Synthesis, Characterization and Application of Zeolitic Imidazole Framework-Mesoporous Silica Nanospheres Composite: A Hybrid Porous Composite for Drug Delivery

Dr. Chandan Adhikari¹ and Dr. Anjan Chakraborty²

¹Department of Chemistry, School of Chemical Engineering and Physical Sciences, Lovely Professional University

²Department of Chemistry, Indian Institute of Technology Indore, Indore.

email: chandan.23667@lpu.co.in
Abstract

Drug delivery system composed of mesoporous silica nanoparticles suffers from many drawbacks. Out of many challenges, two major challenges in drug delivery through mesoporous silica nanoparticles, are premature leakage and fast release of the drug molecules. Because of these, most of the time, efficiency of the drug delivery system become very low. In this work, mesoporous silica nanoparticles (MSN) have been modified using zeolitic imidazole framework through formation of a hybrid system. An anticancer drug Doxorubicin was encapsulated in mesoporous silica nanoparticles. Two zeolitic imidazole frameworks, ZIF-7 and ZIF-8 were prepared and used to form composite with mesoporous silica nanoparticles. The composites were characterized using scanning electron microscope, confocal laser scanning microscope, bright field imaging, powder X-ray diffraction, Fourier transform infrared spectroscopy, thermogravimetric analysis. BET surface analysis was conducted to understand the pore size, pore volume and surface area of the composite materials. The drug delivery study was conducted under pH stimuli as well as in present of liposome. The bare MSN were found to release the drug within 2-3 hours at pH~4 and in presence of liposome. But both the composites were found to control the drug release over a period of 12 hours at pH~4 and over a period of 7 hours in presence of liposome, which are almost 4 times slower release than bare mesoporous silica nanoparticles. This indicates that composite system has enough control on the drug release over the conventional drug delivery through bare mesoporous silica nanoparticles. This phenomenon was explained based on that, the ZIF frameworks act as a shield against the external stimuli and protects the bare silica from contact with the external agent and results in slower drug release. But in case of bare silica due to the absence of this kind of protection, drug release becomes very fast under acidic conditions.

Keywords: Drug Delivery System. Mesoporous Nanoparticles. Controlled Release. Composite Materials. pH Triggered Release.
1. Introduction

Most of the chemotherapeutic drugs suffer from uncontrolled and non-specific delivery that lead to lethal effects to the body thus limits their uses.[1] So; severe side effects of chemotherapeutic drugs to the healthy cells is very common and controlled delivery is very essential for chemotherapeutics.[2] To overcome these adverse effects of uncontrolled delivery, we need a delivery vehicle which will deliver the drug molecules in a controlled way with specific target and high efficiency.[3] An ideal drug delivery systems (DDS) should have high biocompatibility, biodegradability, high loading efficiency, ability to prevent drug leakage, control over the release, high efficiency, less side effects etc.[4-5] To achieve these, several DDS have been developed in the last few decades including both soft materials as well as hard materials.[6] Liposome, polymerosome, micelle, dendrimer, polymer capsules are few examples of soft DDS and few of these have already been approved by FDA.[7-9] Although soft DDS show some promising results, however premature drug leakage followed by uncontrolled drug release remain two major problems that motivated the scientists for the development of other DDS based on hard materials.[10] Hard materials primarily consist of inorganic nanoparticles and inorganic-organic hybrid materials including mesoporous silica nanoparticles, gold nanoparticles, silver nanoparticles, iron oxide nanoparticles, quantum dots, carbon nanotubes, metal organic frameworks etc.[11] Out of these materials, mesoporous silica nanoparticles (MSN) and metal organic frameworks (MOFs) have made their own space in biomaterials science. Several pioneer works show that both MSN and MOFs can be used as promising drug delivery system.[12-13]

Mesoporous silica nanoparticles (MSN) are one of the inorganic porous nanomaterials which have been used in biomedical science since early 2000.[14] Due to the easy synthesis, highly porous structure, high surface area, high pore volume, tunable pore size, high drug loading ability, biocompatibility, easy surface modification, MSN becomes one of the favorite choice of the scientists as DDS for
the last few years.\textsuperscript{[15]} There are several reports in the literature which shows that MSN can be used as potential DDS.\textsuperscript{[16-20]} Although conventional MSN have several advantages but MSN suffer from few problems like drug leakage and fast release of the cargo.\textsuperscript{[21]} Several strategies have been adopted to resolve these issues.\textsuperscript{[22]} Improved MSNs were already prepared by either surface functionalization or by conjugation with different inorganic as well as organic molecules including iron oxide, polymers, polyelectrolytes, quantum dots, gold nanoparticles, cyclodextrin, biomolecules, enzymes etc.\textsuperscript{[23-30]} Several different chemotherapeutic drugs e.g. Doxorubicin, camptothecin, paclitaxel, docetaxel, were already delivered using modified MSN.\textsuperscript{[31-34]} In this chapter, we have developed a new strategy to modify the MSN by making a composite with zeolitic imidazole frameworks (ZIFs).

ZIFs are one of the subclasses of metal organic frameworks, composed of zinc as metal ions and imidazole and imidazole derivatives as organic ligands.\textsuperscript{[35]} Due to high surface area, porous structure, tunable shape and size etc. ZIFs have been used in different field of science including gas storage, gas sensing, catalysis, gas separation etc.\textsuperscript{[36-37]} Although, hundreds of ZIFs have been synthesized during the last few years, only few of them went into the application in DDS.\textsuperscript{[38]} Recently, ZIFs have been used as DDS for various types of drugs because of their biocompatibility, biodegradability, huge cargo loading ability etc.\textsuperscript{[39]} Different types of drugs including Doxorubicin, camptothecin, curcumin, etc. were already delivered through ZIFs which indicate promising future of ZIFs as DDS.\textsuperscript{[40-42]} Although these reports show positive results but still this area is in underdevelopment stage, therefore, more research is needed further so that ZIFs can be used efficiently and practically in biomedical science.

Keeping all these points in mind, here in, we developed a new strategy to improve conventional MSN by combining with ZIFs to make a hybrid system (Scheme 7.1). The main objective of this work was to control premature drug release and prevent drug leakage from the MSN which were very common phenomenon with conventional MSN. We used two ZIFs, ZIF-7 and ZIF-8, to form the composite DDS (ZIFs/MSN DDS). We chose these two ZIFs because both the ZIFs were
known to be biocompatible, biodegradable and have been already used for delivery of chemotherapeutic drugs.\textsuperscript{[39]} We used a prominent anticancer drug doxorubicin (DOX) as a model chemotherapeutics to explore this new ZIFs/MSN composite DDS as a new delivery vehicle. At first, DOX was incorporated in MSN following a previously reported post synthetic drug incorporation method.\textsuperscript{[43]} We used this DOX loaded MSN (MSN@DOX) for composite formation during the synthesis of ZIFs. MSN, MSN@DOX and ZIFs-MSN composite (ZIF-8/MSN@DOX & ZIF-7/MSN@DOX) were characterized by Powder X-ray Diffraction, IR spectroscopy, Thermogravimetric analysis, BET analysis, scanning electron microscopy. We have shown the drug release from MSN@DOX as well as from ZIFs/MSN@DOX under external triggering agents. We used acidic pH (pH\texttextsuperscript{--}5.0) as external stimuli because the microenvironment of the cancerous cells are acidic than normal cells.\textsuperscript{[44]} MSN@DOX shows very fast release of the drug that completed by three hours. On the other hand, composite DDS release the drug slowly in a controlled way over a period of 12 hours which is almost 4 times slower release than normal MSN. So, the composite systems have substantial control on the drug release over the conventional MSN. We hypothesized that the ZIF frameworks act as a shield against the external stimuli and protects the bare silica from contact with the external agent and results in slower drug release. But in case of bare silica due to the absence of this kind of protection, drug release becomes very fast under acidic conditions. We also described that the reason behind the drug release is the rupture of the ZIFs frameworks at acidic conditions because of the detachment of the ligand from the metal center which results in destruction of the frameworks.\textsuperscript{[45]} As soon as ZIF framework collapses, bare silica starts to release the drug slowly at acidic pH. Overall the present work highlights a novel strategy for modification of MSN and prevent drug leakage and uncontrolled drug release. We hope this study will help further in the future development of drug delivery system based on composite materials.
Scheme 1: Pictorial representation of (A) DOX loading in MSN (B) ZIF/MSN@DOX composite formation and stimuli responsive controlled release.
2. Results and discussion:

2.1: SEM imaging of the prepared compounds:

After preparation of the ZIFs and ZIF/MSN composites, we took SEM images to understand the morphology of the ZIFs and ZIF/MSN@DOX composites. Figure 7.1 displays the SEM images of ZIFs and ZIF/MSN@DOX composites. It is clear from the images that the size of ZIF-8 and its composites are close to 1 μM and the size of the ZIF-7 and its composites are close to 200 nm.

![SEM images](image_url)

Figure 1: SEM images of (a) MSN (b) ZIF-8 (c) ZIF-8/MSN (d) ZIF-7 (e) ZIF-7 MSN.
2.2: **Confocal laser scanning microscopy imaging:**

Figure 7.2 shows confocal laser scanning microscopy images of ZIF-8, ZIF-8/MSN, ZIF-7 and ZIF-7/MSN respectively. We used a well-known dye rhodamine-B (Rh-B) for the staining of the compounds. Rh-B absorbs at 540 nm and it gives emission at 640 nm. We used an excitation laser of 559 nm and collected the emission at 625 nm. Figure 7.2 (a & b) show that the size of the particles were ~1 μM and figure 7.2 (c & d) show that the size of the particles were varies from 200 nm to 300 nm. From the images it is clear that the particles have almost equal size distribution, and few are larger due to the aggregation.

**Figure 2:** Confocal laser scanning microscopy images of (a) ZIF-8 (b) ZIF-8/MSN (c) ZIF-7 (d) ZIF-7/MSN.
2.3: **Bright field imaging:**

We also performed bright field imaging of the compounds along with CLSM images and the figure 7.3 shows that the spherical morphology of the compounds which corroborates with the SEM as well as CLSM data.

![Bright field images of (a) ZIF-8 (b) ZIF-8/MSN (c) ZIF-7 (d) ZIF-7/MSN.](image)

**Figure 3:** Bright field images of (a) ZIF-8 (b) ZIF-8/MSN (c) ZIF-7 (d) ZIF-7/MSN.

2.4: **Surface area and pore size analysis of the ZIFs and composites:**

After morphological characterization, we analyze pore properties and surface area of the ZIFs and ZIF-silica composites using nitrogen adsorption-desorption method. Figure 7.4 depicts the nitrogen adsorption-desorption isotherms of ZIF-7, ZIF-7/Silica, ZIF-8 and ZIF-8/silica. The figure shows a type IV isotherm for all the compounds which is typical for mesoporous compounds.\[46\] The Langmuir surface area and BJH (Barrett-Joyner-Halenda) pore diameter and pore volume are summarized in the table 7.4.1.
Figure 4: Nitrogen sorption isotherms of (a) ZIF-7 (b) ZIF-7/Silica (c) ZIF-8 (d) ZIF-8/Silica.

Table: 1

| Compounds     | Pore radius (Å) | Pore diameter (Å) | Pore volume (cc/g) | Surface area [Langmuir] (m²/g) |
|---------------|-----------------|-------------------|--------------------|---------------------------------|
| ZIF-7         | 31.27           | 62.54             | 0.163              | 156.43                          |
| ZIF-7/Silica  | 19.62           | 39.24             | 0.173              | 587.15                          |
| ZIF-8         | 19.68           | 39.36             | 0.299              | 1733.48                         |
| ZIF-8/Silica  | 15.67           | 31.34             | 0.107              | 3025.97                         |

The table reveals that the surface area is higher in case of composite system compared to bare ZIFs for both the cases. This is due to the presence of nanoscaled mesoporous silica nanoparticles into the ZIFs. This data also
supports the formation of the ZIF-silica composite. Another point that needs to be addressed here is that ZIF-7 possesses very low surface area compared to ZIF-8. This is because of the presence of larger pore and hollow cavities in ZIF-7 which lower the surface area.\[47-48\]

2.5: Drug encapsulation in ZIFs and FT-IR studies:

As mentioned in section 7.1 that our aim was not only to synthesize the compounds but also to see the possibility to use them as DDS. For that reason, first we encapsulated DOX in MSN using a previously reported method and the drug loading efficiency was found more than 90% [chapter 5, section 5.2.2, Fig: 5.2]. We used MSN@DOX to form composite with ZIFs. We further characterized ZIFs and composite by IR spectroscopy. Figure 7.5 (a & b) shows IR spectra for ZIF-8, ZIF-8/MSN@DOX, ZIF-7 and ZIF-7/MSN@DOX respectively. The peaks around 1140, 1307, 1383, 1628 cm\(^{-1}\) correspond to N–H of NH\(^{3+}\), C–O, C–O of ether group, whereas the peaks around 1722, 2361, and 3450 cm\(^{-1}\) arises due to C=C, C=O, O–H (carboxylic acid) and O–H of DOX, respectively. The peak comes around 2928 cm\(^{-1}\) due to the presence of N–H group of imidazole of the ZIF-8 and ZIF-7 framework.\[39\]

**Figure 5: IR spectra of (a) ZIF-7 and ZIF-7/Silica (b) ZIF-8 and ZIF-8/Silica**

2.6: Thermogravimetric analysis:

Thermogravimetric analysis was performed to gain an insight into the thermal stability of the compounds. Figure 7.6 shows TGA curve for ZIF-8, ZIF-8 silica, ZIF-7 and ZIF-7 silica. TGA analysis of MSN and MSN@DOX were already
investigated in our earlier study (chapter 5, section 5.2.4, Fig: 5.4). Figure 7.6a reveals that ZIF-8 and ZIF-8/MSN@DOX are stable up to 450 °C. The first weight loss (~15%) in case of ZIF-8/MSN@DOX is due to desorption of physisorbed water molecules on the external surface of mesoporous silica along with the decomposition of organic component i.e. DOX. Figure 7.6 depicts that ZIF-7 and ZIF-7/MSN@DOX are stable up to 560 °C. The little weight loss at the beginning corresponds to the decomposition of organic molecules along with the physisorbed water molecules. The higher thermal stability of the ZIF-7 compared to ZIF-8 is due to the more rigidity of the ZIF-7 framework than ZIF-8.\(^{[39]}\) In both the cases a sharp weight loss (~ 70-80 %) was observed for beyond 600 °C. This weight loss indicates thermal decomposition of the ZIF framework to the corresponding metal oxide at that temperature range.

Figure 7.7 shows PXRD pattern of MSN, ZIF-8, ZIF-8/MSN@DOX, ZIF-7 and ZIF-7/MSN@DOX. The PXRD pattern of pure MSN, ZIF-7 and ZIF-8 matches with the earlier reports.\(^{[49-50]}\) The sharp peak of PXRD pattern indicates narrow size distribution of the synthesized ZIFs.

Therefore, all the above mentioned experiments (CLSM, Bright field, BET, IR, TGA, and PXRD) indicates successful formation of the composites and in the due course of this chapter we will demonstrate the release of the drug molecules under suitable external stimuli for a certain period of time.

Figure 6: TGA curve for (a) ZIF-8 and ZIF-8/MSN@DOX (b) ZIF-7 and ZIF-7/MSN@DOX
2.7: Release of drug molecules under pH stimuli:

After successful characterization of the compounds, we further explored these composites as stimuli responsive DDS for DOX. We choose acidic pH as an external stimulus due to acidic nature of the cancerous cells compared to normal cells. \[51-53\] At first, we studied the drug release from MSN@DOX at pH~4. Certain amount of solid MSN@DOX was immersed into a phosphate buffer of pH~4 and the drug release was monitored by fluorescence emission of the supernatant at different time interval. Figure 7.8a shows emission spectra of DOX at different time varying from 0-2 hours. The gradual increase in the intensity implies the release of drug molecules from the DDS. It is clear from the figure that the drug release was completed within 2 hours with no control. The reasons behind the uncontrolled and fast release from bare MSN were (a) tendency of DOX to come in to the aqueous medium because of higher solubility of DOX in water (b) affinity to form cationic species at lower pH as the pKa of DOX is ~8.\[54\] Figure 7.8 (b & c) show emission spectra of DOX at time varying from 0-14 hours in case of ZIFs/MSN@DOX composite. The results clearly indicate a slow and controlled drug release. The probable reason behind the controlled drug release is that the ZIFs shield bare MSN and hence
protects MSN@DOX to come direct contact with the acidic buffer. As soon as ZIFs framework starts to collapse in acidic pH, \cite{41} MSN@DOX releases the drug molecules slowly and the drug release become controlled. So; the above results clearly indicates that bare MSN fails to control the release of the drug whereas composite system can slower the drug release by six times and the drug release is absolutely controlled here for a period of 20 hours thus making the composite system a superior DDS compared to bare MSN.

**Figure 8:** Fluorescence emission spectra of DOX at pH~4 from (a) MSN@DOX at time varying from 0-2 hours (b) ZIF-8/MSN@DOX (c) ZIF-7/MSN@DOX at tie varying from 0-14 hours (d) Comparison of % of Drug release [in the inset expanded region upto 400 minutes is shown].
2.8: Drug release in presence of biomimetic membrane (liposomes):

Once we studied the pH triggered release, we were keen to understand how the DDS would behave in presence of biomimetic membrane. To explore this we used liposome because liposomes were known as biomimetic membrane for a long time.[55-56] DOX, due to its pKa ~8, remain positively charged at pH lower than 8, so; it is quite expected that in the vicinity of liposomes, DOX would like to stay as positively charged. Because of this we used a negatively charged liposomes composed of DMPC/DMPG (8:2) to facilitate drug release with the help of electrostatic interaction between DOX and liposomes. Figure 7.9 shows the emission spectra of DOX in presence of DMPC/DMPG liposomes. It is clear from the figure 7.9a that the drug release completed within 2 hours in case of MSN@DOX whereas the release has been showered by four times in case of composite system. The probable reasons behind the drug release in presence of liposomes were (a) acidic microenvironment of the liposomes (b) electrostatic interaction of DOX and negatively charged liposomes (c) binding of DOX in the lipid bilayer.[57] These data also supports the phenomenon that the composite system are able to control drug release over a certain period of time unlike bare MSN which has no control over the drug release.
Figure 9: Fluorescence emission spectra of DOX in presence of DMPC/DMPG liposomes (a) MSN@DOX at time varying from 0-2 hours (b) ZIF-8/MSN@DOX (c) ZIF-7/MSN@DOX at tie varying from 0-7 hours.
3: Conclusion:

In conclusion, we have successfully developed a hybrid DDS composed of ZIFs/MSN composite at room temperature employing mild reaction conditions. The hybrid DDS were stimuli responsive and able to release DOX in a controlled manner over a certain period of time under external triggering agents. We found that bare MSN have no control over the drug release whereas composite system can control the release. We explained this phenomenon by the fact that ZIF framework protects the MSN@DOX from the direct contact with the external stimuli by acting as a shield thus controlled the release. The liposome mediated drug release was due to the electrostatic binding of DOX in the lipid bilayer. Overall the present work shows a promising DDS and hope will help in the future development of the DDS based on hybrid materials.
4: Experimental section:

4.1: Materials:
Doxorubicin (DOX), zinc nitrate hexahydrate, 2-methyl imidazole, benzimidazole, dimethyl formamide, mesoporous silica nanospheres (MSN) were purchased from Sigma-Aldrich. Monosodium hydrogen phosphate, disodium hydrogen phosphate, were purchased from Merck and methanol was purchased from Spectrochem. All the chemicals were used without further purification. Deionized (DI) water from a Millipore water (18 MO•cm) purification system was used for all the experiments.

4.2: Preparation of ZIF-7:
ZIF-7 was prepared following a previously reported protocol.[39] In brief, for a typical synthesis of ZIF-7, 100 ml dimethyl formamide (DMF) was taken in a 100 ml round bottom flask (RBF). Then zinc nitrate hexahydrate (3 g) and benzimidazole (10 g) were added to the RBF while continuous stirring. The stirring was continued for another 48 hours. The product was collected by centrifugation (10000 rpm for 15 minutes) and washed with DMF for five times and with methanol for three times to completely remove the unreacted reactants. The product was dried in oven at 100 °C for overnight. The product was kept at room temperature for further experiments.

4.3: Preparation of ZIF-8:
ZIF-8 was prepared following a previously reported protocol with slight modification.[39] For a typical synthesis of ZIF-8, zinc nitrate hexahydrate (600 mg) was dissolved by magnetic stirring in 50 ml of methanol in a 100 ml RBF. In another RBF, 2-methyl imidazole (1.3 g) was dissolved in 50 ml of methanol by hand shaking for 10 minutes. Now, 2 methyl imidazole solution was added to zinc nitrate solution with constant stirring. The reaction was continued for 30 minutes. Product was collected by centrifugation (10000 rpm for 10 minutes) with three times washing with fresh methanol to remove unreacted reactants.
Finally, the product was dried in an oven at 100 °C for overnight. The dried product was kept at room temperature for further experiments.

4.4: Drug loading in MSN:

We followed a previously reported post synthetic drug loading method for the loading of DOX in MSN.\[43\] Briefly, 3g of MSN was taken in a vial and 5 ml of DOX (2 mg/ml) solution was added to it with stirring. The stirring was continued for another 72 hours. Drug loaded MSN (MSN@DOX) was separated by centrifugation (10000 rpm for 10 minutes) and washed five times with water to remove the loosely bound surface attached DOX molecules. MSN@DOX was dried at high vacuum for 24 hours in a dark place (oven drying was avoided as there is a possibility for degradation of DOX at high temperature). The product was kept at room temperature for doing further studies. We used fluorescence spectroscopy to understand drug loading and to calculate efficiency of drug loading (See results and discussion section for more details).

4.5: Preparation of ZIF-8/MSN@DOX:

For ZIF-8/MSN@DOX composite preparation, MSN@DOX (1.5 g) was taken in a RBF and dispersed in 20 ml methanol. Zinc nitrate hexahydrate (600 mg) was added to this and dissolved completely by magnetic stirring for 10 minutes. 2-methyl imidazole (1.3 g) was dissolved in 20 ml methanol. Now, 2-methyl imidazole solution was added to zinc nitrate hexahydrate/MSN@DOX solution with continuous stirring. The stirring was continued for 30 minutes. The product was collected by centrifugation (5000 rpm for 10 minutes) and washed with methanol to remove unreacted reactants. The product was dried at high vacuum (oven drying was avoided as there was a chance for degradation of DOX at high temperature) for two days and kept at room temperature for further experiments.
4.6: **Preparation of ZIF-7/MSN@DOX:**

We prepared ZIF-7/MSN@DOX in the same fashion as in the case of ZIF-8/MSN@DOX. Briefly, MSN@DOX (1.5 g) was taken in a RBF and dispersed in 20 ml methanol. Zinc nitrate hexahydrate (600 mg) was added to this and dissolved completely by magnetic stirring for 10 minutes. Then, benzimidazole (2 g) was added to this solution while constant stirring. The stirring was continued for 48 hours. The product was separated by centrifugation followed by washing with DMF followed by methanol. The composite was dried at high vacuum and kept at room temperature for further studies.

4.7: **Preparation of phosphate buffer of pH~5.0:**

Phosphate buffer of pH 5.0 was prepared by dissolving required amount of monosodium hydrogen phosphate and disodium hydrogen phosphate in Milli Q water. The pH of the buffer was adjusted by pH meter by adding 0.1 N of HCl and 0.1 N of NaOH solution. We followed the same procedure for the preparation of buffer solution of other pH also.

4.8: **In vitro drug release study:**

For in vitro drug release study, three hundred mg of the drug loaded MSN and composite (ZIF-8/MSN@DOX, ZIF-7/MSN@DOX) was immersed into five mL solution of pH~5.0 in an round bottom flask and also the supernatant was collected from time to time. Steadystate visible spectrometry was wont to monitor the drug unharness. The visible radiation emission of the supernatant was measured from time to time. The gradual increase within the visible radiation intensity provides a transparent indication that the drug molecules are taking off from the MSN/composite. We tend to determined that just in case of MSN, drug unharness completed over a period of three hours whereas composite will unharness the medicine over 1 day.

4.9: **Dye staining for confocal laser scanning microscopy imaging:**

For confocal imaging, we tend to selected a wide used dye rhodamine B for staining the compounds. one hundred mg of the compounds were immersed into two mL of one hundred μM dye resolution with constant stirring. The stirring was continued for three
days in a dark place. Dye stained compounds were separated by natural action (7000 revolutions per minute for fifteen minutes. The compounds were dried in high vacuum and unbroken at temperature.

4.10: Instrumentation:

We used field emission scanning electron microscopy (FE-SEM), energy-dispersive X-ray spectroscopy (EDX), confocal laser scanning microscopy, bright field microscopy, infrared spectroscopy, powder X-ray diffraction, and thermogravimetric analysis to characterize all the synthesized compounds. FE-SEM study was conducted in ZEISS Supra 55 field emission scanning electron microscopies (FE-SEM). We used confocal laser scanning microscopy (CLSM) from Olympus (model no: IX83, multiple Ar laser, XY LSM) to study the morphology of the compounds. The wavelength of excitation laser was 559 nm and the emission were collected at 625 nm. IR spectra were taken in a Fourier Transform Infrared Spectrometer (FT-IR), Tensor 27, BRUKER. Powder X-ray diffraction was done in an Automated Multipurpose X-ray Diffractometer from Rigaku SmartLab, X-ray generator: A 3 kW sealed tube X-ray generator (Max. voltage 60kV, Max. current 50mA, with Cu target). Thermogravimetric analysis was done in a TGA instrument from Mettler Toledo, Switzerland, Model: TGA/DSC1. TGA analysis was done under nitrogen flow and the heating rate was 5 °C per minute. State fluorescence spectroscopy using a Fluoromax-4p spectrofluorimeter from Horiba Jobin Yvon (Model: FM-100) was employed to monitor the drug release from the MSN and ZIF/MSN composite.
5: References:

[1] Mark W. T.; James E. D.; Robert L. (2016), Emerging Frontiers in Drug Delivery, *J. Am. Chem. Soc.* 138, 704-717 (DOI: 10.1021/jacs.5b09974).

[2] Gulzar, A.; Gai, S.; Yang, P.; Li, C.; Ansari, B. M.; Lin, J. (2015), Stimuli responsive drug delivery application of polymer and silica in biomedicine, *J. Mater. Chem. B*, 3, 8599-8622 (DOI: 10.1039/C5TB00757G).

[3] Luo, Y. L.; Shiao, Y. S; Huang, Y. F. (2011), Release of Photoactivatable Drugs from Plasmonic Nanoparticles for Targeted Cancer Therapy, *ACS Nano*, 5, 7796–7804 (DOI: 10.1021/nn201592s).

[4] Mai, W. X.; Meng, H. (2013), Mesoporous silica nanoparticles: A multifunctional nano therapeutical system, *Integr. Biol.* 5, 19-28 (DOI: 10.1039/C2IB20137B).

[5] Yih, T. C.; Al-Fandi, M.; Cell. J. (2006), Engineered nanoparticles as precise drug delivery systems, *Biochem.* 97, 1184–1190 (DOI: 10.1002/jcb.20796).

[6] Kamaly, N.; Xiao, Z.; Valencia, P. M. Radovic-Moreno A.F.; Farokhzad O. C. (2012), Targeted polymeric therapeutic nanoparticles: design, development and clinical translation, *Chem Soc Rev.* 7, 2971-3010 (DOI: 10.1039/C2CS15344K).

[7] Farokhzad, O. C.; Langer, R. (2009), Impact of Nanotechnology on Drug Delivery, *ACS Nano*. 3, 16-20 (DOI: 10.1021/nn900002m).

[8] Du, J. Z.; Du, X. J.; Mao, C. Q.; Wang, J. (2011), Tailor-Made Dual pH-Sensitive Polymer–DOXorubicin Nanoparticles for Efficient Anticancer Drug Delivery, *J. Am. Chem. Soc.* 133, 17560-17563 (DOI: 10.1021/ja207150n).

[9] Hu, X.; Hu, J.; Tian, J.; Ge, Z.; Zhang, G.; Luo, K.; Liu, S. (2013), Polyprodrug Amphiphiles: Hierarchical Assemblies for Shape-
Regulated Cellular Internalization, Trafficking, and Drug Delivery, *J. Am. Chem. Soc.* 135, 17617-17629 (DOI: 10.1021/ja409686x).

[10] Maurer, N.; Fenske, D. B.; Cullis, P. R. (2001), Developments in liposomal drug delivery systems, *Expert Opin. Biol. Ther.* 1, 923–947 (DOI: http://dx.doi.org/10.1517/14712598.1.6.923).

[11] Ang, C. Y.; Tan, S. Y.; Zhao, Y. (2014), Recent advances in biocompatible nanocarriers for delivery of chemotherapeutic cargoes towards cancer therapy, *Org. Biomol. Chem.* 12, 4776 (DOI: 10.1039/C4OB00164H).

[12] Horcajada, P.; Gref, R.; Baati, T.; Allan, P. K.; Maurin, G.; Couvreur, P.; Férey, G.; Morris, R. E.; Serre, C. (2012) Metal–Organic Frameworks in Biomedicine, *Chem. Rev.* 112, 1232–1268 (DOI: 10.1021/cr200256v).

[13] Yang, P.; Gaib, S.; Lin, J. (2012), Functionalized mesoporous silica materials for controlled drug delivery, *Chem. Soc. Rev.* 41, 3679–3698 (DOI: 10.1039/C2CS15308D).

[14] Kresge, C. T.; Leonowicz, M. E.; Roth, W. J.; Vartuli, J. C.; Beck, J. S. (1992), Ordered mesoporous molecular sieves synthesized by a liquid-crystal template mechanism, *Nature*, 359, 710–712 (DOI: 10.1038/359710a0).

[15] Colilla, M.; Gonzáleza, B.; Vallet-Regí, M. Mesoporous silica nanoparticles for the design of smart delivery nanodevices, *Biomater. Sci.* 2013, 1, 114–134 (DOI: 10.1039/C2BM00085G).

[16] Hoffmann, F.; Cornelius, M.; Morell, J.; Froba, M. (2006), Ordered mesoporous molecular sieves synthesized by a liquid-crystal template mechanism, *Angew. Chem. Int. Ed.* 45, 3216–3251 (DOI: 10.1038/359710a0).

[17] Ambrogio, M. W.; Thomas, C. R.; Zhao, Y. L.; Zink, J. I. Stoddart, J. F. (2011), Mechanized Silica Nanoparticles: A New Frontier in Theranostic Nanomedicine, *Acc. Chem. Res.* 44, 903–913 (DOI: 10.1021/ar200018x).
[18] Yang, P.; Gai, S.; Lin, J. (2012), Functionalized mesoporous silica materials for controlled drug delivery, *Chem. Soc. Rev.* 41, 3679-3698 (DOI: 10.1039/C2CS15308D).

[19] Hu, J. J.; Liu, L. H.; Li, Z. Y.; Zhuo, R. X.; Zhang, X. Z. (2016), MMP-responsive theranostic nanoplatform based on mesoporous silica nanoparticles for tumor imaging and targeted drug delivery, *J. Mater. Chem. B*, 4, 1932-1940 (DOI: 10.1039/C5TB02490K).

[20] Taghavi, F.; Gholizadeh, M.; Saljooghi, A. S. (2016), Deferasirox loaded on fumed silica nanoparticles used in cancer treatment, *New J. Chem.* 40, 2696-2703 (DOI: 10.1039/C5NJ02790J).

[21] Li, Z.; Barnes, J. C.; Bosoy, A.; Stoddart, J. F.; Zink, J. I. (2012), Mesoporous silica nanoparticles in biomedical applications, *Chem. Soc. Rev.* 41, 2590-2605 (DOI: 10.1039/C1CS15246G).

[22] Kobler, J.; Möller, K.; Bein, T. (2008), Colloidal Suspensions of Functionalized Mesoporous Silica Nanoparticles, *ACS Nano*, 2, 791–799 (DOI: 10.1021/nn700008s).

[23] Lai, C. Y.; Trewyn, B. G.; Jeftinija, D. M.; Jeftinija, K.; Xu, S.; Jeftinija, S.; Lin, V. S. Y. (2003), A Mesoporous Silica Nanosphere-Based Carrier System with Chemically Removable CdS Nanoparticle Caps for Stimuli-Responsive Controlled Release of Neurotransmitters and Drug Molecules, *J. Am. Chem. Soc.* 125, 4451–4459 (DOI: 10.1021/ja028650l).

[24] Huang, S.; Yang, P.; Cheng, Z.; Li, C.; Fan, Y.; Kong, D.; Lin, J. (2008), Synthesis and Characterization of Magnetic FexOy@SBA-15 Composites with Different Morphologies for Controlled Drug Release and Targeting, *J. Phys. Chem. C*. 112, 7130–7137 (DOI: 10.1021/jp800363s).

[25] Zhu, Y.; Shi, J.; Shen, W.; Dong, X.; Feng, J.; Ruan, M.; Li, Y. (2005), Stimuli-responsive controlled drug release from a hollow mesoporous silica sphere/polyelectrolyte multilayer core-shell structure, *Angew. Chem. Int. Ed.* 44, 5083–5087 (DOI: 10.1002/ange.200501500).
[26] Liu, C.; Guo, J.; Yang, W.; Hu, J.; Wang, C.; Fu, S. (2009), Magnetic mesoporous silica microspheres with thermo-sensitive polymer shell for controlled drug release, *J. Mater. Chem.* 19, 4764–4770 (DOI: 10.1039/B902985K).

[27] Ruiz-Hernà, E.; Baeza, A.; Vallet-Regí, M. (2011), Smart Drug Delivery through DNA/Magnetic Nanoparticle Gates, *ACS Nano.* 2011, 5, 1259–1266 (DOI: 10.1021/nn1029229).

[28] Patel, K.; Angelos, S.; Dichtel, W. R.; Coskun, A.; Yang, Y. W.; Zink, J. I.; Stoddart, J. F. (2008), Enzyme-Responsive Snap-Top Covered Silica Nanocontainers, *J. Am. Chem. Soc.*, 130, 2382–2383 (DOI: 10.1021/ja0772086).

[29] Aznar, E.; Dolores Marcos, M.; Martinez Manez, R.; Sancenon, F.; Soto, J.; Amoros, P.; Guillem, C. (2009), pH- and Photo-Switched Release of Guest Molecules from Mesoporous Silica Supports, *J. Am. Chem. Soc.*, 131, 6833–6843 (DOI: 10.1021/ja810011p).

[30] Zhao, W.; Zhang, H.; He, Q.; Li, Y.; Gu, J.; Li, L.; Li. H.; Shi, J. (2011), A glucose-responsive controlled release of insulin system based on enzyme multilayers-coated mesoporous silica particles, *Chem. Commun.*, 47, 9459–9461 (DOI: 10.1039/C1CC12740C).

[31] Zhu, Y.; Ikoma, T.; Hanagata, N.; Kaskel, S. (2010), Rattle-Type Fe3O4@SiO2 Hollow Mesoporous Spheres as Carriers for Drug Delivery, *Small.* 6, 471–478 (DOI: 10.1002/smll.200901403).

[32] Meng, H.; Xue, M.; Xia, T.; Zhao, Y. L.; Tamanoi, F.; Stoddart, J. F.; Zink, J. I.; Nel, A. E. (2010), Autonomous in Vitro Anticancer Drug Release from Mesoporous Silica Nanoparticles by pH-Sensitive Nanovalves, *J. Am. Chem. Soc.*, 132, 12690–12697 (DOI: 10.1021/ja104501a).

[33] Liong, M.; Lu, J.; Kovochich, M.; Xia, T.; Ruehm, S. G.; Nel, A. E.; Tamanoi, F.; Zink, J. I. (2008), Multifunctional Inorganic Nanoparticles for Imaging, Targeting, and Drug Delivery, *ACS Nano.* 2, 889–896 (DOI: 10.1021/nn800072t).
[34] Wu, H.; Liu, G.; Zhang, S.; Shi, J.; Zhang, L.; Chen, Y.; Chen, F.; Chen, H. (2011), Biocompatibility, MR imaging and targeted drug delivery of a rattle-type magnetic mesoporous silica nanosphere system conjugated with PEG and cancer-cell-specific ligands, *J. Mater. Chem.*, 21, 3037–3045 (DOI: 10.1039/C0JM02863K).

[35] Wang, B.; Côte, A. P.; Furukawa, H.; O’Keeffe, M.; Yaghi, O. M. (2008), Colossal cages in zeolitic imidazolate frameworks as selective carbon dioxide reservoirs. *Nature*, 453, 207–211 (DOI: 10.1038/nature06900).

[36] Venna, S. R.; Carreon, M. A. (2010), Highly Permeable Zeolite Imidazolate Framework-8 Membranes for CO2/CH4 Separation. *J. Am. Chem. Soc.*, 132, 76–78 (DOI: 10.1021/ja909263x).

[37] Jiang, H. L.; Liu, B.; Akita, T.; Haruta, M.; Sakurai, H.; Xu, Q. (2009), Au@ZIF-8: CO Oxidation over Gold Nanoparticles Deposited to Metal–Organic Framework. *J. Am. Chem. Soc.*, 131, 11302–11303 (DOI: 10.1021/ja9047653).

[38] Keskin, S.; Kızılcel, S. (2011), Biomedical Applications of Metal Organic Frameworks, *Ind. Eng. Chem. Res.*, 50, 1799–1812 (DOI: 10.1021/ie101312k).

[39] Adhikari, C.; Das, A.; Chakraborty, A. (2015), Zeolitic Imidazole Framework (ZIF) Nanospheres for Easy Encapsulation and Controlled Release of an Anticancer Drug DOXorubicin under Different External Stimuli: A Way toward Smart Drug Delivery System, *Mol. Pharmaceutics*. 12, 3158–3166 (DOI: 10.1021/acs.molpharmaceut.5b00043).

[40] Vasconcelos, I. B.; da Silva, T. G.; Militao, G. C. G.; Soares, T. A.; Rodrigues, N. M.; Rodrigues, M. O.; da Costa, N. B., Jr.; Freire, R. O.; Junior, S. A. (2012), Cytotoxicity and slow release of the anti-cancer drug DOX from ZIF-8. *RSC Adv.* 2, 9437–9442 (DOI: 10.1039/C2RA21087H).
[41] Zhuang, J.; Kuo, C. H.; Chou, L. Y.; Liu, D. Y.; Weerapana, E.; Tsung, C. K. (2014), Optimized Metal Organic-Framework Nanospheres for Drug Delivery: Evaluation of Small-Molecule Encapsulation. ACS Nano. 8, 2812–2819 (DOI: 10.1021/nn406590q).

[42] Zheng, M.; Liu, S.; Guan, X.; Xie, Z. (2014), One-Step Synthesis of Nanoscale Zeolitic Imidazolate Frameworks with High Curcumin Loading for Treatment of Cervical Cancer, ACS Nano. 2014, 8, 2812–2819 (DOI: 10.1021/acsami.5b04315).

[43] Shen, J.; Qianjun. H.; Gao, Y.; Jianlin, S.; Yaping, L. (2011), Mesoporous silica nanoparticles loading DOX reverse multidrug resistance: performance and mechanism. Nanoscale, 3, 4314–4322 (DOI: 10.1039/C1NR10580A).

[44] Gulzar, A.; Gai, S.; Yang, P.; Li, C.; Ansaric, M. B.; Lin, J. (2015), Stimuli responsive drug delivery application of polymer and silica in biomedicine, J. Mater. Chem. B, 3, 8599-8622 (DOI: 10.1039/C5TB00757G).

[45] Zhuang, J.; Kuo, C. H.; Chou, L. Y.; Liu, D. Y.; Weerapana, E.; Tsung, C. K. (2014), Optimized Metal Organic-Framework Nanospheres for Drug Delivery: Evaluation of Small-Molecule Encapsulation. ACS Nano. 8, 2812–2819 (DOI: 10.1021/nn406590q).

[46] Zhang, Y.; Zhou, G.; Sun, B.; Zhao, M.; Zhang, J.; Chen, F. (2014), A cationic–cationic co-surfactant templating route for synthesizing well-defined multilamellar vesicular silica with an adjustable number of layers, Chem. Comm. 50, 2907-2909 (DOI: 10.1039/C3CC49511F).

[47] Li, X.; Liu, X.; Ma, Y.; Li, M.; Zhao, J.; Xin, H.; Zhang, L.; Yang, Y.; Li, C.; Yang, Q. (2012), Engineering the Formation of Secondary Building Blocks within Hollow Interiors, Adv. Mater. 24, 1424-1428 (DOI: 10.1002/adma.201104167).

[48] Zhang, L. X.; Li, P. C.; Liu, X. H.; Du, L. W.; Wang, E. K. (DOI: 2007), The Effect of Template Phase on the Structures of As-Synthesized Silica Nanoparticles with Fragile
Didodecyldimethylammonium Bromide Vesicles as Templates, *Adv. Mater.* 19, 4279-4282 (DOI: 10.1002/adma.200701228).

[49] Li, Y.-S.; Liang, F.-Y.; Bux, H.; Feldhoff, A.; Yang, W. S.; Caro, J. (2010), Molecular Sieve Membrane: Supported Metal–Organic Framework with High Hydrogen Selectivity. *Angew. Chem., Int. Ed.* 2010, 49, 548–551 (DOI: 10.1002/anie.200905645).

[50] Venna, S. R.; Jasinski, J. B.; Carreon, M. A. (2010), Structural Evolution of Zeolitic Imidazolate Framework-8. *J. Am. Chem. Soc.* 2010, 132, 18030–18033 (DOI: 10.1021/ja109268m).

[51] Read, N. W.; Sugden, K. (1987), Gastrointestinal dynamics and pharmacology for the optimum design of controlled-release oral dosage forms. *Critic. Review. Ther. Drug. Car. Syst.*, 4, 221-263 (PMID: 3276406).

[52] Kararli, T.T. (1995), Comparison of the gastrointestinal anatomy, physiology, and biochemistry of humans and commonly used laboratory animals. *Biopharma. Drug. Dispos.* 16, 351-380 (PMID: 8527686).

[53] Schäfer-Korting, M.; Mehnert, W.; Korting, H. C. (2007), Lipid nanoparticles for improved topical application of drugs for skin diseases. *Adv. Drug. Deliv. Rev.*, 59, 427-443 (DOI: http://dx.doi.org/10.1016/j.addr.2007.04.006).

[54] Anand, R., Borghi, F., Manoli, F., Manet, I., Agostoni, V., Reschiglian, P., Gref, R. and Monti, S. (2014), Host–Guest Interactions in Fe (III)-Trimesate MOF Nanoparticles Loaded with DOXorubicin. *J. Phys. Chem. B.* 118, 8532-8539 (DOI: 10.1021/jp503809w).

[55] Bhattacharya, S.; Bajaj, A. (2007), Membrane-forming properties of pseudoglyceryl backbone based gemini lipids possessing oxyethylene spacers. *J. Phys. Chem. B.* 111, 2463-2472 (DOI: 10.1021/jp068383w).

[56] Bhattacharya, S.; Biswas, J. (2009), Understanding membranes through the molecular design of lipids. *Langmuir*, 26, 4642-4654 (DOI: 10.1021/la9011718).
[57] Crommelin, D. J. A.; Van Bloois, L. (1983) Preparation and characterization of DOXorubicin-containing liposomes. II. Loading capacity, long-term stability and DOXorubicin-bilayer interaction mechanism. Int. J. Pharma., 17, 135-144 (DOI: 10.1016/0378-5173(83)90027-3).