 30 | 0.2 |
| $NA_7$ | 1.5 | 2.5 | 30 | 0.3 |
| $NA_8$ | 1.5 | 2.5 | 30 | 0.4 |

pesticidal action of NLP bio-pesticide and release of loaded pesticide (thiram). The bio-pesticidal action of NLP [5,8] and pesticidal action of thiram [24] have been reported in different studies. In this study, we have used thiram as a model pesticide that has been used as a fungicide to control soil fungi that cause root rot diseases of plants [24] and has been used to protect harvested crops, cereals, seeds, fruits, vegetables, and turf crops from deterioration in storage or transport [25,26]. Like other pesticides it has also been reported to have adverse effects on hepatic system [27], the reproductive system [28,29] and on the developmental processes [30].

Therefore, the present study is an attempt to synthesize NLP-alginate based formulations for controlled and sustained release of thiram fungicide. The beads of different composition were prepared by varying amount of neem leaf powder and crosslinker concentration and were characterized by Fourier transform infrared spectroscopy (FTIR), scanning electron micrograph (SEM), energy dispersion analysis by X-rays (EDAX), thermogravimetric analysis and swelling study. The effect of different amounts of neem leaf powder (NLP) and crosslinker concentration on the release dynamics of fungicide has been evaluated.

## 2. Experimental

### 2.1. Materials and methods

Acetonitrile and chloroform were obtained from Merck-Schuchardt, Germany. Neem leaf powder (NLP) was obtained by grinding dry neem leaves collected form Bilaspur District of Himachal Pradesh, India. Sodium alginate was obtained from Loba Chemie Pvt. Ltd., India. Tetramethylthiuram disulphide (thiram) was obtained from Fluka, Switzerland and was used as received.

### 2.2. Synthesis of NLP-alginate beads

Measured amount of neem leaf powder (NLP), alginate and thiram were dispersed in 25 mL hot water and stirred for 15 min to form homogenous solution. This homogeneous solution was then added drop wise, by using 50 mL syringe (needle size 1.2 mm) from 15 cm height, into 100 mL of $CaCl_2$ solution (0.1 M) under constant stirring. The beads formed were removed from $CaCl_2$ solution after 30 min and washed with distilled water and were allowed to dry at room temperature. The optimum reaction parameters for the synthesis of beads were obtained by varying the NLP and concentration of crosslinker. Amount of neem leaf powder was varied from 0 to 2% (w/v) and concentration of crosslinker was varied from 0.1 to 0.4 M during the synthesis of beads (Table 1). The beads of different compositions were designated as different formulations (that is $NA_1$ to $NA_8$) shown in Table 1 and were used to study the release dynamics of the thiram from these formulations.

### 2.3. Beads size measurement

Fifteen completely dried beads from each formulation ($NA_1$ to $NA_8$) were taken and their size was measured by using 12 cm Vernier Calipers. The average bead size of each formulation is presented in Table 2.

### 2.4. Characterization

NLP-alginate beads were characterized by Fourier transform infrared spectroscopy (FTIR) to study the blending of these polysaccharides in the presence of calcium ions, scanning electron micrograph (SEM) to study the surface morphology of the beads, energy dispersion analysis by X-rays (EDAX) to observe the qualitative composition of the beads and thermogravimetric analysis to study the thermal stability of the sample and swelling studies to optimize the matrix for end use such as controlled release of pesticides.

FTIR spectra of NLP, alginate and NLP-alginate beads were recorded in KBr pellets on Nicolet 5700 FTIR. SEM and EDAX were taken by using QUANTA 200 FEG model (Netherlands) after depositing the samples on a brass holder and sputtering them with gold. Thermogravimetric analysis was carried out on a Perkin-Elmer (Pyris Diamond) thermal analyzer in air at a heating rate of 20 °C/min to examine the thermal properties of the samples.

### 2.5. Swelling studies

Swelling studies of the beads were carried out in aqueous medium at (26 ± 1) °C by gravimetric method. Known weight of the beads were taken and immersed in excess of water for 24 h at (26 ± 1) °C and then beads were removed, wiped with tissue paper to remove excess of water and were weighed immediately. The

**Table 2**
Formulation characteristics of NLP-alginate beads.

| Formulation code | Beads formed (g) | Bead diameter (mm) | Entrapment efficiency (%) | Fungicide loading (mg/g of beads) | Equilibrium swelling (%) |
|---|---|---|---|---|---|
| $NA_1$ | 0.84 ± 0.05 | 0.55 ± 0.07 | 97.09 ± 1.32 | 34.67 ± 0.47 | 43.33 ± 2.89 |
| $NA_2$ | 0.98 ± 0.03 | 0.61 ± 0.08 | 97.08 ± 1.13 | 29.72 ± 0.34 | 51.67 ± 2.89 |
| $NA_3$ | 1.09 ± 0.10 | 0.67 ± 0.08 | 98.06 ± 1.07 | 26.99 ± 0.29 | 51.67 ± 2.89 |
| $NA_4$ | 1.16 ± 0.15 | 0.85 ± 0.07 | 97.51 ± 1.09 | 25.22 ± 0.28 | 78.33 ± 2.89 |
| $NA_5$ | 1.07 ± 0.18 | 0.87 ± 0.09 | 97.28 ± 1.23 | 27.28 ± 0.35 | 78.33 ± 2.89 |
| $NA_6$ | 1.24 ± 0.16 | 0.81 ± 0.06 | 97.70 ± 1.17 | 23.64 ± 0.28 | 60.00 ± 5.00 |
| $NA_7$ | 1.52 ± 0.11 | 0.77 ± 0.07 | 97.45 ± 1.20 | 19.23 ± 0.24 | 35.00 ± 5.00 |
| $NA_8$ | 1.68 ± 0.33 | 0.73 ± 0.13 | 97.66 ± 1.10 | 17.44 ± 0.20 | 31.67 ± 2.89 |

**Table 3**
Thermogravimetric analysis of NLP, sodium alginate, $Ca^{2+}$-alginate, NLP-alginate beads.

| Sample | IDT (°C) | FDT (°C) | Decomposition temp (°C) at every 10% weight loss | | | | | | | | | | Residue left (%) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | 10 | 20 | 30 | 40 | 50 | 60 | 70 | 80 | 90 | 100 | |
| NLP | 227 | 489 | 196 | 254 | 283 | 304 | 334 | 414 | 450 | 478 | 489 | - | 10.75 |
| Sodium alginate | 217 | 578 | 87 | 198 | 234 | 247 | 261 | 359 | 554 | 578 | - | - | 23.28 |
| $Ca^{2+}$-alginate | 198 | 787 | 158 | 225 | 273 | 298 | 421 | 537 | 761 | 787 | - | - | 29.34 |
| NLP-alginate | 217 | 791 | 90 | 180 | 260 | 292 | 345 | 514 | 700 | 791 | - | - | 20.91 |

percent equilibrium swelling ($P_s$) of the beads was calculated as

$$P_s = \left[\frac{W_s - W_d}{W_d}\right] \times 100 \qquad \text{(I)}$$

where $W_s$ is weights of swollen beads and $W_d$ is the weight of dried beads. The effect of different composition of the beads on equilibrium swelling was studied.

### 2.6. Fungicide loading and entrapment efficiency

To determine funguicide loading (FL) and entrapment efficiency (EE), calibration curves for pure thiram were constructed as follows.

A number of standard solutions of the reference compound at concentrations encompassing the samples concentrations were measured spectrophotometrically using carry 100Bio UV–vis spectrophotometer by modifying Verma et al. [31] method. Aliquots (0.02–2.0 mL) of standard solution of thiram ($10^{-3}$ M) in acetonitrile were taken in 100 mL separating funnels and diluted to 2 mL with acetonitrile. To each solution was added 5 mL of water, 1 mL of each of EDTA and ammonium buffer (to avoid any interference from $Ca^{2+}$ ions) and tetraacetonitritocopper(I)perchlorate (1 mL, 0.004 M in acetonitrile). Each mixture solution was equilibrated with 5 mL of chloroform for 10 min. The chloroform layer was separated and dried over anhydrous sodium sulphate. The final volume was made to 10 mL with chloroform. The absorbance of the yellow chloroform extract was measured at 432 nm ($\lambda_{max}$ of yellow colored copper-dithiocarbamate complex) against a reagent blank. Calibration curve was constructed by plotting absorbance values against concentration of thiram taken. The Beer's law is obeyed up to 48 μg/mL of thiram. Using the straight line equation $y = mx + c$, the slope and intercept values were 0.03847 and −0.13051, respectively and With Relative standard deviation of 1.4%.

The entrapment efficiency (%) was calculated spectrophotometrically by measuring the thiram contents left in the $CaCl_2$ solution from the beads by the method given above. The entrapment efficiency (%) and FL (per gram of beads) were then calculated as

$$\text{entrapment efficiency} = \left\{\frac{C_1 - C_2}{C_1}\right\} \times 100 \qquad \text{(II)}$$

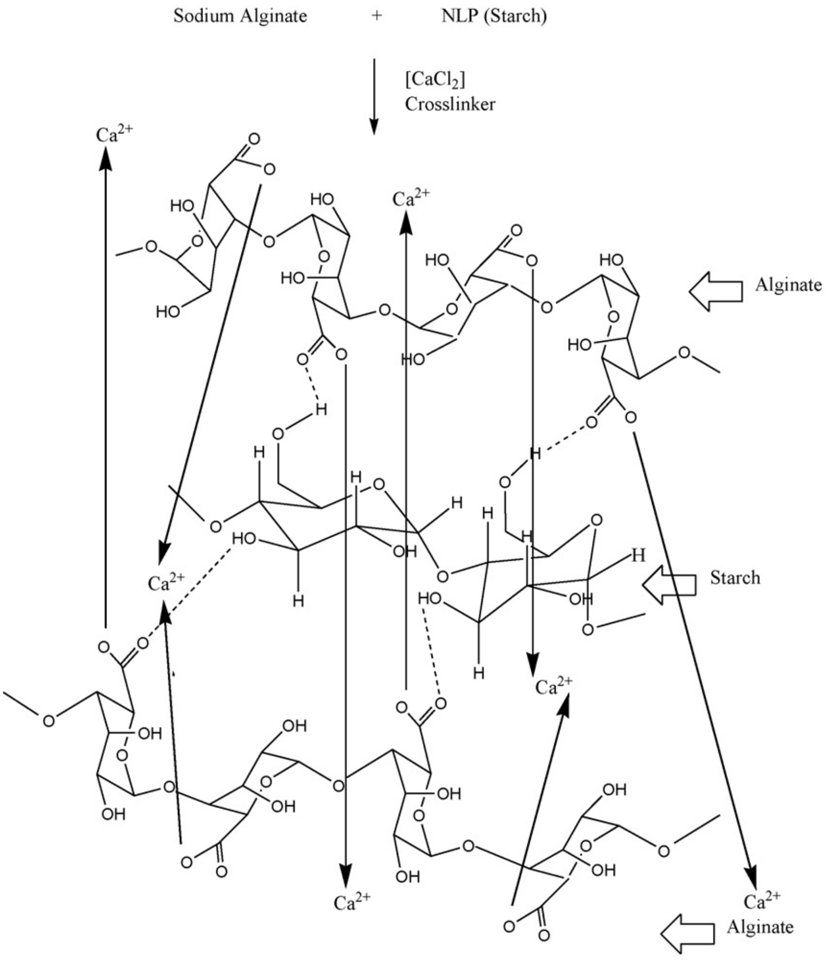

**Plate 1.** Reaction/interactions between alginate, calcium ions and starch (NLP).

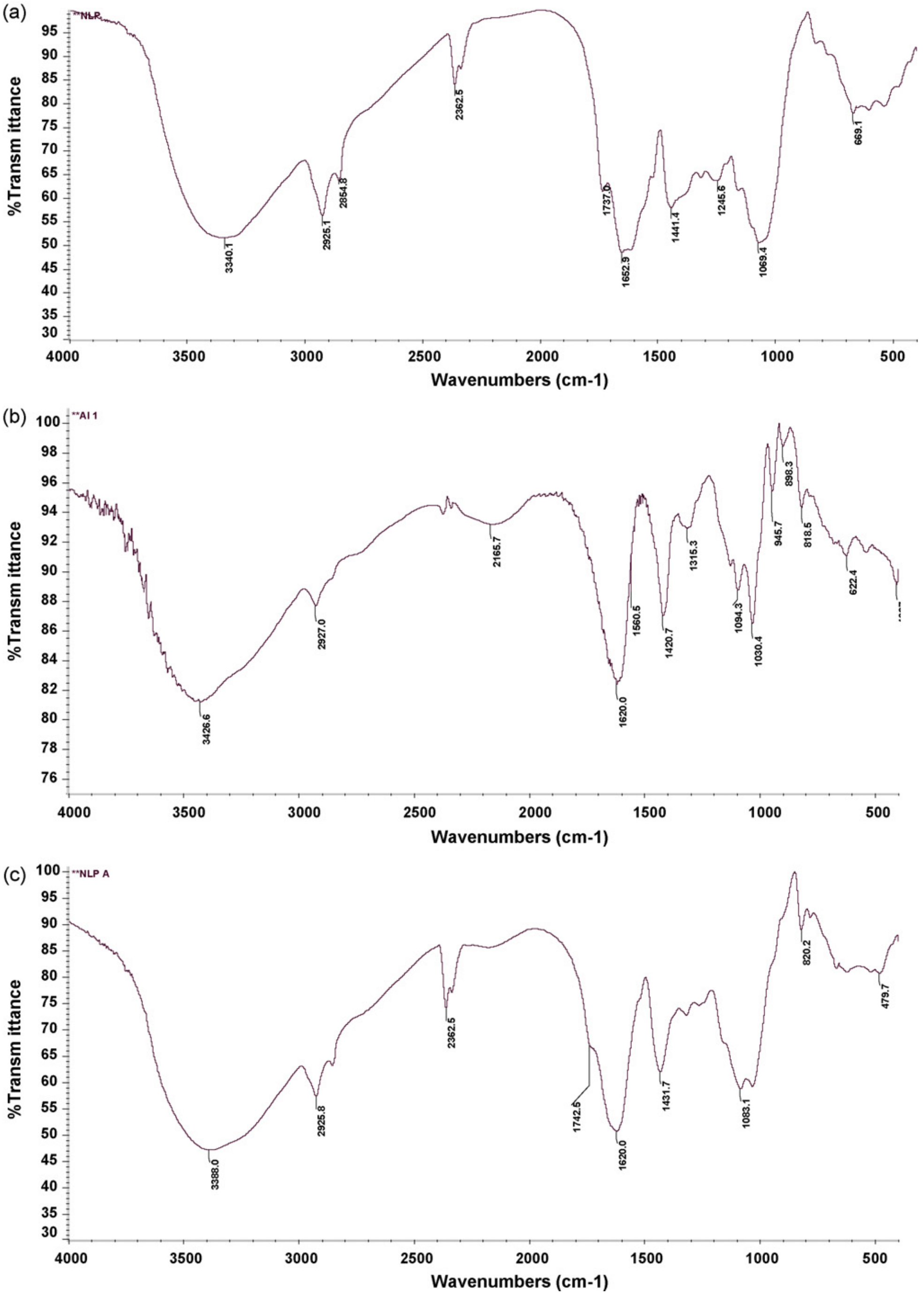

**Fig. 1.** FTIR of (a) neem leaf powder (NLP), (b) alginate and (c) NLP-alginate beads.

$$\text{FL (per g of beads)} = \frac{C_1 - C_2}{W} \qquad \text{(III)}$$

where $C_1$ is known amount of thiram in reaction mixture (theoretical), $C_2$ is amount of thiram left in crosslinker solution (practical) and $W$ is amount of beads formed in grams. The entrapment efficiency and FL (per gram of the beads) for formulations $NA_1$ to $NA_8$ are shown in Table 2.

### *2.7. Release dynamic studies of thiram from NLP-alginate based beads*

In vitro release of the fungicide has been carried out by keeping dried and loaded samples of each formulation (200 mg) in definite volume of releasing medium (10 mL water) at (26 ± 1)°C. The amount of thiram released was measured spectrophotometrically after every 12 up to 204 h and release was measured after 300 h. All the experiments were carried out in triplicate. The effect of different amounts of NLP and crosslinker concentration on fungicide release was studied.

#### *2.7.1. Mathematical modeling of fungicide release*

Mathematical modeling of drug release from swellable polymeric systems will be applied for the release of pesticide from the polymer matrix. Although there are a number of reports dealing with mathematical modeling of drug release from swellable polymeric systems, no single model successfully predicts all the experimental observations. Fickian, non-Fickian and Case II

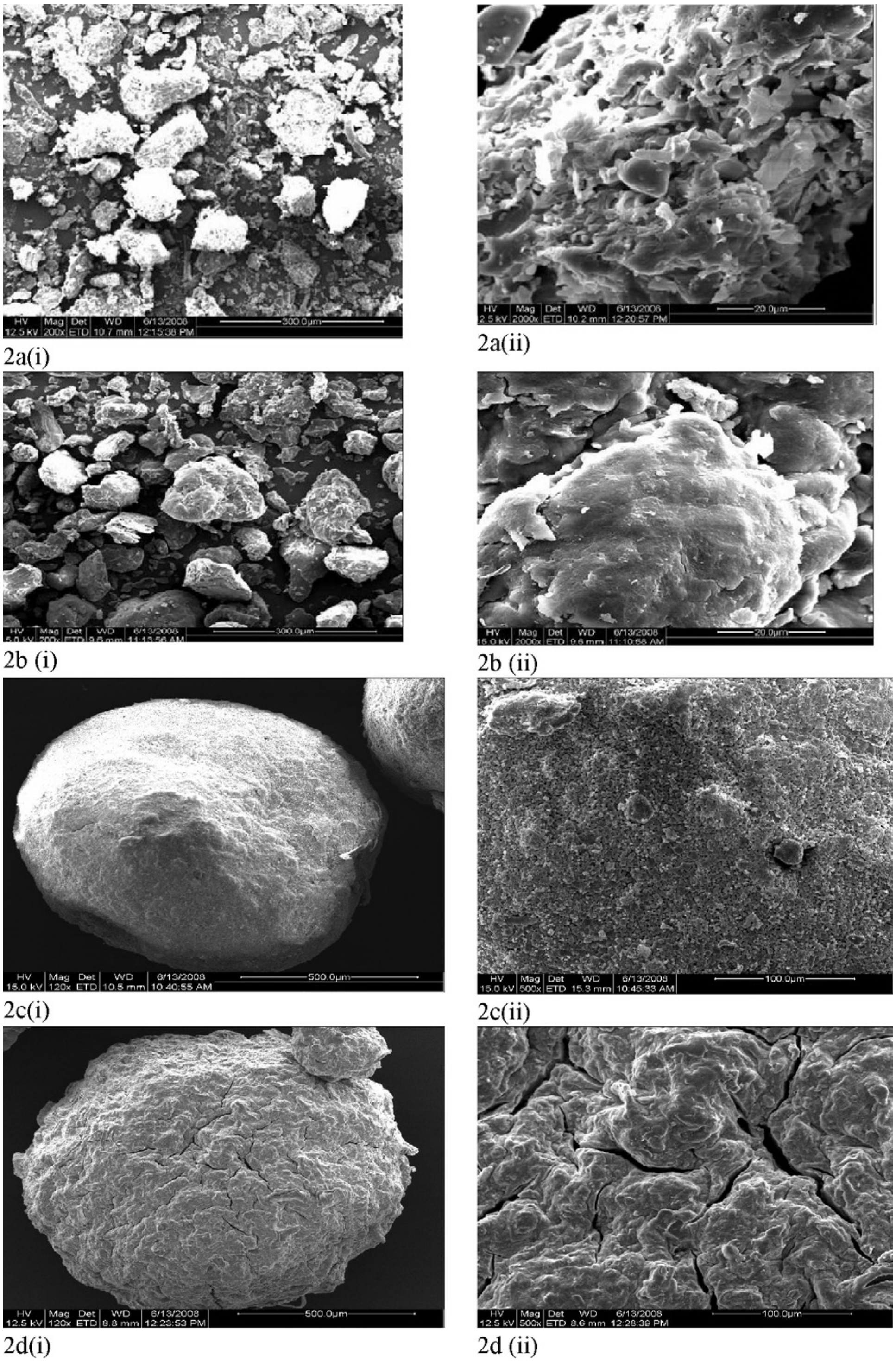

**Fig. 2.** SEMs of (a) NLP {(i) 200×; (ii) 2000×}; (b) sodium alginate {(i) 200×; (ii) 2000×}; (c) $Ca^{2+}$-alginate bead {(i) 120×; (ii) 500×} and (d) NLP-alginate bead {(i) 120×; (ii) 500×}.

diffusion mechanism for swelling of polymers and for the drugs release from the polymers can be calculated from Eq. (IV):

$$\frac{M_t}{M_\infty} = kt^n \qquad \text{(IV)}$$

where $M_t/M_\infty$ is the fractional release of drug in time $t$, '$k$' is the constant characteristic of the drug–polymer system, and '$n$' is the diffusion exponent characteristic of the release mechanism. For Normal Fickian diffusion the value of $n = 0.5$, Case II diffusion $n = 1.0$ and non-Fickian $n = 0.5$–$1.0$ [32–35]. The values of diffusion exponent '$n$' and gel characteristic constant '$k$' have been evaluated for the release dynamics of fungicide from the beads and results are presented in Table 4, along with the correlation coefficient '$r$'.

## 3. Results and discussion

### 3.1. Effect reactions parameters on formulation characteristics

It has been observed from Table 2 that the amount of NLP-alginate beads increased with increase in contents of NLP in the

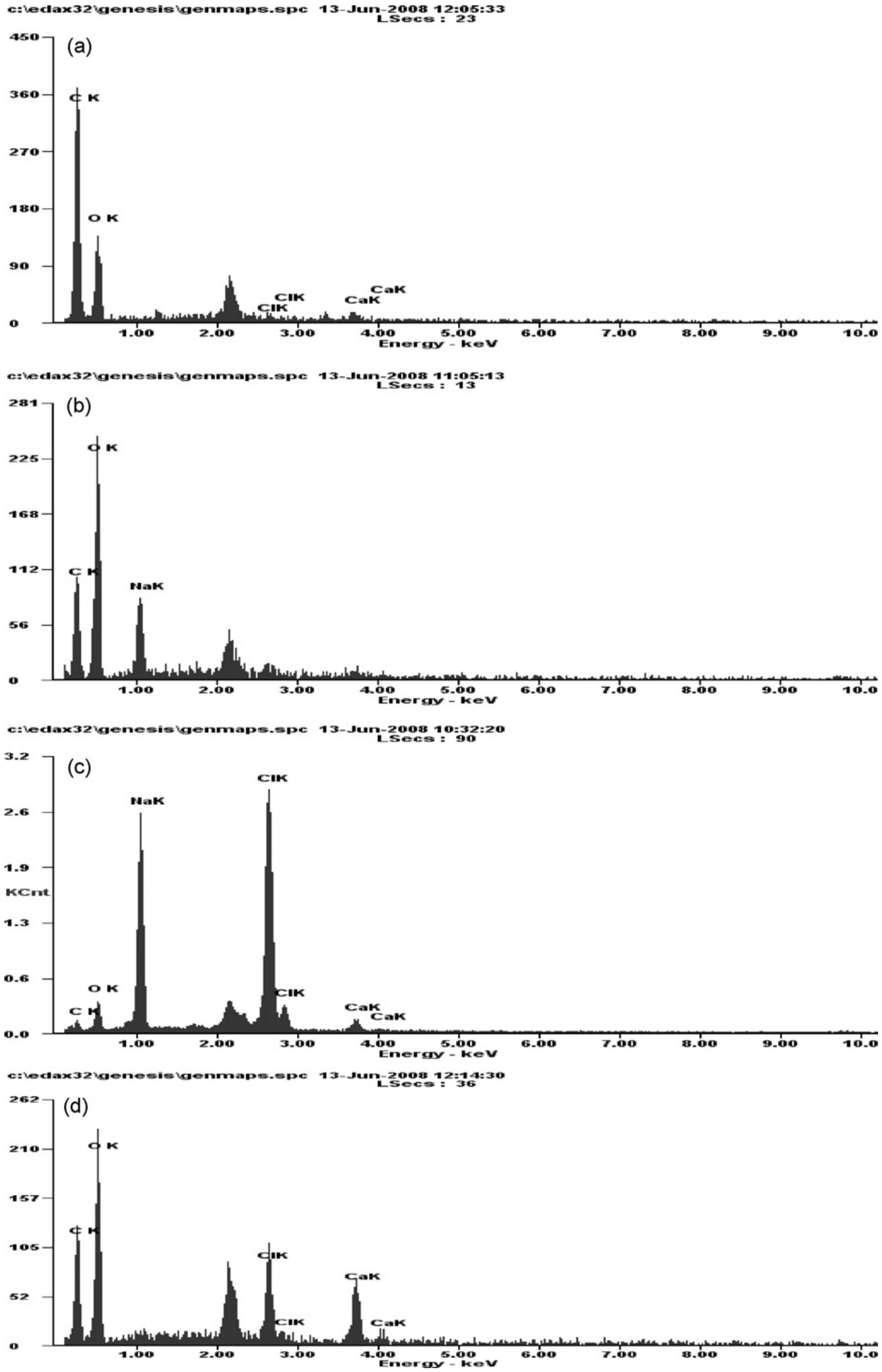

**Fig. 3.** EDAX of (a) NLP, (b) sodium alginate, (c) $Ca^{2+}$-alginate beads and (d) NLP-alginate beads.

feed solution during synthesis. The increase in yield was almost equivalent to the amount of NLP used during the synthesis. Further with increase in crosslinker concentration in the reaction solution has also increased the yield of the product. This may be due to the more and more crosslinking in the presence of calcium ions which get incorporated into the beads. The main constituent in neem leaves is carbohydrate (starch) and the interaction between alginate and neem leaf powder might be similar to the interaction between alginate and starch. The main reaction between alginate, calcium ions and starch content of NLP is shown in Plate 1. Since FL is related to amount of beads formed therefore its value for different bead formulations was different. The beads were spherical in shape and their diameters varied from (0.55 ± 0.07) mm to (0.87 ± 0.09) mm in different formulations. The size of beads increased with increase in amount of NLP in the beads and decreased with increase in crosslinker concentration (Table 2). The increase in size with increase in NLP content can be attributed to the presence of NLP as filler in the beads where as decrease in size with increase in crosslinker concentration can be explained as more and more carboxylate groups in alginate bind to metal ions, which increase the degree of crosslinking, and decrease the size of the beads [36]. Percentage entrapment efficiency has been observed about 97% in all the formulation (Table 2).

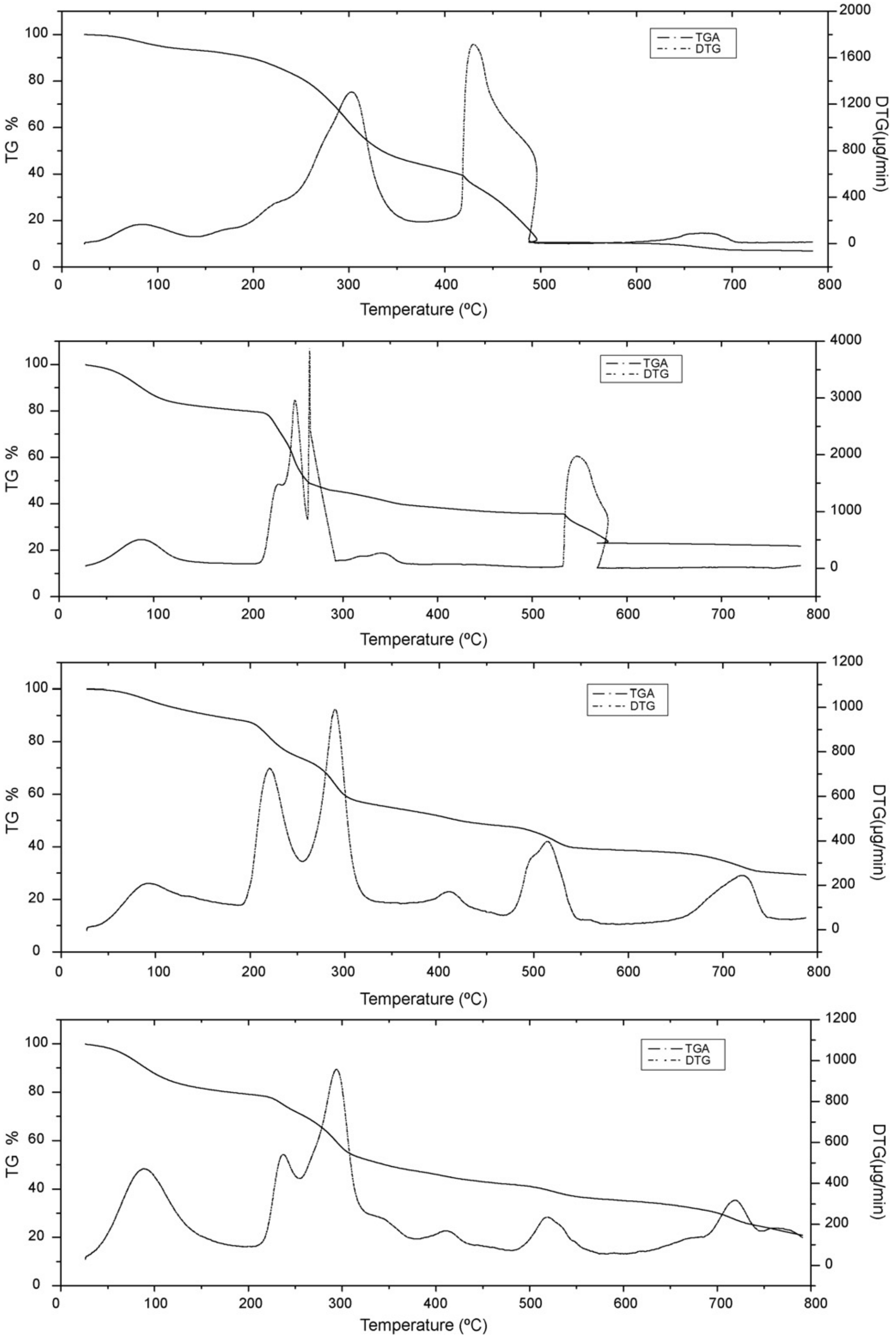

**Fig. 4.** (a) Thermogram of NLP observed from TGA. (b) Thermogram of sodium alginate observed from TGA. (c) Thermogram of $Ca^{2+}$-alginate bead observed from TGA. (d) Thermogram of NLP-alginate bead observed from TGA.

## *3.2. Characterization*

### *3.2.1. Fourier transform infrared spectroscopy*

FTIR spectra of neem leaf powder (NLP), alginate and NLP-alginate crosslinked beads were recorded in KBr pellets and are presented in Fig. 1a–c, respectively. The strong and broad absorption band has been observed between 3600 and 3200 $cm^{-1}$ due to –OH stretching which is the characteristic of the natural polysaccharides in all the three cases. In case of Fig. 1b and c, the presence of strong asymmetric stretching absorption band between 1650 $cm^{-1}$ and weaker symmetric stretching band near 1420 $cm^{-1}$ has been observed which is supporting the presence of carboxylate anion of sodium alginate and calcium alginate.

### 3.2.2. SEM and EDAX analysis

Scanning electron micrographs were obtained by scanning the surface of samples with high energy beam of electrons. These electrons interact with atoms that constitute the sample and produce the signal which contains the information about surface topology. The SEMs of NLP, sodium alginate, $Ca^{2+}$-alginate and NLP-alginate beads at different magnification are shown in Fig. 2a–d, respectively. The micrographs (at 200× magnification) of powdered NLP and sodium alginate have shown the assemblage of fine particles which were irregular in shape and size. However, in case of NLP micrographs at 2000× magnification, the particles have shown a large number of steps, kinks and broken edges in them [37] where as in sodium alginate the particles have shown a smooth and fine surface. From Fig. 2c and d, it is cleared that the beads have spherical shape (120× magnifications). Further, the calcium alginate beads have smooth surface where as NLP-alginate beads have rough surface which might be due to the presence of NLP (500× magnification). The SEMs have also revealed that NLP-alginate beads were larger in size than calcium alginate beads.

The same high energy beams of electrons which were used in SEM analysis have also interacted with the deep region of the sample. This interaction has emitted the X-rays (secondary fluorescence) which were characteristics of the elements present in the sample. This forms the basis of energy dispersion analysis by X-rays (EDAX) and allows a qualitative analysis of chemical composition of sample. The EDAX of neem leaf powder (NLP), sodium alginate, $Ca^{2+}$-alginate and NLP-alginate beads are shown in Fig. 3a–d, respectively. The EDAX spectrum of all species shows the presence of carbon and hydrogen which are the main constituents in polysaccharides. The elemental peak for calcium in $Ca^{2+}$-alginate and NLP-alginate beads are supporting the fact that crosslinking is due to $Ca^{2+}$ ions in the beads.

### 3.2.3. Thermogravimetric analysis

The primary thermograms of NLP, sodium alginate, $Ca^{2+}$-alginate and NLP-alginate beads are shown in Fig. 4a–d, respectively. These were obtained in the range of 25–800 °C under air atmosphere at 20 °C/min heating rate. In general, decomposition of polysaccharides consists of four phases; each phase corresponds to the characteristic decomposition pattern of that polysaccharide. These include desorption of physically absorbed water, removal of structural water (dehydration reactions), depolymerization accompanied by the rupture of C–O and C–C bonds in the ring units resulting in the evolution of CO, $CO_2$ and $H_2O$ and finally the formation of polynuclear aromatic and graphitic carbon structures [38]. In case of alginates TGA, the temperature range between 40 and 160 °C corresponds to the loss of different types of water molecules depending up on their interaction with the polysaccharides [39]. In the present studies, the actual polymeric decomposition has started after the loss of 14.43%, 21.04%, 12.44% and 21.67% water in the NLP, sodium alginate, $Ca^{2+}$-alginate and NLP-alginate beads, respectively and their respective initial decomposition temperatures have been observed at 227, 217, 198 and 217 °C. The decomposition temperature corresponding to 10% weight loss in each sample along with the initial decomposition temperature (IDT) and final decomposition temperature (FDT) are shown in Table 3. The final decomposition temperatures have been observed at 489, 578, 787 and 791 °C, respectively for NLP, sodium alginate, $Ca^{2+}$-alginate and NLP-alginate beads. In each case residue left at FDT is 10.75%, 23.28%, 29.34% and 20.91%, respectively. The higher residue left in case of sodium alginate, $Ca^{2+}$-alginate and NLP-alginate beads can be attributed to the oxides and carbonates of sodium and calcium. Further, from differential themogravimetry (DTG), the temperatures of maximum rate of weight loss ($T_{max}$) have been observed at 430, 265, 290 and 293 °C, respectively for NLP, Sodium alginate, $Ca^{2+}$-alginate and NLP-alginate beads.

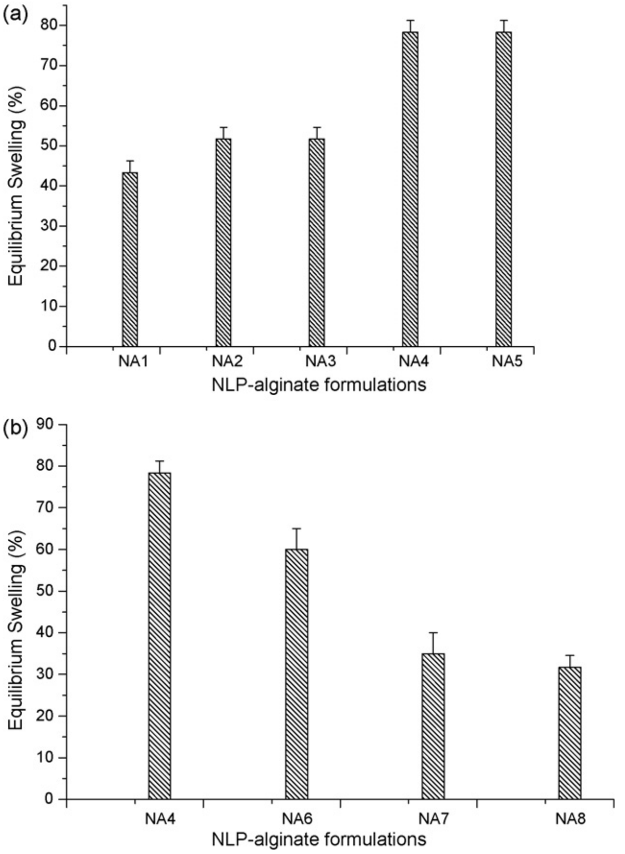

**Fig. 5.** (a) Effect of amounts of neem leaf powder (NLP) on % equilibrium swelling of NLP-alginate beads in distilled water. [NLP content (w/v): 0%($NA_1$); 0.5%($NA_2$); 1.0%($NA_3$); 1.5%($NA_4$); 2.0%($NA_5$). Content: 2.5% (w/v); [$CaCl_2$]: 0.1 M]. (b) Effect of crosslinker concentration on % equilibrium swelling of NLP-alginate beads in distilled water. [[$CaCl_2$]: 0.1 M ($NA_4$); 0.2 M ($NA_6$); 0.3 M ($NA_7$); 0.4 M ($NA_8$). Alginate content: 2.5% (w/v); NLP content: 1.5% (w/v).]

### 3.2.4. Swelling studies

The percentage equilibrium swelling of each formulation was studied after 24 h and the effect of NLP and crosslinker concentration on the equilibrium swelling has been determined in each case and results are presented in Fig. 5a and b, respectively (Table 2). The overall equilibrium swelling of NLP-alginate beads ($NA_1$ to $NA_8$) varied significantly from (31.67 ± 2.89)% to (78.33 ± 2.89)%. Swelling increases with increase in NLP contents in the formulation. As the percentage of NLP is increased from 0 to 2% (w/v) in NLP-alginate beads, the percentage swelling is increased considerably from (43.33 ± 2.89)% to (78.33 ± 2.89)% (Fig. 5a). This may be due to the hydrophilic nature of the carbohydrate (starch and cellulose, the main constituent in neem leaf) present in the beads. As the amount of NLP increases, the number of interaction of –OH groups present in it with water increases which have increased the swelling of the beads. Decrease in swelling has been observed with increase in crosslinker concentration in the beads due to increase in crosslinking density and decrease in pore size. Maximum (78.33 ± 2.89)% swelling has been noted in the formulation prepared in 0.1 M $CaCl_2$ solution (Fig. 5b).

## 3.3. Release dynamics thiram fungicide from NLP-alginate beads

Release of thiram from the different formulations prepared with different NLP contents and crosslinker concentration has been studied and results are presented in Figs. 6 and 7, respectively. The

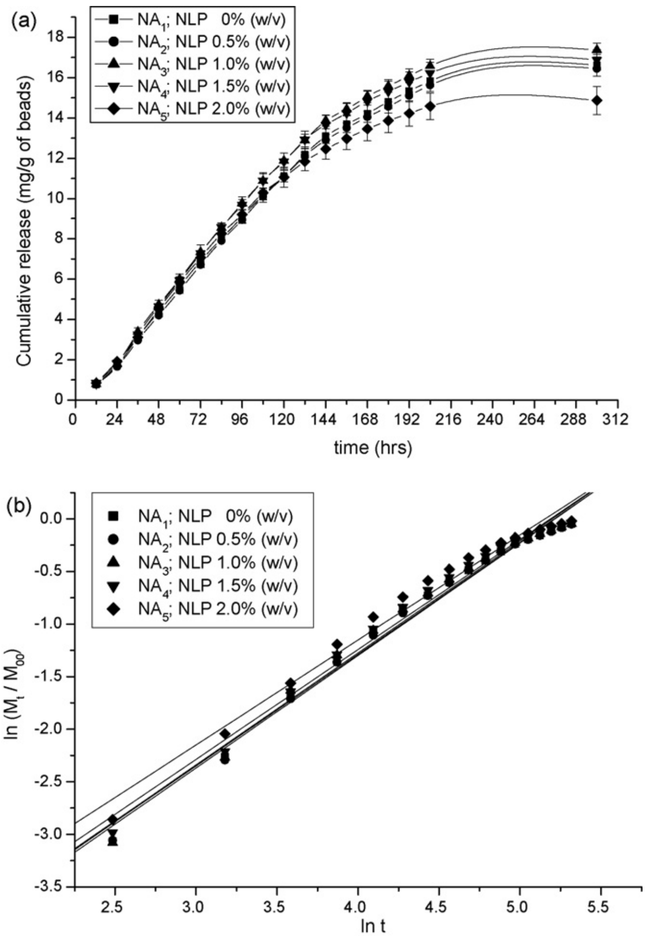

**Fig. 6.** (a) Release dynamics of thiram from fungicide loaded samples of NLP-alginate beads prepared with different contents of neem leaf powder (NLP). [Alginate content: 2.5% (w/v); [$CaCl_2$]: 0.1 M.] (b) Plot of $\ln M_t/M_\infty$ versus $\ln t$ for the evaluation of '*n*' and '*k*' for the release dynamics of thiram from fungicide loaded samples of NLP-alginate beads prepared with different amounts of neem leaf powder (NLP). [Alginate content: 2.5% (w/v); [$CaCl_2$]: 0.1 M.]

release of thiram from the beads prepared with different NLP contents has shown irregular trends with increase in NLP contents in the formulations. In general, the increase in NLP contents in the formulation has increased the release of fungicide. Maximum 16.61, 16.44, 17.37, 16.88 and 14.87 mg of thiram release were observed, respectively in formulations namely $NA_1$, $NA_2$, $NA_3$, $NA_4$ and $NA_5$ (Fig. 6a). Initial rate of release of thiram from the formulations has been observed more than the latter stages. These observations show that the cumulative release of thiram from the beads occurs in sustained manner, which is a primary requisite for rational use of agrochemicals from this type of devices. The 50% of the total release of thiram from the loaded NLP-alginate beads prepared with different NLP contents, viz., $NA_1$, $NA_2$, $NA_3$, $NA_4$ and $NA_5$ occurred in 83.36, 82.81, 81.54, 79.39 and 73.62 h, respectively (Fig. 6a).

From the slope and intercept of the plot of $\ln M_t/M_\infty$ versus $\ln t$ (Fig. 6b), the diffusion exponent '*n*' and gel characteristic constant '*k*' for the release of fungicide from the beads prepared with different NLP content has been evaluated to study the mechanism of release of thiram from the loaded formulations of NLP-alginate beads. The results obtained from this figures are presented in Table 4. The values of '*n*' for NLP-alginate beads prepared with different NLP contents ($NA_1$ to $NA_5$) have been observed >1 which shows that the release of thiram from these formulations occurred through Case II diffusion mechanism. The Case II diffusion mechanism is relaxation-controlled transport and it occur when the rate of diffusion of water molecules in the polymer matrix is very rapid as compared with relaxation of polymeric chains and the rate of water penetration is controlled by polymer relaxation and release of chemical occurs as it diffuses out when the polymer swells by absorbing water [34–35].

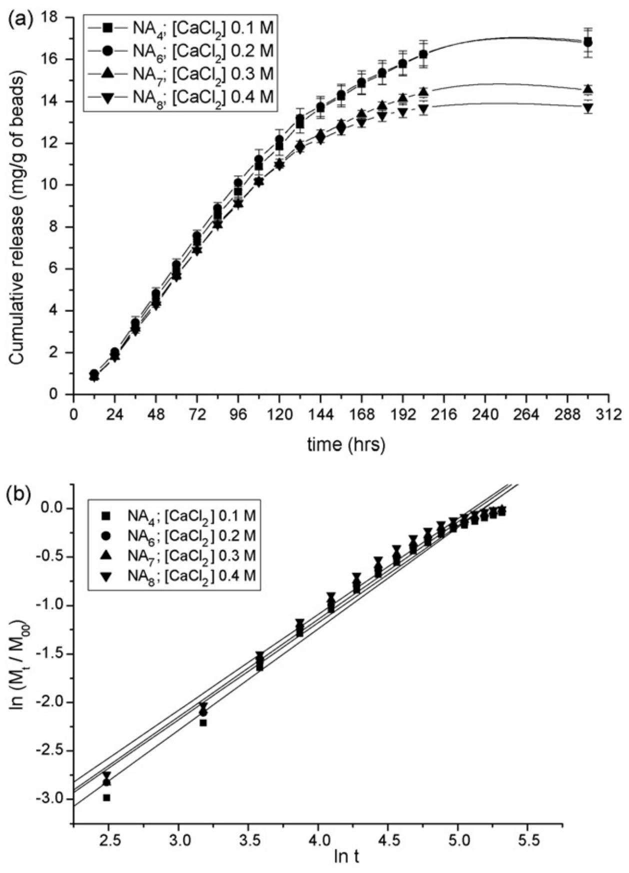

**Fig. 7.** (a) Release dynamics of thiram from fungicide loaded samples of NLP-alginate beads prepared with different crosslinker concentration. [Alginate content: 2.5% (w/v); NLP content: 1.5% (w/v).] (b) Plot of $\ln M_t/M_\infty$ versus $\ln t$ for the evaluation of '*n*' and '*k*' for the release dynamics of thiram from fungicide loaded samples of NLP-alginate beads prepared with different crosslinker concentration. [Alginate content: 2.5% (w/v); NLP content: 1.5% (w/v).]

The effect of crosslinker concentration on the release profile of thiram from fungicide loaded beads is presented in Fig. 7a. It is clear from the figure that the release of fungicide from the formulation decreases with increase in crosslinker concentration in the polymer matrix. These results are corresponding to the swelling of these formulations. Maximum 16.88, 16.80, 14.54 and 13.75 mg of thiram release have occurred in the formulations prepared in different crosslinker concentration [$CaCl_2$], viz., $NA_4$, $NA_6$, $NA_7$ and $NA_8$, respectively. The rate of release of thiram decreased with

**Table 4**
Results of diffusion exponent '*n*', gel characteristic constant '*k*' and correlation coefficient '*r*' for the release of thiram from fungicide loaded samples of NLP-alginate beads.

| Formulation code | $k \times 10^2$ | *n* | *r* | Diffusion mechanism |
|---|---|---|---|---|
| $NA_1$ | 0.39 | 1.06 | 0.993 | Case II |
| $NA_2$ | 0.38 | 1.07 | 0.994 | Case II |
| $NA_3$ | 0.40 | 1.06 | 0.992 | Case II |
| $NA_4$ | 0.44 | 1.05 | 0.992 | Case II |
| $NA_5$ | 0.58 | 1.00 | 0.999 | Case II |
| $NA_6$ | 0.57 | 1.00 | 0.992 | Case II |
| $NA_7$ | 0.57 | 1.00 | 0.991 | Case II |
| $NA_8$ | 0.65 | 0.99 | 0.989 | Non-Fickian |

increase in crosslinker concentration in the beads. The 50% of the total release of thiram from of the loaded NLP-alginate beads prepared with different crosslinker concentration, viz., $NA_4$, $NA_6$, $NA_7$ and $NA_8$ occurred in 79.39, 76.74, 72.93 and 69.22 h, respectively (Fig. 7a). From Fig. 7b, value of '*n*' and '*k*' has been calculated which have showed the Case II mechanism for release of thiram in the most of formulations (Table 4).

## 4. Conclusion

It is concluded from the forgone discussion that the composition of the formulation has exerted very strong effect on the swelling of the beads and release pattern of the thiram from these beads. It is further concluded that increase in NLP contents after certain limit in these formulations has increased the release of the fungicide. However, increase in crosslinker concentration in the formulations has decreased the release of thiram. The release of thiram from these beads has occurred in very controlled and sustained manner, which is the primary requisite for the use of agrochemicals to control the environment, ecosystem and health hazards. Hence, these polymeric beads may be utilized for the safe handling of pesticide, to reduce their toxic effects, and to make their better delivery [40]. The release of fungicide from most of the formulations occurred through Case II diffusion mechanism. It may be proposed that use of NLP in the formulations may increase the potential of these delivery devices due to its inherent nature as bio-pesticide and release of loaded pesticide in slow manner. Synergic action can also not be ignored.

## Acknowledgements

One of the author (Atul Gupta) wish to thanks the Council of Scientific and Industrial Research, New Delhi, India for providing the financial assistance for this work (Grant No. 9/237(0115)/2007-EMR-I).

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