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OPTIMIZATION OF SOL-GEL HYBRID MEDIATED HOLLOW FIBER-SOLID PHASE MICROEXTRACTION FOR THE ANALYSIS OF SELECTED ALDEHYDES

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Abstract. Hexanal and heptanal are two promising aldehydes that have been found in biological samples of lung cancer patients. In general, aldehydes can be detected in trace level as low as part per billion or part per trillion levels. Since reliable technique is needed to detect the analyte of interest especially in complex matrices, a new hybrid silica-based material as the adsorbent of hollow fiber-solid phase microextraction (HF-SPME) was synthesized. In the present work, the sol-gel hybrid material was held in the pores of HF by in situ gelation and was used to extract the two test aldehydes using direct immersion sampling mode. Several sol-gel parameters affecting the extraction performance of the in-house sol-gel hybrid material were optimized. The extraction efficiencies of the new silica-based hybrid material for the target aldehydes were based on peak area response obtained from gas chromatography-flame ionization detector. The best extraction efficiencies of hexanal and heptanal were obtained using a mol ratio of 1:4 for the two precursors used, 6 mmol water, 6 mmol methanol and 2.5 mmol trifluoroacetic acid for in situ gelation HF-SPME. The new in situ sol-gel silica-based adsorbent material exhibits high potential for HF-SPME of hexanal and heptanal.

Keywords Sol gel hybrid, adsorbent, hollow fiber-solid phase microextraction, aldehydes, gas chromatography-flame ionization detector

1.0 INTRODUCTION

Lung cancer is ranked as the number one killer cancer in Malaysia, and worldwide. To diagnosis lung cancer, a few tests can be carried out such as chest X-ray, biopsy, a sputum test and pulmonary function test. Lung cancer diagnosis which relies upon volatile organic compounds (VOCs) is an alternative approach which offers noninvasive, simple, inexpensive, and easy-to-use diagnostic tool [1]. Alkanes, benzene derivatives and aldehydes were among the 28 VOCs biomarker that can be found in lung cancer [2, 3]. Hexanal and heptanal are common aldehydes found in breath [4, 5] and biological samples [6, 7] of lung cancer patients. The two aldehydes can be detected in elevated levels in lung cancer patients as compared to healthy human.

The presence of VOCs at trace levels is cumbersome. Owing to the complexity of sample matrices and relatively low concentration of these biomarker compounds, sample clean-up and enrichment procedure are needed to improve the sensitivity of method. The well-known extraction method used to analyse the biomarker compounds of lung cancer is solid phase microextraction (SPME) [4-7]. The common SPME fibers used for analysis of VOCs of lung cancer are polydimethylsiloxane/divinylbenzene (PDMS/DVB) [4, 6, 7] and carboxen-polydimethylsiloxane (CAR-PDMS) [5]. SPME offers the advantage of one step extraction and preconcentration which is rapid for sampling. However, SPME has certain limitations such as the commercially available fibers are not chemically stable due to by mere physical deposition or partial crosslinking of the polymer coating. In addition, the ordinary SPME fibers suffer from low recommended operating temperature, fragile, limited lifetime and expensive [8].

Sol-gel process is simple as compared to the other sorbent synthesis techniques. Some inherent advantages of sol-gel process are better homogeneity and purity of materials, preparation can be done at ambient temperature, possibility of creating hybrid organic-inorganic materials and capable to achieve enhanced stationary phase stability [9]. On the other hand, the use of sol-gel hybrid materials proved to be very useful for ultra-trace analysis [10-12] and since endless materials can be produced using sol-gel technique, the material produced is expected to overcome the limitations observed by commercial sorbents.

Introducing extracting phase of SPME via sol-gel technology has gained great interest among researchers. Recently, Es'haghi *et al*. [13] has come out with a new SPME approach namely hollow fiber solid phase microextraction (HF-SPME). The significant of this technique is, it eliminates the main drawbacks of the ordinary SPME fibers such as sample carryover, breakage and stripping of the fibers and favourable for analysis in aqueous samples. The possible memory effect can be overcome since the HF-SPME device is discarded after single use. This technique is simple and practical where the sol solution is held (injected) into the hollow fiber (HF) segment and the sol solution can be left at ambient temperature to allow in situ gelation. This approach has been successfully applied for determination of phenobarbital [13], organophosphorus pesticides [14, 15], selected aflatoxins [16], non-steroidal anti-inflammatory drugs [17] and diethylstilbestrol [18].

In this work, a new sol-gel hybrid based on methyltrimethoxysilane-(3-mercaptopropyl)trimethoxysilane (MTMOS-MPTMOS) as a new extractant of HF-SPME was synthesized in-house to assess its potential and effectiveness in determining hexanal and heptanal in aqueous sample. Fine-tuning of sol solution components is the main key to obtain the desired sol-gel product. In order to achieve satisfactory performance of the new sol-gel silica-based hybrid material in extracting hexanal and heptanal, several parameters affecting the sol-gel components were optimized. Selection of optimal sol-gel conditions were based on extraction efficiencies which was measured by peak area response obtained using gas chromatography-flame ionization detector (GC-FID).

2.0 EXPERIMENTAL

2.1 Reagents and Materials

Methyltrimethoxysilane (MTMOS), (3-mercaptopropyl)trimethoxysilane (MPTMOS), trifluoroacetic acid (95%) and polymethylhydroxysiloxane (PMHS) were obtained from Aldrich (St. Louis, MO, USA) while HPLC grade methanol was purchased from Fisher Scientific (Leicestershire, UK). Acetonitrile, tetrahydrofuran and ethanol used for solvent optimization were supplied from Merck (Darmstadt, Germany). Deionized water was produced from Milipore water purification system (Molsheim, France). Accurel Q3/2 polypropylene tubular membranes from Membrana (Wuppertal, Germany) with a wall thickness of 200 μm, pore size of 0.2 μm, and an id of 600 μm were used for HF-SPME device. Test analytes were hexanal and heptanal. Both aldehydes were purchased from Merck Schuchardt (Hohenbrunn, Germany). Stock solutions (1000 μg/L) of hexanal and heptanal were prepared by dissolving the calculated amounts in methanol and fresh working solutions were prepared daily by further dilution of the stock solution in methanol.

2.2 Preparation of sol-gel hybrid

Sol solution was prepared by mixing appropriate volumes (μL) of precursors (MTMOS and MPTMOS), water, TFA (95%) as acid-catalyst, MeOH and PMHS in a 2 mL centrifuge tube to produce the desired mol ratios. The mixtures were vortexed for 2 min followed by centrifugation at 12 000 rpm for 5 min. Then homogenous solutions were injected into the HF to allow for in situ gelation.

2.3 HF-SPME procedure

The polypropylene (PP) HF was cut into ~ 2 cm segments and cleaned with acetone. It was then air dried before being injected with 6 μL of the sol solution. Both sides of the HF devices were heat sealed before used. To ensure that the sol held in the HF was solidified, it was left at ambient temperature for at least overnight and the HF-SPME devices were then kept in an oven at 100^{o}C for further drying. Extraction was performed by direct immersion of the HF-SPME devices in 10 mL aqueous sample that was spiked with 25 ppb of each hexanal and heptanal. Extraction was carried out for 40 min at a stirring rate of 600 rpm. After extraction, the HF-SPME device was transferred into another small vial and desorption process was performed by ultra sonication for 20 min with 500 μL methanol as desorption solvent.

2.4 Sol-gel process optimization

The mol ratio of MTMOS was tested from 0.5 mmol to 4 mmol with keeping the other amount constant at 1 mmol MPTMOS, 6 mmol water, 6 mmol MeOH, and 2.5 mmol TFA (95%). For optimization of MPTMOS, the mol ratio was varied from 1 mmol to 7 mmol with kept constant the mol ratio of MTMOS at 1 mmol and the other components are as above-mentioned. Effect of water content was evaluated from 3 mmol to 9 mmol in sol solution containing of 1 mmol MTMOS, 4 mmol MPTMOS, 6 mmol MeOH and 2.5 mmol TFA (95%).

2.5 Gas chromatography

A 7820A Agilent GC from Agilent Technologies (Waldbronn, Germany) equipped with a HP-5 (5% phenyl methyl siloxane) column (30 m × 0.32 mm ID and 0.23 μm film thickness), a split/splitless injector and flame ionization detector (FID) was used for analysis of extracted hexanal and heptanal. Helium was used as the carrier gas at a flow rate of 1 mL/min. The flow rate of air as oxidant was set at 400 mL/min while the flow rate of hydrogen was set at 30 mL/min. All gaseous sources were supplied from Linde Malaysia (Kuala Lumpur, Malaysia). The oven temperature program was as follows: initial temperature of 50°C, ramp to 80°C at a rate of 10°C/min, then to 180°C at a rate of 40°C/min to obtain a run time of 5.5 min. The inlet temperature used was 250°C and the FID temperature was set at 280°C.

3.0 RESULTS AND DISCUSSION

3.1 Precursors

Precursors are the backbones of sol-gel polymer. Synthesis of extracting phase via sol-gel process using only precursors is much easier, faster, cheaper and more controllable as compared to addition of coating polymer [19]. In this study, the precursors used are methyltrimethoxysilane (MTMOS) and 3-mercaptopropyltrimethoxysilane (MPTMOS). The mol ratios of both precursors were evaluated to achieve high selectivity and efficiency extraction. Initially the main precursor MTMOS was evaluated at different mol ratios (0.5- 4 mmol) while the MPTMOS amount was kept constant at 1 mmol. Trifluoroacetic acid (TFA) as acid catalyst and methanol as solvent were fixed at a 2.5 and 6.0 mmol, respectively. PMHS as a deactivation agent (25 µL) was added to each sol solutions. The best response based on the peak area was achieved by employing 1 mmol MTMOS (**Figure 3.1**). The peak areas response for both analytes reduced as the mol ratio of MTMOS: MPTMOS was increased from 2:1, 3:1 and 4:1. The probable reason is that when higher mol ratio of MTMOS: MPTMOS was used, it gave a more 'non-polar' character to the sol-gel hybrid which cause the decrease in extraction efficiency for both hexanal and heptanal. Thus, a mol ratio of 1:1 MTMOS: MPTMOS was selected as the optimum ratio and used for subsequent studies.

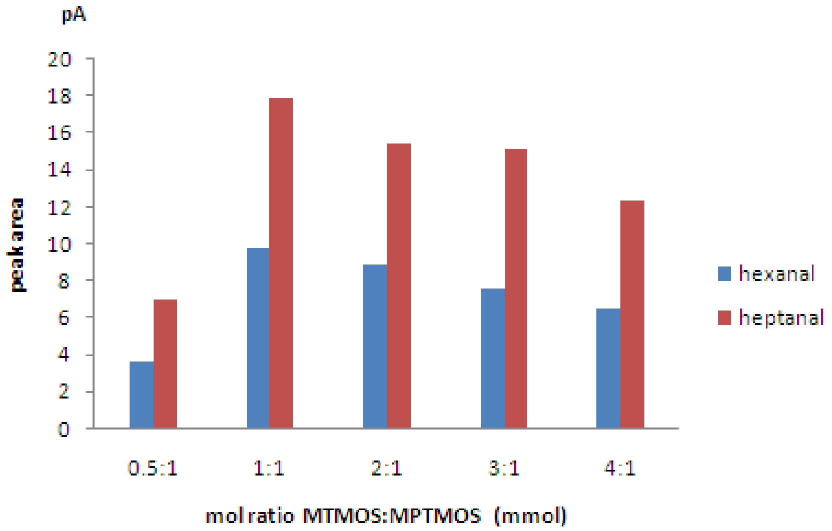

Figure 3.1: Effect of different mol ratios of MTMOS: MPTMOS HF-SPME on the extraction efficiency of hexanal and heptanal. Sol-gel synthesis conditions: TFA (2.5 mmol), MeOH (6 mmol), water (6 mmol) and PMHS (25 µL). HF-SPME extraction conditions: Each test analytes were spiked at 25 ppb in 10 mL aqueous sample and extraction were carried out as follows: 40 min extraction time, 600 rpm stirring rate and 20 min desorption time using 500 µL methanol.

Increasing the mol ratios of MTMOS: MPTMOS from 1:1 to 1:4 gradually increased the peak area response of hexanal and heptanal **(Figure 3.2)**. The addition of MPTMOS that possess thiol (-SH) moiety can alter the polarity of the sol-gel hybrid formed thus enhance the possibility to extract the two mid-polar hexanal and heptanal more effectively. The hybrid sol-gel has organically modified silica to give some interaction towards the test analytes through Van der Waals and dipole-dipole interaction. However, further increase of MPTMOS from 5 mmol to 7 mmol resulted in a decreased of extraction performance for the two aldehydes. At higher mol ratios of MTMOS: MPTMOS, the increase number of thiol moieties led to covalent formation of disulphide (-S-S). This behavior decreased the number density of the bridging oxygens -Si-O-Si- as the network backbone and may alter the features of the sol-gel material. The addition of high amount of MPTMOS led to longer gelation time (more than 12 hr). Therefore, the mol ratio 1:4 MTMOS: MPTMOS was selected as the optimum sol-gel composition.

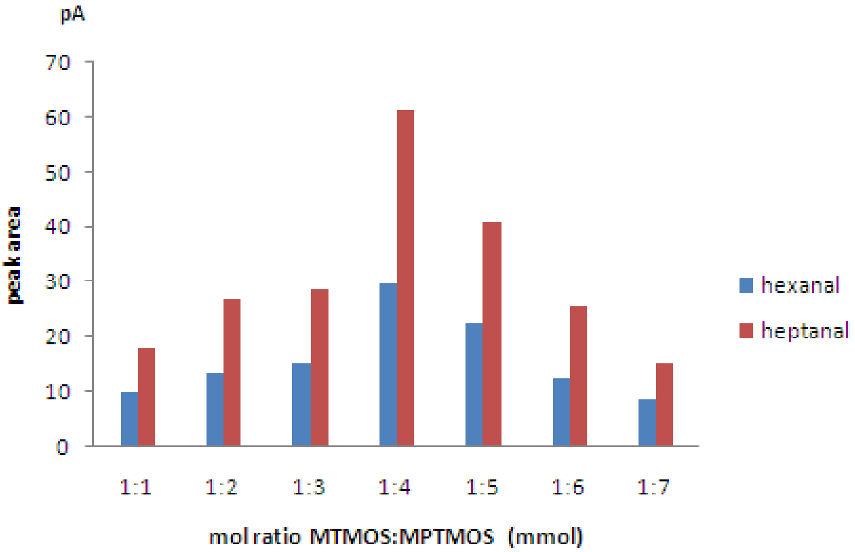

Figure 3.2: Effect of different mol ratios of MTMOS: MPTMOS HF-SPME on the extraction efficiency of hexanal and heptanal. Other conditions are as in Figure 3.1.

3.2 Effect of water content

Water content contributes to significant influence on the structure and properties of sol-gel material since water takes part in hydrolysis and condensation reaction. In the present work, the sol-gel hybrid 1:4 mol ratio MTMOS:MPTMOS materials were prepared using different mol ratios of water varying from 3 mmol to 9 mmol keeping the other amount constant (6 mmol MeOH, 2.5 mmol TFA). **Figure 3.3** shows that employing 3 mmol of water resulted in the lowest peak areas for hexanal and heptanal. Under acidic condition, the rate of hydrolysis reaction is slow as compared to basic condition. Addition of small amount of water content in sol system may alter the resulting sol-gel product. The precursor will compete with each other to be hydrolyzed and the precursor with higher reactivity will take part in hydrolysis. The low extraction performance seen at low mol ratio might be due to MTMOS dominating the hydrolysis and polycondensation reaction.

When the water content was increased from 3 mmol to 6 mmol, higher peak area responses were observed for hexanal and heptanal. Based on stoichiometric calculation, both MTMOS and MPTMOS have three methoxy groups each that can be hydrolyzed. This is parallel with the results obtained as the addition of 6 mmol of water in the sol system gave the highest peak area response. An increase in water content also led to shorter gelling time. This might be due to the presence of more water in sol solution, faster hydrolysis and polycondensation reaction can occur. Further increase of water content from 7 to 9 mmol resulted in a reduction of extraction performance of the sol-gel hybrid material. We should note that complete hydrolysis rarely occurred since condensation reaction may take place between silanol group and a methoxy group or between two silanols to produce methanol and water, respectively [20]. Thus, the addition of higher volume of water content may not necessary owing to elimination of water during condensation may take part in reaction to produce the 3-D network. Therefore, 6 mmol of water was selected as the optimum amount for further investigation on sol-gel synthesis optimization.

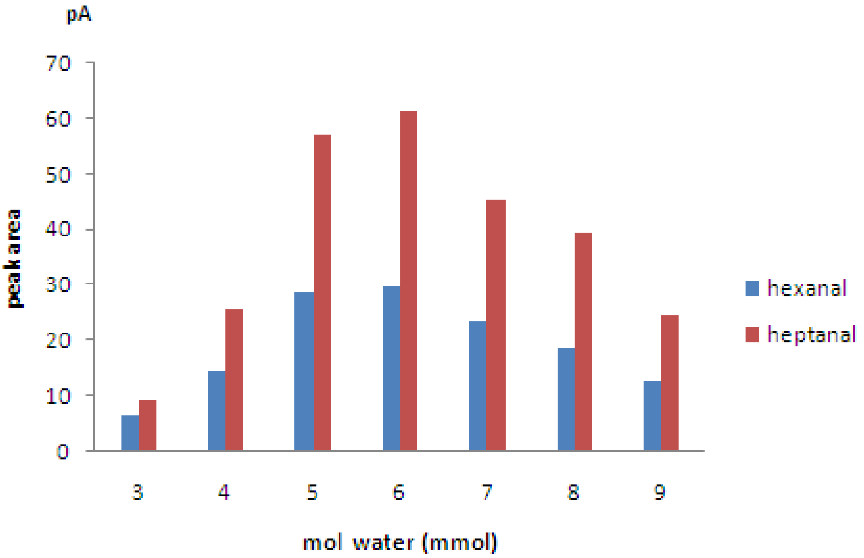

Figure 3.3: Effect of different mol ratios of water in 1:4 MTMOS: MPTMOS HF-SPME on the extraction efficiency of hexanal and heptanal. Other conditions are as in Figure 3.1.

4.0 CONCLUSIONS

In situ gelation of a new sol-gel silica-based hybrid sorbent HF-SPME was successfully used in the optimization of several sol-gel process parameters for extraction of two aldehyedes namely hexanal and heptanal prior to GC-FID analysis. Separations of hexanal and heptanal were achieved within 6 minutes with good resolution. It was found that optimum extraction efficiencies was accomplished using sol solution containing 1 mmol MTMOS, 4 mmol MPTMOS, 6 mmol H_2O, 6 mmol MeOH as solvent and 2.5 mmol TFA 95% as catalyst with 25 µL PMHS (25) as deactivation agent. The in situ gelation sol-gel silica-based hybrid HF-SPME device makes the optimization of sol-gel components become more convenient and faster. This in situ gelation HF-SPME is a promising technique for determination of hexanal and heptanal in aqueous solution.

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REFERENCES

[1] Hakim, M., Broza Y.Y., Barash, O., Peled, N., Phillips, M., Amann, A. and Haick, H. (2012). Volatile Organic Compounds of Lung Cancer and Possible Biochemical Pathways. *Chem Rev*. 112, 5949-5966.

[2] Gordon, S.M., Szidon, J.P., Krotoszynski, B.K., Gibbons, R.D., O'Neill, H.J. (1985). Volatile organic compounds in exhaled air from patients with lung cancer. *Clin.Chem*. 31,1278-1282.

[3] O'Neill, H.J., Gordon, S.M., O'Neill, M.H., Gibbons, R.D., Szidon, J.P. (1988). Computerized classification technique for screening for the presence of breath biomarkers in lung cancer. *Clin. Chem*. 34, 1613-1618.

[4] Poli D, Goldoni M, Corradi M. Acampa, O., Carbognani, P., Internullo, E., Casalini, A., Mutti, A.(2010). Determination of aldehydes in exhaled breath of patients with lung cancer by means of on-fiber derivatization SPME-GC/MS. *J Chromatogr B*. 878, 2643-2651.

[5] Rudnicka, J., Kow alkow ski, T., Ligor, T., Buszw eski, B. (2011). Determination of volatile organic compounds as biomarkers of lung cancer by SPME–GC–TOF/MS and chemometrics. *J. Chromatogr B*. 879, 3360-3366.

[6] Deng, C., Li, N., Zhang, X. (2004). Development of headspace solid-phase microextraction with on-fiber derivatization for determination of hexanal and heptanal in human blood. *J. Chromatogr B*. 813, 47-52.

[7] Guadagni, R., Miraglia, N., Simonelli, A., Silvestre, A., Lamberti, M., Feola, D., Acampora, A., and Sannolo, N. (2011). Solid-phase microextraction–gas chromatography–mass spectrometry method validation for the determination of endogenous substances: Urinary hexanal and heptanal as lung tumor biomarkers. *Anal. Chim. Acta*. 701, 29–36.

[8] Sarafraz-Yazdi, A. Dizavandi, Z.R., and Amiri, A. (2012). Determination of phenolic compounds in water and urine samples using solid-phase microextraction based on sol–gel technique prior to GC-FID. *Anal. Methods*. 4, 4316–4325.

[9] Kumar, A., Gaurav, Malik, A.K., Tewary, D.K., and Singh, B. (2008). A review on development of solid phase microextraction fibers by sol-gel methods and their applications. *Anal. Chim. Acta*. 610, 1-14.

[10] Wan Ibrahim W.A, Veloo, K.V., Sanagi, M.M. (2012). Novel Sol-Gel Hybrid Methyltrimethoxysilane-Tetramethoxy-silane as Solid Phase Extraction Sorbent of Organophosphorus Pesticides, *J. Chromatogr. A*. 1229, 55-62.

[11] Wan Ibrahim W.A, Abdul Keyon, A.S., Prastomo, N., Matsuda, A. (2011). Synthesis and characterization of polydimethylsiloxane-cyanopropyl-triethoxysilane-derived hybrid coating for stir bar sorptive extraction, *J. Sol-Gel Sci. Technol*. 59, 128-134.

[12] Wan Ibrahim, W.A., Wan Ismail, W.N., and Sanagi, M.M. (2013). Selective and Simultaneous Solid Phase Extraction of Polar and Non-Polar Organophosphorus Pesticides Using Sol-Gel Hybrid Silica-Based Sorbent. *J. Teknologi*. 62, 383–87.

[13] Es'haghi, Z., Rezaeifar, Z., Rounaghi, G-H., Nezhadi, Z.A., and Golsefidi, M.A. (2011). Synthesis and application of a novel solid-phase microextraction adsorbent: Hollow fiber supported carbon nanotube reinforced sol-gel for determination of phenobarbital. *Anal. Chim. Acta*. 689, 122-128

[14] Ebrahimi, M., Es'haghi, Z., Samadi, F., and Hosseini, M-S. (2012). Rational design of heteropolyacid-based nanosorbent for hollow fiber solid phase microextraction of organophosphorus residues in hair samples. *J. Chromatogr A*. 1225, 37-44.

[15] Ebrahimi, M., Es'haghi, Z., Samadi, F., and Hosseini, M-S. (2011). Ionic liquid mediated sol–gel sorbents for hollow fiber solid-phase microextraction of pesticide residues in water and hair samples. *J. Chromatogr A*. 1218, 8313-8321.

[16] Es'haghi, Z.,orayaei, H., Samadi, F., Masrournia, M., and Bakherad, Z. (2011). Fabrication of a novel nanocomposite based on sol–gel process for hollow fiber-solid phase microextraction of aflatoxins: B1 and B2, in cereals combined with high performance liquid chromatography–diode array detection. *J. Chromatogr B*. 879, 3034-3040.

[17] Song, X-Y, Shi, Y-P and Chen, J (2012). A novel extraction technique based on carbon nanotubes reinforced hollow fiber solid/liquid microextraction for the measurement of piroxicam and diclofenac combined with high performance liquid chromatography. *Talanta*. 100, 153-161.

[18] Yang, Y., Chen, J., and Shi, Y-P. (2012). Determination of diethylstilbestrol in milk using carbon nanotube-reinforced hollow fiber solid-phase microextraction combined with high-performance liquid chromatography. *Talanta*. 97, 222-228.

[19] Bagheri, H., Piri-Moghadam, H., and Ahdi, T. (2012). Role of precursors and coating polymers in sol-gel chemistry toward enhanced selectivity and efficiency in solid phase microextraction. *Anal. Chim. Acta*. 742, 45-53.

[20] Schubert, U. Hüsing, N., and Lorenz, A. (1995). Hybrid Inorganic-Organic Materials by Sol-Gel Processing of Organofunctional Metal Alkoxides. *Chem. Mater*. 7, 2010-2027.

[21] Koodali, R.T., Mokhtari J., Sorensen, C.M., Klanbunde, K..J. (2013). Role of solvents in the gelation process: can light scattering studies shed some light? *J. Sol-Gel Sci. Technol*. 66, 43-49.