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Development of a rhodamine–benzimidazol hybrid derivative as a novel FRET based chemosensor selective for trace level water†

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A newly designed rhodamine–benzimidazol hybrid molecule has been developed as a FRET-based chemosensor for the selective detection of trace level water in both polar protic and aprotic organic solvents.

During the past few decades, considerable attention has been paid to the detection and quantitative analysis of trace water in organic solvents because of their indispensable applications in food, textile, ceramics, electronic, pharmaceutical, petroleum, and environmental monitoring industries.[1] Previously, water content in samples was measured up to the ≈1 ppm level using the Karl Fischer coulometric titration method.[2] Although this method has been improved through modification, it still has several disadvantages including a long measuring time, the use of toxic and expensive chemical reagents, difficulty in endpoint determination and interference by other common species. Nowadays, optical water sensors based on fluorescence intensity and naked eye detection are particularly attractive on account of their high sensitivity and selectivity, quick, inexpensive, and easy fabrication and non-destructive properties as well as their capability of remote and *in situ* monitoring of optical measurements.[3–10] In previous reports, several approaches have been used to facilitate sensitive water detection in many polar aprotic and polar protic solvents, including intramolecular charge transfer,[4] proton transfer,[5] photo-induced electron transfer,[6] water-induced decomplexation of dyes with anions,[7] water-induced interpolymer p-stacking aggregation,[8] solvatochromism,[9] *etc.*[10] In most cases, however, the strong polarity and hydrogen bonding of the sensing medium are key in the determination of water by these sensors as well as in lowering their LOD. As a result, there is still an increasing demand for highly sensitive optical sensors for the detection of water in polar protic solvents including a new detection principle for the development of new optical water sensors.

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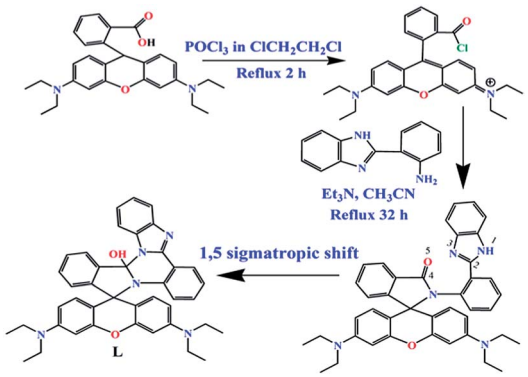
To the best of our knowledge, an optical water sensor based on a FRET mechanism (not hydrogen bonding) is a new approach that has not yet been reported. The utilization of this novel strategy for water detection has several advantages. The selectivity of this rhodamine-based probe (**L**) towards water is extremely high (Fig. S1†) and independent of solvent type (non-polar, polar, aprotic and protic). This probe also responds through FRET, which provides a good detection limit.[11] Fluorescence techniques have become powerful tools for sensing different analytes in trace amounts because of their simplicity, high sensitivity and real-time monitoring with a short response time.[12] Among them, FRET, a distance-dependent radiationless energy transfer from an excited donor fluorophore to a suitable acceptor fluorophore, is now an important physical phenomenon with considerable interest for studying molecular level interactions in biological systems. FRET also has potential applications in optoelectronic and thin film device development due to its sensitivity to distance and intramolecular phenomena, which is resistant to outside perturbation. The distance over which energy can be transferred is dependent on the spectral characteristics of the fluorophores, but is generally in the range of 10–100 Å. However, the efficiency of energy transfer also depends on the relative orientation of transition dipoles of both the donor and acceptor, and the extent of spectral overlap between the donor emission and acceptor absorption.[11]

To synthesize the probe **L**, rhodamine B acid was first converted to its activated acid chloride form by the conventional method.[13] The mixture of the produced rhodamine B acid chloride and added *2-amino phenyl benzimidazole* (2-APBZ) was then refluxed for 32 h (Scheme 1 and ESI†). The crystallized product (**L**) obtained from dry acetonitrile was characterised using physico-chemical and spectroscopic tools (Fig. S2–S4†). The 1,5 sigmatropic shift[14] is responsible for obtaining the final rearranged cyclic organic moeity (**L**), which is well supported by IR, ^{1}HNMR and ^{13}CNMR data.

The addition of water to a solution of **L** in dry organic solvents facilitates the formation of the fluorescent zwitterionic structure $\mathbf{L_1}$ through the protonation of the imidazole nitrogen

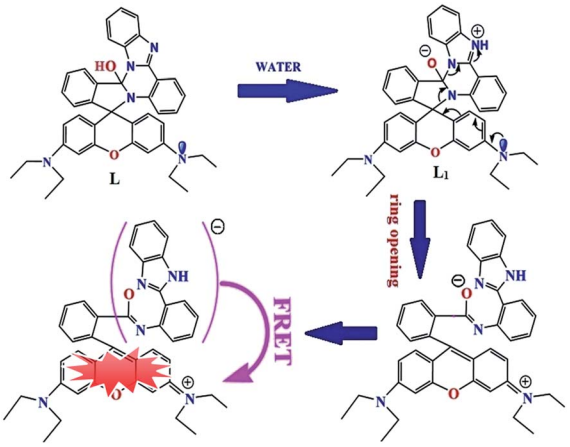


Scheme 1 Synthesis of fluorescence FRET sensor L.

of the *2-amino phenyl benzimidazole* (2-APBZ) unit and deprotonation of the alcoholic hydrogen (Fig. S5 and S6†) Thus, a spirolactone ring opening[15] occurred *via* extended conjugation from the NEt_2 lone pair to the 2-APBZ unit, causing FRET to operate in the molecule (Scheme 2, ESI†) and creating red fluorescence. A peak at *ca.* 3449 cm^{-1} attributable to ν_{OH} was observed in the IR spectrum of **L** in dry acetonitrile, but was absent in the spectrum of **L** in 20% (v/v) water in acetonitrile. Alternatively, the peak at *ca.* 3158 cm^{-1} attributable to ν_{NH} was obtained in later case but not in the former case (Fig. S2†). ^{1}HNMR titration was carried using D_2O (Fig. S5 and S6†). Here, the signal at *ca.* $\delta = 12.64$ ppm was observed in the spectrum of **L** in dry DMSO-d_6 but was absent when the spectrum was acquired by adding a few drops of D_2O into DMSO-d_6. Otherwise, some corresponding peaks shifted slightly towards downfield and some remained unaffected. The peak at approximately 73 ppm due to the sp^3 hybridized carbon was observed in the ^{13}CNMR spectrum of **L** in dry DMSO-d_6, which also supported the existence of –OH attached to sp^3 carbon instead of carbonyl carbon (Fig. S4b†).

To calculate the detection limit (DL) and quantitation limit (QL),[6] the calibration curves[11] (Fig. 2) in the lower region were obtained. From the curve slope (S) and the standard deviation (σ_{zero}) of seven replicate (of each dry solvent) measurements at the zero level, the DL and QL were estimated using as $3\sigma/S$ and $10\sigma/S$, respectively. These values are quite significant compared to previous reports (*viz.* Table 1).

Fluorescence and absorption spectra of **L** were measured in dry organic solvents (acetonitrile, methanol, DMSO and THF) containing various concentrations of water. A new peak generated at approximately 575 nm increased in intensity (~10 times) with increasing water content in acetonitrile, methanol or DMSO. An isoemissive point at around 547 nm (*viz.* Fig. 1a, c and e) along with the steady decrease in fluorescence intensity due to the benzimidazole unit were also observed. In the low water content region below 11 (v/v)%, as shown in Fig. S7,† the fluorescence intensities increased almost linearly with increasing water content in the organic solvent. In contrast, the corresponding absorption spectra exhibited significant changes in absorbance; the spectra showed a ~20 nm blue shift (Fig. 1b, d and f), which is significant evidence supporting the occurrence of FRET.[16] In the absence of water, the rhodamine moiety adopted a closed, non-fluorescent spirolactam form corresponding to a weak spectral overlap between the *2-amino phenyl benzimidazole* (2-APBZ) emission and the rhodamine absorption. Eventually, FRET was suppressed, and only the blue emission of the donor was observed upon excitation of the 2-APBZ chromophore at 350 nm. The water induced ring opening



Scheme 2 Probable mechanistic pathways of sensor **L** for detection of water in organic solvents.

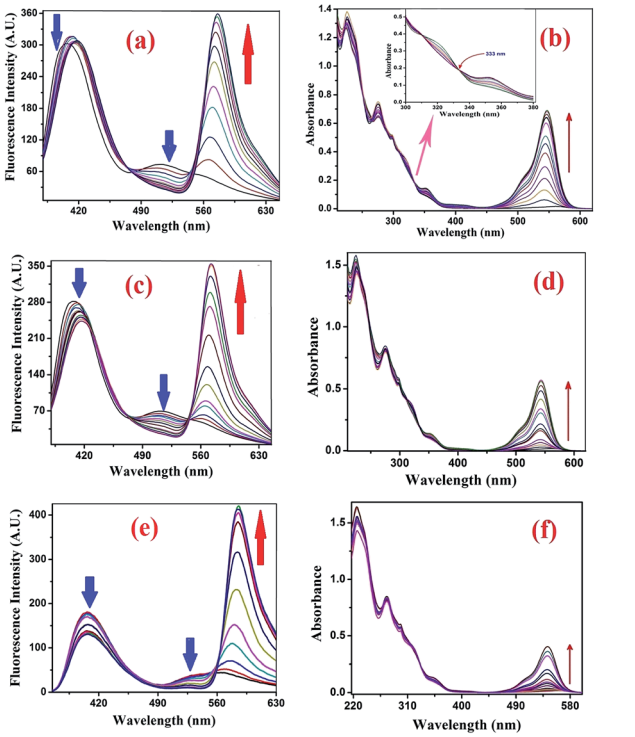


Fig. 1 (a) Fluorescence ($\lambda_{ex} = 350$ nm) and (b) absorption spectra of **L** ($c = 2.0 \times 10^{-5}$ M) in acetonitrile containing water (1.43–20% v/v). (c) Fluorescence ($\lambda_{ex} = 350$ nm) and (d) absorption spectra of **L** ($c = 2.0 \times 10^{-5}$ M) in methanol containing water (1.43–25.7% v/v). (e) Fluorescence ($\lambda_{ex} = 350$ nm) and (f) absorption spectra of **L** ($c = 2.0 \times 10^{-5}$ M) in DMSO containing water (1.43–28.6% v/v).

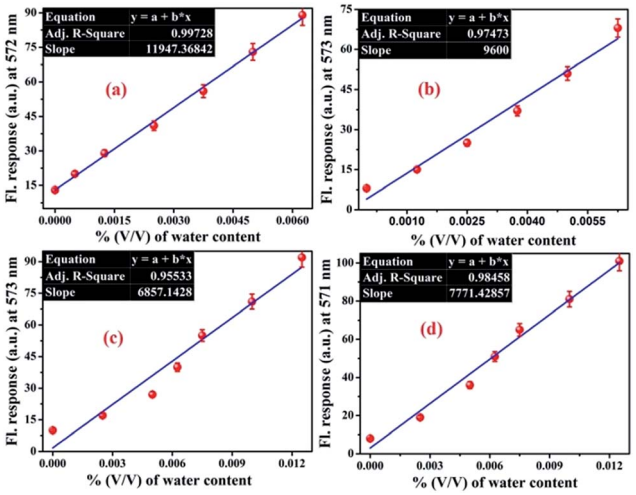


Fig. 2 Calibration curves in the lower concentration range, with error bars for calculating the DL and QL of L as a function of water content in (a) acetonitrile, (b) methanol, (c) DMSO and (d) THF.

Table 1 Comparison of the detection limit of this FRET-based probe (L) with previously reported water sensors

Detection limit (DL)	Quantitation limit (QL)	Ref.
0.038 v/v% (MeCN)	—	4a
0.02 v/v% (THF)	—	4b
0.006 v/v% (MeCN)	—	4c
0.002 v/v% (MeCN)	—	4d
0.0035 v/v% (THF)	—	5a
0.1 wt% (MeCN)	0.4 wt% (MeCN)	6a
0.1 v/v% (EtOH)	0.3 v/v% (EtOH)	8
0.02 v/v% (MeCN)	0.07 v/v% (MeCN)	
0.008 v/v% (DMF)	0.03 v/v% (DMF)	
0.0026 v/v% (MeOH)	0.0085 v/v% (MeOH)	This work
0.0032 v/v% (MeCN)	0.0108 v/v% (MeCN)	
0.0044 v/v% (DMSO)	0.0147 v/v% (DMSO)	
0.0033 v/v% (THF)	0.0105 v/v% (THF)	

of the fluorescent rhodamine moiety, generated the intense absorption in the 2-APBZ emission region. As a result, the spectral overlap was improved, and excitation of the 2-APBZ chromophore resulted in the intense red emission of rhodamine owing to FRET.

In the case of **L**, the fluorescence quantum yield was calculated in HPLC grade organic solvents in presence of water. From these measurements, it is clear that the fluorescence quantum yield increases from 0.77 to 10.21 (14 times, for acetonitrile, at $\lambda = 575$ nm), from 0.77 to 9.32 (12 times, for methanol, at $\lambda = 575$ nm) and from 0.77 to 8.28 (10 times, for DMSO, at $\lambda = 575$ nm) upon the addition of 30% (v/v) water. For this reason, the probe (**L**) exhibits fluorescence and visual colour changes with increasing water content (Fig. S8†).

In support of this FRET process, a fluorescence life time experiment was performed. In this study, the probe (**L**) was placed in dry DMSO (10 μM) and excited at the donor excitation position at 350 nm (from UV-Vis peak of 2-APBZ at 345 nm)[17]. Fig. 3 showed a decrease trend in lifetime at the donor emission position (420 nm) with increasing water content in dry DMSO from 4.05 ns to 3.51 ns, which is in good agreement with the FRET process. A significant spectral overlap was also observed between 2-APBZ and the rhodamine-B moeity (Fig. 4). Again, the Förster distance between donor and acceptor was also calculated (*ca.* 52 Å; ESI†), and supports the occurrence of FRET.

In summary, a new rhodamine–benzimidazol hybrid molecule has been established as a dual channel chemosensor for the detection of trace level water in both protic and aprotic organic solvents through a FRET pathway. This probe is highly selective and sensitive towards water because water has the maximum dielectric constant among all polar protic solvents. For this reason, only water is able to create a dipole in the molecule through zwitterion formation (*viz.* Scheme 2). This dipolar charge is responsible for ring opening (through extended conjugation), favouring the FRET process.

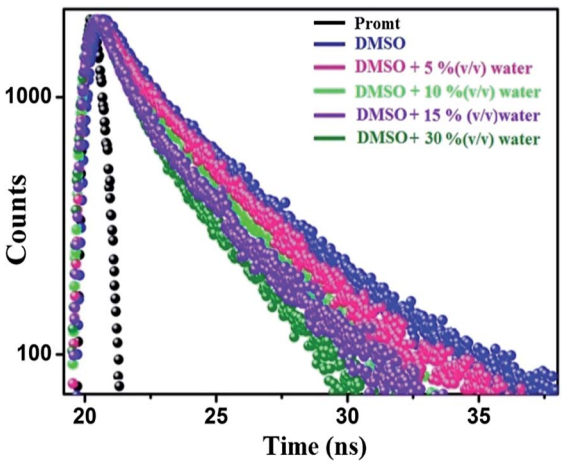


Fig. 3 Fluorescence lifetime decay profiles of L at 420 nm (donor emission position) with increasing water content.

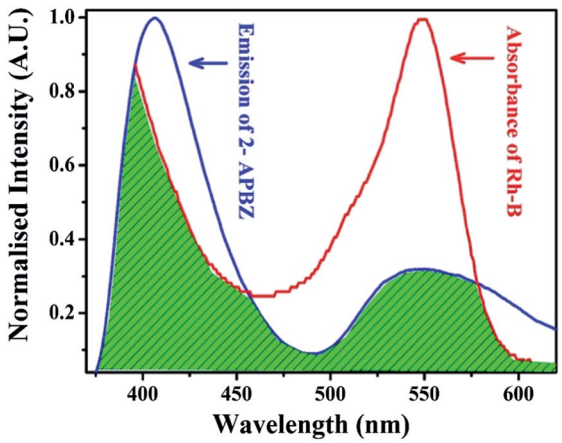


Fig. 4 Overlap spectra of donor emission and acceptor absorbance of L (10 μM) in dry acetonitrile.

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Notes and references

1 (*a*) J. W. Rhim, *Food Sci. Biotechnol.*, 2004, **13**, 528; (*b*) M. E. Grismer, *Soil Sci.*, 1992, **154**, 482; (*c*) F. Garcia-Golding, M. Giallorenzo, N. Moreno and V. Chang, *Sens. Actuators, A*, 1995, **337**, 46; (*d*) L. J. Wadso, *J. Therm. Anal.*, 1997, **49**, 1053; (*e*) E. Traversa, *Sens. Actuators, B*, 1995, **23**, 135; (*f*) V. Sedivec and J. Flek, *Handbook of Analysis of Organic Solvents*, John Wiley, Inc., New York, 1976; (*g*) Q. Wang, X. Li, L. Wang, Y. Cheng and G. Xie, *Ind. Eng. Chem. Res.*, 2005, **44**, 4518; (*h*) S. Al-Zuhair, K. V. Jayaraman, S. Krishnan and W. H. Chan, *Biochem. Eng. J.*, 2006, **30**, 212; (*i*) T. N. P. Nguyen and K. J. Kim, *Ind. Eng. Chem. Res.*, 2010, **49**, 4842.

2 K. Fischer, *Angew. Chem.*, 1935, **48**, 394.

3 (*a*) M. Q. Bai and W. R. Seitz, *Talanta*, 1994, **41**, 993; (*b*) X. Yang, C. G. Niu, Z. J. Shang, G. L. Shen and R. Q. Yu, *Sens. Actuators, B*, 2001, **75**, 43; (*c*) M. A. Kessler, J. G. Gailer and O. S. Wolfbeis, *Sens. Actuators, B*, 1991, **3**, 267; (*d*) B. Bag and A. Pal, *Org. Biomol. Chem.*, 2011, **9**, 915.

4 (*a*) C. G. Niu, L. M. Li, P. Z. Qin, G. M. Zeng and Y. Zhang, *Anal. Sci.*, 2010, **26**, 671; (*b*) Z. W. Li, Q. W. Yang, R. X. Chang, G. C. Ma, M. X. Chen and W. Q. Zhang, *Dyes Pigm.*, 2011, **88**, 307; (*c*) C. G. Niu, P. G. Qin, G. M. Zeng, X. Q. Gui and A. L. Guan, *Anal. Bioanal. Chem.*, 2007, **387**, 1067; (*d*) C. G. Niu, A. L. Guan, G. M. Zeng, Y. G. Liu and Z. W. Li, *Anal. Chim. Acta*, 2006, **577**, 264; (*e*) D. Citterio, K. Minamihashi, Y. Kuniyoshi, H. Hisamoto, S. Sasaki and K. Suzuki, *Anal. Chem.*, 2001, **73**, 5339.

5 (*a*) J. S. Kim, M. G. Choi, Y. Huh, M. H. Kim, S. H. Kim, S. Y. Wang and S. K. Chang, *Bull. Korean Chem. Soc.*, 2006, **27**, 2058; (*b*) W. H. Liu, Y. Wang, W. J. Jin, G. L. Shen and R. Q. Yu, *Anal. Chim. Acta*, 1999, **383**, 299; (*c*) H. Mishra, V. Misra, M. S. Mehata, T. C. Pant and H. B. Tripathi, *J. Phys. Chem. A*, 2004, **108**, 2346.

6 (*a*) Y. Ooyama, M. Sumomogi, T. Nagano, K. Kushimoto, K. Komaguchi, I. Imae and Y. Harima, *Org. Biomol. Chem.*, 2011, **9**, 1314; (*b*) Y. Ooyama, A. Matsugasako, K. Oka, T. Nagano, M. Sumomogi, K. Komaguchi, I. Imae and Y. Harima, *Chem. Commun.*, 2011, **47**, 4448; (*c*) Y. Ooyama, H. Egawa and K. Yoshida, *Eur. J. Org. Chem.*, 2008, 5239; (*d*) Y. Ooyama, A. Matsugasako, T. Nagano, K. Oka, K. Kushimoto, K. Komaguchi, I. Imae and Y. Harima, *J. Photochem. Photobiol., A*, 2011, **222**, 52; (*e*) Y. Ooyama, H. Egawa and K. Yoshida, *Dyes Pigm.*, 2009, **82**, 58.

7 (*a*) J. O. Moon, Y. H. Kim, M. G. Choi and S. K. Chang, *Bull. Korean Chem. Soc.*, 2011, **32**, 3517; (*b*) Y. H. Kim, Y. K. Han and J. M. Kang, *Bull. Korean Chem. Soc.*, 2011, **32**, 4244; (*c*) Y. H. Kim, M. G. Choi, H. G. Im, S. Ahn, W. Shim and S. K. Chang, *Dyes Pigm.*, 2012, **92**, 1199.

8 Q. L. Deng, Y. L. Li, J. H. Wu, Y. Liu, G. Z. Fang, S. Wang and Y. K. Zhang, *Chem. Commun.*, 2012, **48**, 3009.

9 (*a*) S. Cha, M. G. Choi, H. R. Jeon and S. K. Chang, *Sens. Actuators, B*, 2011, **157**, 14; (*b*) S. Ishihara, J. Labuta, T. Sikorsky, J. V. Burda, N. Okamoto, H. Abe, K. Arigaaf and J. P. Hill, *Chem. Commun.*, 2012, **48**, 3933; (*c*) C. Pinheiro, J. C. Lima and A. J. Parola, *Sens. Actuators, B*, 2006, **114**, 978; (*d*) K. N. Kim, K. C. Song, J. H. Noh and S. K. Chang, *Bull. Korean Chem. Soc.*, 2009, **30**, 197; (*e*) H. Hisamoto, Y. Manabe, H. Yanai, H. Tohma, T. Yamada and K. Suzuki, *Anal. Chem.*, 1998, **70**, 1255; (*f*) K. N. Kim, K. C. Song, J. H. Noh and S. K. Chang, *Bull. Korean Chem. Soc.*, 2009, **30**, 197.

10 G. Men, G. Zhang, C. Liang, H. Liu, B. Yang, Y. Pan, Z. Wang and S. Jiang, *Analyst*, 2013, **138**, 2847.

11 S. Pal, B. Sen, M. Mukherjee, K. Dhara, E. Zangrando, S. K. Mandal, A. R. Khuda-Bukhsh and P. Chattopadhyay, *Analyst*, 2014, **139**, 1628.

12 (*a*) S. Sen, T. Mukherjee, B. Chattopadhyay, A. Moirangthem, A. Basu, J. Marek and P. Chattopadhyay, *Analyst*, 2012, **137**, 3975; (*b*) S. Sen, S. Sarkar, B. Chattopadhyay, A. Moirangthem, A. Basu, K. Dhara and P. Chattopadhyay, *Analyst*, 2012, **137**, 3335; (*c*) S. Sen, T. Mukherjee, S. Sarkar, S. K. Mukhopadhyay and P. Chattopadhyay, *Analyst*, 2011, **136**, 4839; (*d*) U. C. Saha, K. Dhara, B. Chattopadhyay, S. K. Mandal, S. Mondal, S. Sen, M. Mukherjee, S. V. Smaalen and P. Chattopadhyay, *Org. Lett.*, 2011, **13**, 4510; (*e*) U. C. Saha, B. Chattopadhyay, K. Dhara, S. K. Mandal, S. Sarkar, A. R. Khuda-Bukhsh, M. Mukherjee, M. Helliwell and P. Chattopadhyay, *Inorg. Chem.*, 2011, **50**, 1213; (*f*) K. Dhara, U. C. Saha, A. Dan, M. Manassero, S. Sarkar and P. Chattopadhyay, *Chem. Commun.*, 2010, **46**, 1754.

13 P. Du and S. J. Lippard, *Inorg. Chem.*, 2010, **49**, 10753.

14 (*a*) M. Mukherjee, B. Sen, S. Pal, M. S. Hundal, S. K. Mandal, A. R. Khuda-Bukhsh and P. Chattopadhyay, *RSC Adv.*, 2013, **3**, 19978; (*b*) S. Sen, S. Sarkar, B. Chattopadhyay, A. Moirangthem, A. Basu, K. Dhara and P. Chattopadhyay, *Analyst*, 2012, **137**, 3335; (*c*) H. Paul, T. Mukherjee, M. G. B. Drew and P. Chattopadhyay, *J. Coord. Chem.*, 2012, **65**, 1289; (*d*) U. C. Saha, B. Chattopadhyay, K. Dhara, S. K. Mandal, S. Sarkar, A. R. Khuda-Bukhsh, M. Mukherjee, M. Helliwell and P. Chattopadhyay, *Inorg. Chem.*, 2011, **50**, 1213.

15 B. Sen, M. Mukherjee, S. Pal, K. Dhara, S. K. Mandal, A. R. Khuda-Bukhsh and P. Chattopadhyay, *RSC Adv.*, 2014, **4**, 14919 and ref. therein.

16 (*a*) Z. Zhou, M. Yu, H. Yang, K. Huang, F. Li, T. Yi and C. Huang, *Chem. Commun.*, 2008, 3387; (*b*) S. Saha, P. Mahato, U. G. Reddy, E. Suresh, A. Chakrabarty, M. Baidya, S. K. Ghosh and A. Das, *Inorg. Chem.*, 2012, **51**, 336.

17 S. Santra and S. K. Dogra, *Chem. Phys.*, 1998, **226**, 285.