Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2016, 8(7):702-707

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Optical and Structural study of ZnSe Nano rods-Tryptophan conjugate

T. K. Das[1], A. K. Bhunia[1,2*], T. Kamilya[3] and S. Saha[1]

[1]Department of Physics & Technophysics, Vidyasagar University, Paschim Medinipur- 721102, India
[2]Department of Physics, Government general Degree College at Gopiballabpur-II, Beliaberh, Paschim Medinipur- 721517, India
[3]Department of Physics, Narajole Raj College, Paschim Medinipur- 721211, India

ABSTRACT

A simple wet chemical method has been successfully used to grow zinc selenide nanorods (ZnSe NRs). The structural characteristics were investigated through TEM and XRD. The crystal unit cell of the nanorods is found to be a mixture of hexagonal phase and cubic phase and the average size of the ZnSe NR ≈10 nm. The photoluminescence spectrum shows shallow deep level visible emission due to various defect states. The interaction as well as the formation of bioconjugate of Tryptophan (Trp) and Zinc Selenide nanorods is investigated using optical spectroscopy and HRTEM images. UV–VIS and fluorescence spectra show that a spontaneous binding process occurred between Trp and Zinc Selenide nanorod. A small red shift of the absorption peak of Trp is observed due to binding of Trp with ZnSe NRs. Zinc Selenide nano rods quench the fluorescence emission of tryptophan. The Stern–Volmer quenching constant, the binding constant and the number of binding sites were also calculated.

Keywords: ZnSe nanorod; Triptophan; photoluminescence spectra; absorption spectra; high resolution transmission electron microscopy.

INTRODUCTION

The semiconductor nanorods exhibit structural, optical, luminescence and photo conducting properties that are very different from their bulk properties. It is very attractive because of their possible application in solar cell, photo detector, laser, LED, high density magnetic information storage and many others in semiconductor industries [1–7]. A2B6 semiconductor nanorods play an important role having application in nano devices [8, 9,31]. ZnSe (bulk band gap 2.6 eV at 300 K) has huge potential in this aspect. Their growth techniques are relatively cheap. Their characteristic absorption of light is in the visible range. There are various methods to prepare ZnSe nanorods [10–19] . Some of the above mentioned methods have some draw backs. Used precursors are unstable causing environmental hazards and required very high temperatures. These methods are not cost effective also. In the present work a chemical reduction method is followed at room temperature. Sodium borohydride is used to initiate the reaction between $ZnCl_2$ and selenium at room temperature. The grown samples are dispersed in ethanol to characterize it structurally and optically. Size dependent tunable optical property which is of great interest in the NPs based drug delivery, bio-imaging, and biomedical research [20, 21,32,33]. However, before going into realizing these applications, it is very much essential to understand the way of interaction of bare ZnSe NPs with protein. The study of protein-NPs conjugation will provide us with the information of the phenomena occurring at the protein-NPs interface at the molecular level as well as the information about conformational changes of protein occurring at the protein-NPs interface. Whenever NPs come in physiological fluid systems they are surrounded by the protein molecules. The NPs then get associated with the protein molecules and a dynamic layer of proteins is formed on the surface of NPs. This conjugated system is known as “NPs-protein corona.”[16]. Tryptophan (TRP), an important protein fluorophore and one of the 20 standard amino acids as well as an essential amino acid in the human diet, has

long been the subject of photophysical studies. We hereby present the interaction between TRP with a nanoparticles (ZnSe). The fluorescence response of Trp is associated with the Π –Π* transition of its indole functional group with the absorption and fluorescence wavelengths of about 278 and 348 nm, respectively [23, 24]. Trp molecule (inset of Fig. 6) can offer at least three major sites for interaction: (i) salt-bridge interaction involving the carboxylic group, $–COO^-$, (ii) charge–solvent structure interaction involving the indole ring, and (iii) amine site interaction involving the $–NH_2$ group [25]. In order to recognize the physical basis of the biological activity of ZnSe NRs under conditions of environmental exposure in a better way, we have focused our aim to analyze the formation of ZnSe NRs-Tryptophan corona and interaction of ZnSe NRs with L-Tryptophan major spectroscopic along with microscopic techniques. In this paper the Absorption and fluorescence quenching technique was applied to study the interaction of Trp with ZnSe nanomaterials. From the plot of log $[(F_0 -F)/F]$ vs. log[Q] and by using the binding constants, the numbers of binding sites and the nature of the interacting forces among the reactants have been discussed. The TEM analysis showed the corona formation mechanism.

EXPERIMENTAL SECTION

Anhydrous $ZnCl_2$ (99.999%) (360 mg), selenium powder (99.999%) (208 mg) and stoichiometric amount of sodium borohydride (98%) (150 mg) were purchased from Alfa-Aesar to prepare sample. Ethylene-di-amine (99%) used as capping agent. Sodium borohydride were taken to initiate the reaction at room temperature. The reaction was carried out at 30^0C. The stirring was continued for three hours at a particular speed. The Tryptophan solution with predetermined concentration of Tryptophan was prepared by using triple distilled water, deionized with a Milli-Q water purification system from Millipore, U.S.A. The pH and the resistivity of freshly prepared water were 6.8 and 18.2MΩ cm, respectively. The so prepared ZnSe NRs were dispersed in Millipore water using ultra sonication for 15 min. The concentration of the ZnSe was varied from 100 µM to 700µM. Trp-ZnSe NRs mixed solutions were prepared by mixing 50 µM Trp with ZnSe NRs, ranging from 100 µM to 700µM with proper ratio. X-ray diffraction was carried using Rigaku X-ray diffractometer system over $20 < 2\theta < 80$ using Cu-kα radiation of wavelength λ=1.54Å. For further structural characterization transmission electron microscopy images were taken in a JEOL JEM-2100F microscope with the accelerating voltage of 200 kV. For TEM study a very small amount of the powder sample was first dispersed in water by ultra-sonication . A drop of that solution was taken on a carbon coated grid for TEM imaging. The room temperature PL spectrum was recorded in PERKIN ELMER LS- 55 with a Xenon lamp with the excitations of 330nm. The optical absorption spectra of the samples were taken by using Shimadzu-Pharmaspec-1700 UV-VIS, after ultra-sonication of the samples in water.

RESULTS AND DISCUSSION

Absorption Spectra

The tryptophan exhibits absorption peak at ~278 nm (Fig. 1(a)) due to the Π-Π* transition of aromatic amino acid residues. The UV visible spectra presented in Figures 2 illustrate the effect of binding of ZnSe NRs with with Trp. In fig. 1(b)- (f) our results showed that the tryptophan absorbance (278 nm) increases in increases with C_{ZnSe} . The increase in intensity of absorbance of Trp in the presence of ZnSe may be due to binding ofTrp with ZnSe NRs and the formation of the ground state complex [26].

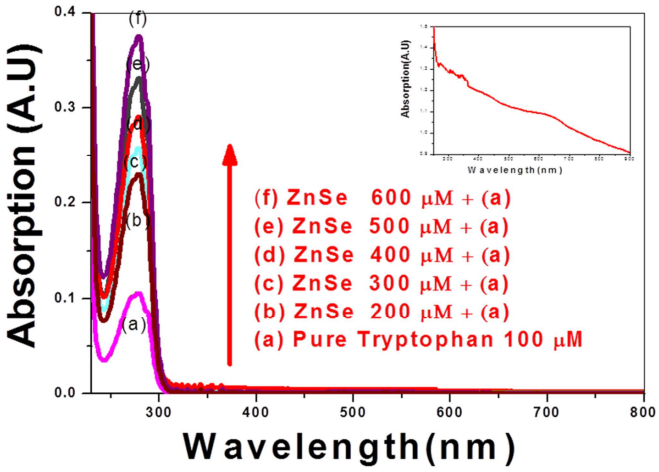

Figure 1: Absorption spectra of (a) pure Tryptophan(100 µM),(b) Tryptophan with 200 µM ZnSe NRs ,(c) Tryptophan with 300 µM ZnSe NRs, (d) Tryptophan with 400 µM ZnSe NRs,(e) Tryptophan with 500 µM ZnSe NRs,(f) Tryptophan with 600 µM ZnSe NRs; Inset shows absorption spectra of pure ZnSe NRs(0.01 mg/mL)

X-ray diffraction (XRD)

A typical XRD pattern of the ZnSe NRs is shown in figure 2. The unit cell of the crystal was found to be mixture of hexagonal phase and cubic phase with the presence of the peaks (111), (220), (311), (400), (331). Furthermore, the intensities of different diffraction peaks are different, which indicates that the growth of various planes (direction) is different.

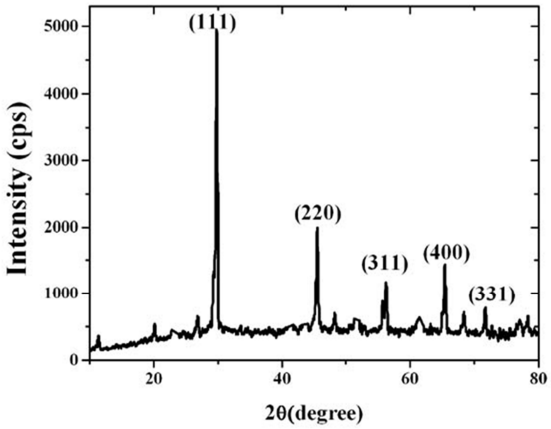

Figure 2: The XRD pattern of the sample ZnSe NRs

Fluorescence Spectroscopy

Figure 3(A) shows the emission spectra of pure ZnSe nanorods. The photoluminescence spectrum shows shallow deep level visible emission around 400 nm due to various defect states. The ZnSe NRs-Trp binding kinetics equilibrium has been analyzed by fluorescence quenching measurements. The addition of ZnSe NRs of different concentrations (C_{ZnSe}) with Trp results a change in the maximum fluorescence emission spectrum intensity (I_{max}), suggesting the occurrence of fluorescence quenching process (Fig. 3(B)) [28-30]. The quenching occurs via the adsorption and interaction of the Tryptophan residues accessible to the metallic surface of the ZnSe NRs. A small red shift (~4 nm) of Trp emission with C_{ZnSe} also signifies the unfolding as well as denaturation of Trp in the presence of ZnSe NRs. The binding constant *K* along with the number of binding sites (*n*) between Trp and ZnSe NRs are $1.94X10^3\ M^{-1}$ and 2.11 respectively. This indicates that a positive cooperative take place [27]. In favor of positive cooperative reaction, *n*›1, reveals that once one Trp molecule is bound to the NPs, its affinity for the nanoparticle gradually increases in a superliner fashion. Schematic of the Trp-ZnSe NPs binding is shown in Figure 6.

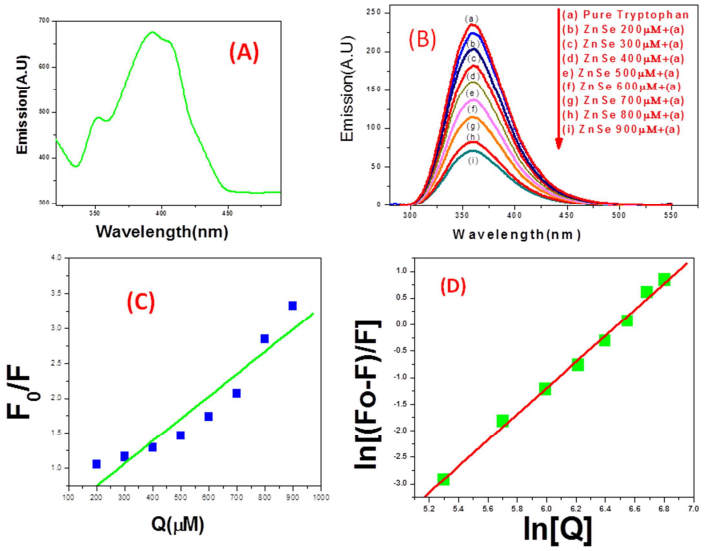

Figure 3: Emission spectra of (A) pure ZnSe NRs ,(B)(a) pure Tryptophan(50 µM) (b)Tryptophan with 200 µM ZnSe NRs , (c) Tryptophan with 300 µM ZnSe NRs, (d) Tryptophan with 400 µM ZnSe NRs,(e) Tryptophan with 500 µM ZnSe NRs, (f) Tryptophan with 600 µM ZnSe NRs ; (C) F0/F vs Q(µM); (D) ln [(F_0-F)/F] vs ln[Q]

TEM

Figure 4 (a) shows the nanostructure of pure ZnSe with diameter 10 nm. Figure 4(b),2(c)2(d) shows the behavior of the ZnSe nanorods inside Trp molecules. HRTEM of 4(d) and 5(a) clearly indicate that core ZnSe nanorod is coated by shell Tryptophan. The SEAD pattern of pure ZnSe nanorods are crystalline [fig.5(b)] in nature but when they are coated by Trp molecules they shows amorphous in nature [fig.5(c)]. This images (fig. 4 and fig.5) clearly represents that the core ZnSe NRs are fully covered with Trp along with shell thickness of ~ 8 nm, matched with dimension of Trp (~8 nm), calculated from protein data bank [22]. Schematic of the Trp-ZnSe NPs conjugation is shown in Figure 6.

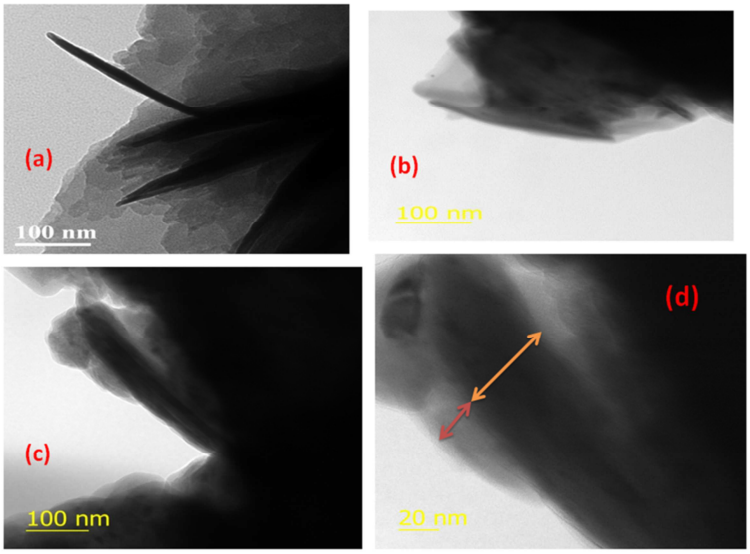

Figure 4: HRTEM of (a) pure ZnSe NRs; (b) ZnSe-Tryptophan conjugate ; (c) ZnSe-Tryptophan conjugate ; (d) core ZnSe coated with shell Tryptophan

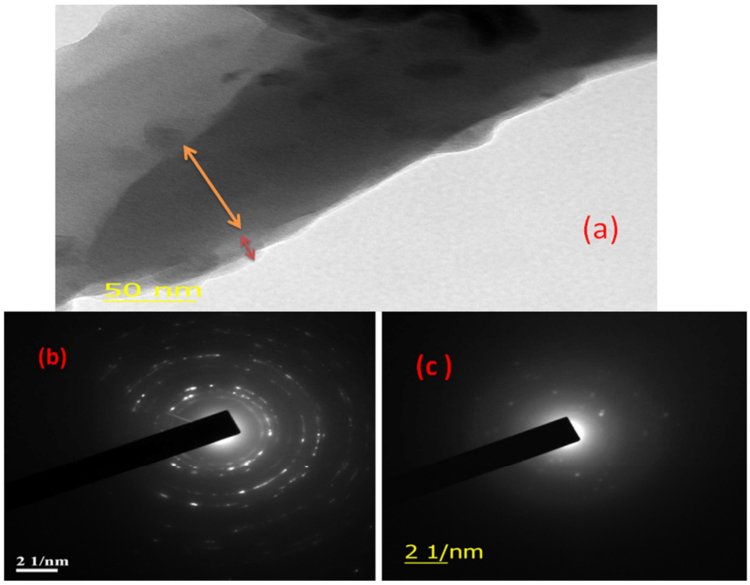

Figure 5: (a) HRTEM of core ZnSe with shell Tryptophan; (b) SAED of pure ZnSe NRs; (c) SAED of ZnSe-Tryptophan conjugate

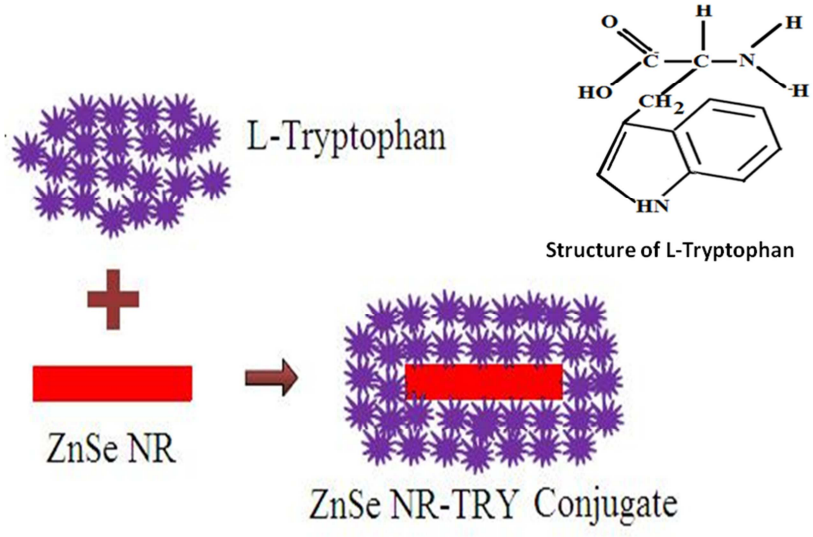

Figure 6. Schematic representation for interaction and conjugate formation of ZnSe NRs and Tryptophan; Inset shows structure of L-Tryptophan

CONCLUSION

In conclusion, we have synthesized Zinc Selenide nanorods of average diameter ~10 nm using a simple wet chemical method. The XRD pattern of the as synthesized samples shows a mixture of hexagonal phase and cubic phase. The emission quenching of the nanoprods-Trp system showed a strong interaction phenomenon. The interaction between ZnSe nanorods with Tryptophan showed positive cooperative reaction phenomenon. The TEM picture along with SAED pattern of the NRs-Trp indicates that the NRs are completely covered by Tryptophan molecules. The present investigation provides important insight into the interaction of the physiologically important protein fluorophore Tryptophan with semiconductor ZnSe nanorods and possesses potential applications in biotechnology.

Acknowledgements
Authors are grateful to UGC and DST for their constant financial assistance through SAP and FIST programme to Department of Physics and Technophysics of Vidyasagar University.

REFERENCES

[1] R. Lv, C. Cao, H. Zhai, D. Wang, S. Liu, H. Zhu, *Solid State Commn*, **2004,**130, 241.
[2] G.W. Orr, L.J. Barbaur, J.L. Atwood, *Science*, **1999**,285, 1049.
[3] J. Goldberger, R. He, Y. Zhang, *Nature* ,**2003**, 422 ,599.
[4] Z.L. Wang, R.P. Gao, J.L. Gole, *Adv. Mater*, **2000**,12, 1938
[5] C.N.R. Rao, A. Gobindaraj, F.L. Deepak, *Appl. Phys. Lett*., **2001**, 78, 1853.
[6] Y. Xiong, Y. Xie, J. Yang, *J. Mater. Chem*, **2002**,12, 3712.
[7] L. Dloczik, R. Engelhardt, K. Ernst, *Appl. Phys. Lett*, **2001**,78, 3687.
[8] P.G. Collins, A. Zettl, H. Bando, *Science*, **1997,** 27,8100.
[9] E.W. Wong, P.E. Sheehan, C.M. Lieber, *Science* , **1997,** 277,1971.
[10] Y.T. Chen, J.B. Ding, Y. Guo, *Mater. Chem. Phys*, **2002,** 77, 734.
[11] D. Xu, Y. Xu, D. Chen, *Adv. Mater*, **2000**,12, 520.
[12] J. Yang, J.H. Zeng, S.H. Yu, *Chem. Mater*, **2000,** 12, 3259.
[13] Y.D. Li, H.W. Liao, Y. Ding, *Chem. Mater*, **1998**,10, 2301.
[14] X.H. Liao, H. Wang, J.J. Zhu, *Mater. Res. Bull*, **2001**,36, 2339.
[15] N.G. Chopra, R.J. Luyken, K. Cherry, *Science*, **1995,**269, 966.
[16] B.A. Korgel, D. Fitzmaurice, *Adv. Matter*, **1998**,10, 661.
[17] O. Ray, A.A. Sirenko, J.J. Berry, *Appl. Phys. Lett*, **2000,** 76, 1167.
[18] H.P.Wagner, H.P. Tranitz, R. Schuster, *Phys. Rev. B* , **2001**,63, 155311.
[19] C.H.C. Wallace, X. Sha, S. Yuxiu, *J. Phys. D: Appl. Phys*, **2009**,42, 125410.
[20] J. W. Rasmussen, E. Martinez, P. Louka, and D. G. Wingett, Expert. *Opin. Drug Deliv* ,**2010**, 7,1063.
[21] S. Saha and A. K. Bhunia, *Journal of Physical Sciences* ,**2014,** 19, 109.

[22] Protein Data Bank.
[23] H. Du, R. A. Fuh, J. Li, A. Corkan and J. S. Lindsey, *Photochem. Photobiol*, **1998**,68, 141.
[24] J. T. Vivian and P. R. Callis, *Biophys. J*, **2001** ,80, 2093.
[25] RCSB PBB:RCSB Protein Data Bank
[26] A.K. Bhunia, P. K. Samanta, S. Saha, and T. Kamilya. *Appl. Phys. Lett*, **2013**, 103, 143701.
[27] A.K. Bhunia, P. K. Samanta, S. Saha, and T. Kamilya, *Appl. Phys. Lett*, **2014**, 104, 123703.
[28] X. C. Shen, X. Y. Liou, L. P. Ye, H. Liang, and Z. Y. Wang, J. *Colloid Interface Sci*, **2007,** 311, 400.
[29] D. J. Gao, Y. Tian, S. Y. Bi, Y. H. Chen, A. M. Yu, and H. Q. Zhang, *Spectrochim. Acta A*,**2005** , 62, 1203.
[30] N. Chadborn, J. Bryant, A. J. Bain, and P. O'Shea, *Biophys. J*, **1999**, 76, 2198.
[31] M.W DeGroot, Khadka Chhatra, Ro Harald ¨ sner, and Corrigan J F *Journal of Cluster Science*, **2006 ,**17, 97
[32] C V Kumar, A Buranaprapuk, J Thota, *Journal of Chemical Sciences,* **2002 ,**114 , 579.
[33] R Kurapati, U V Reddy, A Raichur M, and N Suryaprakash, *J. Chem. Sci*, **2016 ,**128 ,325.