An efficient 2-aminothiazolesalicylaldehyde Fluorescent Chemosensor for Fe²⁺ ion Detection and Potential Inhibitor of NUDT5 Signaling Hormone for Breast Cancer Cell and Molecular Keypad Lock Application

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Abstract

A novel thiazole phenol conjugate 2-aminothiazolesalicylaldehyde (receptor1) was designed and synthesized for the first time through a single step process via schiff base condensation reaction. The formation of receptor1 was confirmed by FTIR, ^{13}C NMR and ^1H NMR. The receptor1 complexing with various metal ions studied through fluorescence spectroscopy showed its selectivity towards Fe^{2+} ion following a reverse photo induced electron transfer (PET) process compared to all other potentially competing ions. Furthermore, the receptor1 showed excellent reversibility property on adding EDTA solution. The binding affinity between receptor1- Fe^{2+} ion was found to be $K_a = 2.22 \times 10^2 \text{ M}^{-1}$ and the sensing affinity of receptor1 towards Fe^{2+} ion at nanolevel concentration was $33.7 \times 10^{-9} \text{ M}$ which is lower than the detection limit of existing Schiff base. The receptor1 was used to detect Fe^{2+} ion in different water samples. Using molecular docking the chelating function of ligand (receptor1) was assessed as a potential inhibitor of NUDT5, a silence hormone signaling for breast cancer. The excellent reversibility property is used to construct INHIBIT molecular logic gate.

Introduction

The fluorometric studies have great impact on the detection of heavy metal ion in the biological and environmental system in recent years [1]. Iron is the most abundant metal found in the earth's crust next to aluminium. It is a major mineral required for human body for the development of immune system, synthesis of haemoglobin and transfer of electrons in photo physical process [2-6]. Due to its significant catalytic property iron is used as a catalyst in numerous reactions such as DNA and RNA synthesis, enzyme catalysis, and cellular metabolism [7]. Intake of excess iron may cause neuro degeneration such as Parkinson's disease, Alzheimer's, Huntington's and Menke's diseases [8, 11]. On the other hand, iron deficiency reduces the defence mechanism that paves a way for the entry of deadly viruses that cause diseases like pneumonia, typhoid, COVID-19 and so on. Its deficiency may also cause liver damages, diabetes, anaemia, kidney damages and heart diseases [9-13]. In biological system the distinction between ferric and ferrous ion is rather difficult in the detection process of iron [14-22], the schiff base compound like 2-aminothiazolesalicylaldehyde plays a major role in detecting the ferric ion in particular. In schiff base compound the presence of π -conjugated aromatic chromogenic group helps to sense cation and anion molecule [23]. Among the entire heterocyclic group, the sulphur containing thiazole group has a greatest impact in medicinal field due to its anti-cancer, anti-tubercular and antimicrobial property [24]. For the past few years biosensor, chromogenic sensor, mass sensor, electrochemical sensor was reported for sensing toxic metals in the environment [25]. Among these techniques fluorescent chemosensor method is highly efficient due to its specific selectivity of ion, responds for both solid and liquid phase, responds at short time and cost efficient [26]. In this article, 2-aminothiazolesalicylaldehyde (receptor1) was synthesized by conjugating thiazole with salicylaldehyde to form C=N imine based schiff base compound, which showed fluorescence quenching with Fe^{2+} ion through a probable reverse photo induced electron transfer (PET) process. Therefore, the reversibility property (turn on-off-on-off) of receptor1 with Fe^{2+} and EDTA was favourable for establishment of INHIBIT molecular logic gate and

molecular keypad lock. The receptor1 was subjected to the detection of Fe^{2+} ion in real water sample analysis. The synthesized receptor1 was assessed for its ability to interact with protein receptor through molecular docking, which is an important tool for drug discovery. The receptor1 was docked with NUDT5 (PDB ID: 5NWH) which is a hormone, signaling for breast cancer cells.

Experimental

Materials and instruments

Grade quality solvents and reagents were purchased commercially. Highly purified compound such as 2-aminothiazole and salicylaldehyde was purchased from Hi-media. The ^1H and ^{13}C NMR spectra were recorded from Bruker 400 MHz and 100 MHz spectrometer using DMSO-d_6 solvent and Tri methyl silane (TMS) as an internal standard. Absorption studies were carried out using Shimadzu UV-240 spectrophotometer. The fluorescence study was recorded on Jasco FP-8200 spectro fluorimeter. The absorption and emission spectra were recorded at $24 \pm 1^\circ\text{C}$.

Synthesis of 2-aminothiazolesalicylaldehyde

2-Aminothiazole (200 mg, 1 mmol) was added to salicylaldehyde (0.5 mL, 1 mmol) dissolved in 30 mL of methanol. To this mixture 3-4 mL of glacial acetic acid was added with constant stirring for 17 hours at room temperature [27]. The preliminary test for the product formation was assessed initially by thin-layer chromatography. After 15 days the evaporation of the solvent was noticed along with oily brownish yellow layer on the reaction mixture and after 22nd day the reaction mixture turned into brownish yellow coloured compound. The formation of 2-aminothiazolesalicylaldehyde (Scheme 1) here after named as receptor1 was confirmed by FTIR, ^{13}C NMR and ^1H NMR. The yield of the product was 88%.

UV-Vis absorption studies

The UV-Vis absorption studies were carried out to fix the excitation wavelength for the synthesized compound and thereby assess the photo physical property through fluorescence studies. The stock solution was prepared by dissolving receptor1 (2 mg) in 5 mL of DMF having concentration of 2×10^{-3} M. About 50 mM of HEPES (4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid) buffer dissolved in DMF and H_2O at 1:1 ratio was used to maintain the pH. The concentrated solution of 2×10^{-5} M and 2×10^{-6} M was prepared from the stock solution to observe the change in absorbance. The study was carried out at neutral pH 7.4.

Fluorescence titration

To carry the fluorescence titration the stock solution of receptor1 was prepared at the concentration of 4×10^{-6} M (2 mg, 0.01 mmol). The metal nitrate salts of 4×10^{-6} M concentration of Ag^+ , Zn^{2+} , Fe^{2+} , Fe^{3+} , K^+ , Li^+ , Na^+ (1 mg, 0.01mmol) Ba^{2+} , Ca^{2+} , Co^{2+} , Cu^{2+} , Ni^{2+} , Sr^{2+} (2 mg, 0.01mmol) Al^{3+} , Pb^{2+} , Hg^{2+} , Zr^{2+} , Mg^{2+} , Mn^{2+} (3 mg, 0.01mmol) Bi^{3+} , Cr^{3+} (4 mg, 0.01mmol) Cd^{2+} (7 mg, 0.01mmol) was weighed and

dissolved into 25 mL of DMF-H₂O, 1:1 v/v, HEPES 50 mM buffer solution. To the 2 mL of receptor1, 2 mL of metal solution was added to give 100 equivalents. The fluorescence intensity was recorded for each metal bound with receptor1 at the excitation wavelength of (λ_{ex}) 275 nm.

Competition with other metal ions

The interference of other metal ions during the sensing of Fe²⁺ using receptor1 was studied by incorporating various metal ion solution of 100 equivalents of each metal salt (0.01 mmol) (M = Ag⁺, Al³⁺, Ba²⁺, Bi³⁺, Ca²⁺, Cd²⁺, Co²⁺, Cr³⁺, Cu²⁺, Fe³⁺, Hg²⁺, K⁺, Li⁺, Mg²⁺, Mn²⁺, Na⁺, Ni²⁺, Pb²⁺, Sr²⁺, Zr²⁺ and Zn²⁺) dissolved in 25 mL of (DMF-H₂O, 1:1 v/v, HEPES 50 mM) buffer solution. The fluorescence intensity was recorded at the excited wavelength (λ_{ex}) 275 nm. In each vial 2 mL of receptor1 and 2 mL of each metallic solution was added and fluorescence was noted. To each vial 2 mL of Fe²⁺ ion solution was added to assess the interference behaviour.

Job's plot measurements.

To assess the stoichiometry of the receptor1-Fe²⁺ complex equal concentration of receptor1 (host) and Fe²⁺ (guest) of strength 1x10⁻⁶ M was prepared. The receptor 1 and the metal ion solution were mixed at the ratio of 0:4, 0.1:3.9, 0.2:3.8, 0.3:3.7, 0.4:3.6, 0.5:3.5, 0.6:3.4, 0.7:3.3, 0.8:3.2, 0.9:3.1 and 1:3 mL. The fluorescence analysis was carried out by transferring 4 mL of above solution to cuvette. The binding stoichiometric was evaluated using Jobs plot [28] and nonlinear fitting curve.

Quantum yield measurement

The quantum yield (Φ) was calculated using quinine sulphate ($\Phi_R = 0.54$) as a standard, the equation for calculating quantum yield [29, 30] is

$$\Phi_U = \Phi_R \times F_U/F_S \times A_S/A_U \times \eta_U^2/\eta_S^2 \quad (1)$$

where, Φ_R is standard value (Quinine Sulphate), F_U and F_S is the integrated fluorescence intensity of unknown and standard sample, A_S and A_U is absorbance of standard and unknown sample and η_U^2 is refractive index of solvent in which unknown sample was dissolved (1.4305) and η_S^2 is the refractive index of solvent in which standard was dissolved (1.33). The quantum yield of receptor1 (Φ_U) is 0.0126 and for receptor1-Fe²⁺ is 0.0036 at 275 nm.

Docking parameters

Molecular docking plays a vital role in drug discovery. It is a useful platform for predicting the protein ligand interaction through binding mechanism. The structure of the protein-ligand was accomplished using Schrodinger maestro software. Structure of the schiff bases was sketched using 2D-chemsketch. The docking study exposes the most suitable binding sites in the target protein for the ligand to bind. The molecular docking is a flexible process where it does not requires any prior knowledge on ligand

conformation but this docking study helps to gain information about the active site of the synthesized ligand (receptor1). The receptor1 docks with NUDT5, a silence hormone signaling for breast cancer cell. NUDT5 is a nucleotide metabolizing enzyme linked with ADP ribose and 8-oxoguanine metabolism. NUDT5 influence on the hormones that regulates the gene pool and increases the breast cancer cells. Here, the structure based receptor1 bound with NUDT5 could inhibit the activity of NUDT5 and thereby inhibit the proliferation of breast cancer cells.

Results And Discussion

Design and synthesis of 2-aminothiazolesalicylaldehyde

Receptor1 was synthesized by condensation of 2-Hydroxy benzaldehyde and 2-amino thiazole in methanol with 88% yield as shown in scheme 1 and characterized by ^1H -NMR and ^{13}C -NMR. In receptor1 benzaldehyde group acts as fluorophore, imine group act as spacer, and heterocyclic thiazole group containing N and S atom acts as a binder. The imine group links the fluorophore and binding unit. From FTIR analysis the presence of skeletal stretching of aromatic group is confirmed from the peak at 1600.92 cm^{-1} , the peak at 1192.21 cm^{-1} showed C-O stretching vibration of alcohol and the peak at 1492.00 cm^{-1} confirms C-C stretching vibration, the peak at 756.00 cm^{-1} confirms C-S stretching vibration, the peak at 2742.78 cm^{-1} confirms C-H stretching vibration. The peak at 1658.78 cm^{-1} confirms the presence of C=N stretching band (Fig. S1) [31]. The total number of carbon was confirmed by ^{13}C NMR. The chemical shift were noticed at (100 MHz, DMSO: d_6 , ppm) 130.14, 122.72, 128.32, 117.70, 154.62, 128.53, 140.48, 139.36, 128.96, 129.07. The total number of hydrogen was confirmed by ^1H NMR. The chemical shift of ^1H NMR (400 MHz, DMSO: d_6 , ppm) (J , Hz): 7.01, 7.12, 7.15 & 7.17 (d, 4H, Ar-H), 6.94 (s, 1H, CH=N), 6.85&6.94 (d, 2H, CH_2) and 9.23 (s, 1H, 1-OH) (Fig. S2 & S3) [32]. Analytically Calculated for $\text{C}_{10}\text{H}_8\text{N}_2\text{SO}$ (204.23): C, 120.10; H, 8.06; N, 28.01.

Selectivity Studies

The selectivity studies are performed to assess the sensing behaviour of receptor1 for a selective detection of metal ion. The metal ions such as Ag^+ , Al^{3+} , Ba^{2+} , Bi^{3+} , Ca^{2+} , Cd^{2+} , Co^{2+} , Cr^{3+} , Cu^{2+} , Fe^{2+} , Fe^{3+} , Hg^{2+} , K^+ , Li^+ , Mg^{2+} , Mn^{2+} , Na^+ , Ni^{2+} , Pb^{2+} , Sr^{2+} , Zr^{2+} and Zn^{2+} are taken in the form of nitrate salts and added to receptor1 and buffer solution. The excitation wavelength is fixed as 275 nm from UV-Vis spectroscopy. The receptor1 responded to a drastic quenching effect with Fe^{2+} and Ag^+ ions as shown in (Fig.1). However, other metal ions showed no significant changes during the identical condition. As a result, the receptor1 showed a potential response for Fe^{2+} ion with a turn on-off fluorescence through a possible reverse PET mechanism.

Proposed Binding Mechanism

In PET based sensors, a fluorophore is usually connected via a spacer to a binding unit containing a relatively high energy non-bonding electron pair found in nitrogen donors, capable of transferring an

electron to the excited fluorophore to quench its fluorescence (off-on) [33]. The quenching takes place because of the deactivation of the excited state of fluorophore by the addition of electron to its excited state frontier orbitals from the functional group of the fluorophore that quench the fluorescence intramolecularly [34]. This leads to the nonemissive state of fluorophore. Here, in receptor1 the free electron pair of nitrogen present in thiazole and C=N group attached to the fluorophore has strong fluorescence emission, and binding of cation results in weak fluorescence emission because the Fe^{2+} ion bound to receptor1 is paramagnetic in nature with unfilled d-subshell that strongly quench the fluorescence intensity of the fluorophore through electron transfer between receptor1 and Fe^{2+} ion [35]. The Fe^{2+} ion bound to the fluorophore acts as a signal transduction unit and also bound with a thiazole moiety and spacer [36]. There is no possibility of Fe^{2+} ion to bind with sulphur because of the weak interaction between Fe^{2+} and sulphur [37]. Therefore, the quenching response of receptor1 towards Fe^{2+} ascribed to electrons of fluorophore transferring to binding unit by metal ion complexation (reverse-PET process), resulting in fluorescence quenching (Scheme 2) [38].

Interference Study

Interference study was carried out to predict if any competitive metal ion interferes during complexation of receptor1 with Fe^{2+} . Fig.2. shows a change in the fluorescence intensity with respect to the addition of metal ion to receptor1. The emission intensity of receptor1 with various metal ion such as Al^{3+} , Ba^{2+} , Bi^{3+} , Ca^{2+} , Cd^{2+} , Co^{2+} , Cr^{3+} , Cu^{2+} , Fe^{3+} , Hg^{2+} , K^+ , Li^+ , Mg^{2+} , Mn^{2+} , Na^+ , Ni^{2+} , Pb^{2+} , Sr^{2+} , Zr^{2+} and Zn^{2+} is on par to the emission intensity of receptor1 alone, whereas the emission intensity of receptor1- Ag^+ is low compared to the emission intensity of other receptor1-metal ion complex. This is because the electronic configuration of Ag^+ ion in 4d subshell is completely filled and results in diamagnetic property, which leads to the decrease in emission intensity as there is no transfer of electron taking place between Ag^+ and receptor1. The response of receptor1 specifically towards Fe^{2+} is based on HSAB (Hard-Soft and Acid-Base) theory, the borderline base (C=N) group is able to coordinate with a borderline acid Fe^{2+} ion [39, 40]. The receptor1 showed a quenching in fluorescence intensity when it is complexed with Fe^{2+} is due to the reverse PET mechanism. The tested interfering metal ions showed no obvious interference when receptor1 is complexed with Fe^{2+} ion (Fig. 2). This result implies that receptor1 could be used for the selective detection of Fe^{2+} ion in various applications.

Binding and stoichiometry studies

In order to find the binding ability of receptor1 with Fe^{2+} ion, the fluorescence study was carried out by gently increasing the concentration of Fe^{2+} ion from 0-85 equivalents. A decrease in fluorescence intensity was observed with increase in Fe^{2+} ion concentration (Fig. 3). The fluorescence intensity was almost saturated upon addition of 85 equivalents of Fe^{2+} . The binding ratio of ligand and complex was predicted by Job's plot. The Job's plot confirms the binding mode of the host-guest complex, the maximum mole fraction of complex was 0.5 at excitation wavelength 275 nm (Fig. 4). Job's plot

concludes that the receptor1 interacts with Fe^{2+} at 1:1 stoichiometry. The stoichiometry binding of receptor and metal was predicted theoretically with the help of Benesi-Hildebrand non-linear fitting curve (Fig. 5) [41-43]. The association constant (K_a) of receptor1- Fe^{2+} ion is $2.22 \times 10^2 \text{ M}^{-1}$. The limit of detection (LOD) calculated using $3\sigma/S$ is $33.7 \times 10^{-9} \text{ M}$, where σ is the standard deviation of the blank signal, and S is the slope of the linear calibration plot indicating correlation coefficient (R^2) value of 0.9863 derived from the linear graph plot (Fig. 5).

Reversibility studies

The synthesized chemosensor has a very good reversible property that could be applied for various applications. The reversible property of receptor1 is analysed using a chelating agent EDTA (Ethylenediaminetetraacetic acid). Figure 6a. depicts the reversible property of receptor1. The fluorescence intensity is very high for receptor1. On addition of Fe^{2+} ion to the receptor1 the fluorescence intensity is quenched due to the complexation of receptor1 with Fe^{2+} ion. Further on addition of EDTA to receptor1 and Fe^{2+} ion solution the fluorescence intensity is enhanced. This results shows that receptor1 has a good reversible property. This is confirmed further by the addition of Fe^{2+} ion and EDTA several times to assess the reproducibility of the reversible property. The change in intensity with respect to Fe^{2+} and EDTA is due to the complexation of EDTA and Fe^{2+} ion with receptor1. The cycle was repeated for 10 times (Fig. 6b).

Effect of pH and Time

The pH study is carried out to assess the potential of the synthesized receptor1 to work under various environmental conditions. The effect of pH on the receptor1 and receptor1- Fe^{2+} complex is done by adjusting the pH from acidic to basic (2 to 11) condition. The fluorescence intensity of receptor1- Fe^{2+} complex decreases in acidic pH and increases at neutral pH and again decreases at basic pH (Fig. 7). The decrease in fluorescence intensity at acidic pH is because the imine ($\text{C}=\text{N}$) group of receptor1 gets protonated due to the increase in H^+ ion concentration and cleavage may take place between the fluorophore and heterocyclic group that hinders the binding of Fe^{2+} ion with receptor1 [44]. On the other hand, even at basic condition the fluorescence intensity decreases. This is because of the deprotonation of H^+ ion from phenolic group of fluorophore due to the increase in the concentration of OH^- ion. After deprotonation, the Na^+ ion occupies the phenoxide group and results in O^-Na^+ which hinders the binding of Fe^{2+} ion with receptor1 leading to a decrease in fluorescence intensity. The change in fluorescence intensity at acidic and basic condition is due to the presence of fluorophore group in receptor1. The increased fluorescence intensity at 7.4 indicates that the receptor1 works at physiological pH 7.4 which is suitable for biological and environmental application. Hence all the other experiments are carried out at pH 7.4.

A study was carried out further to assess the time taken for the complex formation between receptor1 and Fe^{2+} [45]. The decrease in fluorescence intensity from 0 to 4th minute showed that the complexation

between receptor1 and Fe^{2+} ion was unstable. After 4th minute the saturation in graph indicates the stable complexation between receptor1 and Fe^{2+} ion. The solution was kept further for 60 minutes and the fluorescence intensity was noted. There were no desirable changes after four minutes (Fig. 8). Hence, the result shows that the receptor1 favours for detection of Fe^{2+} ion in short interval of time.

Application studies

Determination of Fe^{2+} ion in water sample

The synthesized receptor1 was subjected to the detection of Fe^{2+} ion in water sample. Three different water samples (Bore well water (B1-3 μM , B2-6 μM), Tap water (T1-3 μM , T2-6 μM), and RO drinking water (D1-3 μM , D2-6 μM)) are collected from Coimbatore Institute of Technology. The sample water is spiked with Fe^{2+} ion concentration of 3 and 6 μM . About 2 mL of 4×10^{-6} M concentration of receptor1 was added to the spiked water sample. The recovery percentage of Fe^{2+} ion from spiked water sample after adding receptor1 is calculated from fluorescence spectroscopy (Fig. S4). The receptor1 showed the enhanced fluorescence intensity but on adding the spiked water sample with receptor1 the quenching in fluorescence intensity is noticed. The difference in the fluorescence intensity is due to the complexation of Fe^{2+} ion with receptor1. The quenching in intensity proves that the receptor1 has the ability to attract Fe^{2+} ion from spiked water sample. The recovery percentage of Fe^{2+} ion for B1, B2 is 96%, 99.3%, for T1, T2 is 100.7%, 98.8% and for D1, D2 is 101.3%, 99.5% respectively (Table 1). The results proved that receptor1 could be practically applicable for selective detection of Fe^{2+} ion in real water sample analysis.

Table 1. Fe^{2+} ion recovered from real water sample spiked with Fe^{2+} ion using receptor1.

CIT Water Sample		Fe ²⁺ added (μM)	Fe ²⁺ analysed from fluorescence spectra (μM) mean ^[a] ± SD ^[b]	Recovery (%)	Relative error (%)
Borewell 1	water	3.0	2.88 ± 0.15	96.0	-4.0
Borewell 2	water	6.0	5.96 ± 0.05	99.3	-0.7
Tap water 1		3.0	3.02 ± 0.05	100.7	0.7
Tap water 2		6.0	5.93 ± 0.03	98.8	-1.2
RO drinking water 1		3.0	3.04 ± 0.14	101.3	1.3
RO drinking water 2		6.0	5.97 ± 0.06	99.5	-0.5

^a mean of three measurement.

^b standard deviation.

Molecular Logic Gate

The application of molecular logic gates has attracted the scientist in sensor field [46, 47]. The reversible property of receptor1 helped to construct a molecular logic gate using two binary inputs. The absence and presence of Fe²⁺ and EDTA was represented by '0' and '1' (i.e. presence of Fe²⁺ and EDTA as 1 and the absence of Fe²⁺ and EDTA as 0). The output is monitored at 420 nm. Based on the change in fluorescence intensity the truth table was constructed. The output '0' denotes the quenching intensity (fluorescence-off) and '1' denotes the enhanced intensity (fluorescence-on). On the basis of truth table the INHIBIT logic gate was constructed as shown in Figure 9. Thus the addition of Fe²⁺ to receptor1 shows

the weak fluorescence and on adding EDTA to receptor1-Fe²⁺ complex the fluorescence intensity is enhanced (Table 2).

Table 2. Truth table of INHIBIT logic gate

Input 1 (Fe ²⁺)	Input 2 (EDTA)	Output (λem 420nm)
0	0	1
1	0	0
0	1	1
1	1	1

Molecular docking Studies

Protein-ligand interaction plays a significant role in structure based drug designing. Molecular docking technique reveals the interaction between the ligand and protein [48]. The synthesized receptor1 acts as a ligand and it is subjected to molecular docking using Schrodinger maestro software to assess its interactions towards protein. The performance of receptor1 was high when it is docked with NUDT5, a macromolecule that act as a silence hormone signaling in breast cancer (PDB IB: 5NWH) [49]. The hydroxyl group of receptor1 interacts with the side chain residue ASP274. The hydrophobic interaction was noted between the thaizole moiety and benzene ring of the schiff base interacting with the amino acid residues such as LEU178, VAL174, LEU169, LEU164, PHE144, VAL147, LEU210, LEU208 and ALA273(Fig. 11). The docking score of receptor1 with protein is -5.23 kcal mol⁻¹. The docking score of net negative energy represents a stable complex. The stable complex between receptor1 and amino acid residues are held by means of electrostatic interaction. Such interactions are due to the distribution of asymmetric charges in the receptor1 molecule containing heteroatoms such as N, O, and S. The docking result indicates that the receptor1 has shown good affinity towards NUDT5 target. Thus, receptor1 can act as an inhibitor for blocking NUDT5 activity.

Molecular keypad lock

The excellent reversible property helped us to propose a sequence of code for molecular keypad to lock. Three different chemical input was given as receptor1 'R', Fe²⁺ metal ion 'F' and EDTA 'E'. The six feasible input fusions are RFE, REF, FRE, FER, ERF, and EFR. In these combination, RFE resulted for enhanced fluorogenic output whereas the other combination such as REF,FRE,FER,ERF,EFR result in quenching intensity as output as shown in Figure 10. The keypad contains the alphabet from A to Z and the password RFE unlocks the keypad. At the same time, the other codes results for wrong password. Due to this reason, this type of sensing will safeguard the molecular level information.

Conclusion

A schiff base receptor1 is synthesized using salicylaldehyde and thiazole moieties. The receptor1 is highly selective towards Fe^{2+} ion showing quenching with turn off-on process following reverse PET mechanism. The association constant of receptor1- Fe^{2+} ion is $2.22 \times 10^2 \text{ M}^{-1}$ and limit of detection (LOD) is $33.7 \times 10^{-9} \text{ M}$. The binding ratio of receptor1- Fe^{2+} is proposed to be 1:1 by Job's plot. The receptor1- Fe^{2+} ion are found to be reversible with EDTA solution. Based on this property, constructed a molecular logic gate with two inputs and one output and this type of receptor1 can be successfully applied for the mimicking behaviour of molecular keypad lock which has a great utilisation in security devices. The real time application of receptor1 is demonstrated in detection of Fe^{2+} ion in water samples and molecular docking. Receptor1 on docking with potent inhibitor of NUDT5 target shows the good docking score of $-5.23 \text{ kcal mol}^{-1}$. The new selective fluorescent receptor1 could be able to sense Fe^{2+} ion in biological and environmental system.

Abbreviations

FTIR - Fourier Transform Infrared Spectroscopy, ^{13}C NMR-Carbon-13 Nuclear Magnetic Resonance, ^1H NMR- Proton Nuclear Magnetic Resonance, PET- photo induced electron transfer, NUDT5- [Nudix hydrolases](#).

Declarations

I hereby declare that this is the original work done by the stated authors and has not been submitted elsewhere for publication.

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Scheme

Please see the Supplementary Files for the Scheme 1 and 2.

Figures

Figure 1

Fluorescence spectra to assess the response of various metal ion (100 equivalents. of each, $\lambda_{\text{ex}} = 275$ nm) towards receptor 1 of concentration 4×10^{-6} M solution (DMF- H_2O , 1:1 v/v, HEPES 50 mM, pH = 7.4)

Figure 2

Metal ions competition analysis of receptor1 (4×10^{-6} M) solution (DMF-H₂O, 1:1 v/v, HEPES=50 mM, pH=7.4) in the presence of various metal ions (100 equivalents. of each, $\lambda_{\text{ex}} = 275$ nm). The green bar represents the receptor1+ metal ion and red bar represents receptor1+metal ion+Fe²⁺ ion that shows the quenching fluorescence intensity

Figure 3

Fluorescence titration spectrum of receptor1 (4×10^{-6} M) solution (DMF-H₂O, 1:1 v/v, HEPES 50 mM, pH=7.4) upon addition of different concentration of Fe²⁺ (0-85 equivalents. $\lambda_{\text{ex}} = 275$ nm).

Figure 4

Job's plot of receptor1 and Fe^{2+} in DMF- H_2O solution (1:1 v/v, HEPES 50 mM, pH=7.4).

Figure 5

Benesi-Hildebrand plot of receptor1 (4 μM) with Fe^{2+} in DMF- H_2O solution (1:1 v/v, HEPES 50 mM, pH=7.4) ($\lambda_{ex} = 275$ nm).

Figure 6

a Reversibility spectra of receptor1 in the presence of Fe^{2+} ion and EDTA in DMF- H_2O solution (1:1 v/v, HEPES 50 mM, pH=7.4) ($\lambda_{\text{ex}} = 275$ nm). **b** Sequential addition of Fe^{2+} and EDTA showing reversibility behaviour.

Figure 7

Effect of change in pH of a) Receptor1 b) Receptor1- Fe^{2+} .

Figure 8

Time taken for the complexation of receptor1- Fe^{2+} in DMF- H_2O solution (1:1 v/v, HEPES 50 mM, pH=7.4) ($\lambda_{\text{ex}} = 275 \text{ nm}$).

Figure 9

Figure 10

Output for recepor1, corresponding to six possible input combinations of receptor1 series at 420 nm. Inset: a molecular keypad lock generating emission at 420 nm at correct password RFE entered. The keys R, F and E hold the relevant input receptor1, Fe^{2+} and EDTA, respectively.

Figure 11

2D Binding activity of receptor1 with NUDT5.

Supplementary Files

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