Stabilization and Protection of Biomaterial through Biomineralization via Surface Interaction: An Astrobiological Approach

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Abstract: A Cytosine (CYN), core constituent of both RNA and DNA, is a well-known functional biomaterial of living system. Adsorption of such molecule on primigentional mineral surfaces transacts a very specific and selective role in biocompatibility and biomineralization. The adsorption behaviour of CYN, with change in pH, concentration, time and temperature on siliceous Montmorillonite (MMT) surface, a prominent nanoclay of smectite group with specific surface area, was studied systematically and methodically using different spectral techniques (viz. UV, FTIR, SEM and XRD). Biogenic metal dication [M2+= Mg2+, Ca2+, Fe2+ and Cu2+] entrapped nanocomposites of MMT were prepared separately by cation-exchange approach. The results reveal significant role of MMT and biogenic metal ions on the quantity of CYN adsorbed. The adsorption isotherm evinces monolayer formation of the biomaterial on the surface of MMT and MMT-M2+ compounds. The Langmuir adsorption parameters (K_L & X_m) delineated appreciable interaction of CYN on MMT-Fe2+ surface as compared to MMT alone and other metal confined MMTs. The present work throws light on affinity of MMT nanoclay (having multifarious applications in biological systems) involving biogenic metal ions on stabilization and protection of biomaterials during prebiotic chemical evolution of biomolecules. The results embarks better adsorption capability of transition metal ion incorporated MMT than the clay without metal ion and with alkaline Earth dications due to better coordination properties of transition metal ions involving their d-orbitals for stable complex formation with negative sites of cytosine molecules as ligands. Since metal ions are key components of various enzymes which can catalyse a number of biochemical reactions in the living systems, the present study give better understanding regarding the pathways of aggregation of biomonomers on solid surfaces which themselves might have catalysed the formation of complex biomolecules via polymerization and condensation in enzyme free environment of early Earth and also influences implant biocompatibility.

Keywords: Adsorption; Cytosine; Montmorillonite; Biogenic metal ions; Langmuir adsorption parameters; Prebiotic chemical evolution.

1. Introduction

Life can be seen as the result of chemical activity of biocompatible and water soluble bionanoassemblies such as proteins, nucleic acid bases, sugars and phosphate molecules. In early periods of the Earth, Life is supposed to be originated from lifeless environment through chemical evolutionary processes. Proteins and nucleic acids constitute the main matrix of living systems. How
did these complex biomolecules, capable of self-replication and self-organisation, appear abiotically on primitive Earth from their monomeric building blocks is a question of paramount importance [1-8]. The fundamental pathways from non-living matter to life forming molecules, starting with the build-up of nucleobases, over the synthesis of nucleotides and finally to the formation of RNA and DNA, are still shrouded in mystery.

Bernal in 1951 independently speculated and stressed the potential importance of clays as solid mineral surfaces for concentration of biomolecules via adsorption [9]. Clay minerals have the capacity to concentrate, protect and catalyse the polymerisation of enormous organic molecules due to their fine particle size, specific surface area and charge characteristics [10-14]. Recently, layered clay minerals are reported to be advantageous for adsorption [15-18] and phyllosilicates containing Fe-Mg strongly bind the nucleotides by exchanging their ligands and thereby preserving the genetic information inside the inorganic templates. In succession the polymer formation needs excessive assemblence of nucleotides under favoured conditions of thermodynamics [19, 20].

Montmorillonite a prominent nanoclay with confined metal dication especially known to have particular affinities for organic molecules by adsorbing them on and in between their silicate layers [21-24]. Metal entrapped smectite clays provide good adsorbent sites for the frequent concentration of biologically active molecules [25-28] and also can act as catalyst for their polymerization as it might have happened on contemporaneous primeval Earth [29-35]. In silico surface interaction studies of nucleobases, nucleotides and their polymers with MMT and boron nitride nanotubes were also reported [36-39] along with electronic properties of metal semiconductor structures with cytosine biopolymer [40,41].

An extensive contemplation of the affinities of clay minerals towards surface interaction of biomolecules has fascinated the minds of great thinkers in past few decades [42-49, 50 (and references therein)]. Currently, several inquisitive researchers are engaged in exploring various aspects of MMT nanoclay as a drug carrier system with multitudinous applications and its biocompatibility outcomes [51-54]. Among all four nucleic acid bases, cytosine (CYN), a potent biomolecule and a viable functional biogenic material [55], seems to be significant from surface interaction point of view during the course of formation of genetic macromolecules (DNA and RNA) and their monomers [56-58]. Therefore, CYN is taken as model for adsorption studies on the prominent nanoclay MMT, with and without entrapped metal diions [Mg$^{2+}$, Ca$^{2+}$, Fe$^{3+}$ and Cu$^{2+}$], which may relevant to stabilization and protection of biomolecule through biomineralization in prebiotic scenario of Earth.

Metal ions have been associated with biological systems for billions of years, through the interplay of biogeochemical weathering and metabolic pathways involving the products of that weathering. A number of metal ions play a vital role in a vast number of widely differing biological processes essential for the activity of enzymes [59]. All life forms have an absolute requirement for metals, as metals play critical roles in fundamental processes, including osmotic regulation, catalysis, metabolism, biomineralization, and signalling [60]. It is increasingly recognized that metals are involved in cellular and subcellular functions and also used as inorganic drugs for many diseases [61].

The present study throws light upon adsorption of CYN on prebiotically available MMT and its biogenic metal dication modified nanocomposites showing the inevitability of adsorption phenomenon in primitive era of the Earth as well as its biological implications.

2. Materials and Methods

2.1. Procurement of chemicals

Analytical grade reagents were used during all experimentation. Montmorillonite (MMT K10) was procured from Sigma Aldrich, Germany; Cytosine (CYN) from Loba Chemie, India; CaCl$_2$ from CDH, India and H$_2$O$_2$, MgCl$_2$, FeCl$_3$, CuCl$_2$ from Qualigens, India. All solutions were prepared in triple distilled deionised water ($\mu$H$_2$O = pH 6.8±0.2) and borosilicate glasswares were used throughout the work.
2.2. Preparation and purification of sensitizers/adsorbents

Preparation and purification of MMT and MMT-M²⁺ i.e., metal [Mg²⁺, Ca²⁺, Fe²⁺ and Cu²⁺] entraped MMT nanocomposites were carried out as described in earlier reports [25, 28, 62]. After drying in vacuum desiccators the adsorbents were grounded, sieved to mesh sized for further experimentation (Scheme I).

Scheme I. Schematic presentation of various steps involved in the preparation as well as purification of adsorbents and their characterization before and after adsorption.

2.3 Experimental procedure

The adsorption studies of CYN (Figure 1) separately on MMT and MMT confined with biogenic metal ions [Mg²⁺, Ca²⁺, Fe²⁺ and Cu²⁺] were carried out in aqueous environment as a function of pH (4.0-9.3) and concentrations (1.0×10⁻⁶M – 9.0×10⁻⁴M) of adsorbate at room temperature in order to obtain saturation point by adding relevant buffer i.e., acetic acid-acetate (pH 4.0-6.0) or boric acid-borax (6.6-9.3), to the adsorbate solution (3ml each) containing different adsorbents MMT/MMT-M²⁺ [M²⁺= Mg²⁺, Ca²⁺, Fe²⁺, Cu²⁺] (30mg each), kept in separate conical flasks (10 ml), stirred mechanically for 15min and allowed to stand at room temperature to find out the adsorption capability of the given adsorbents.
In each experiment the adsorption of CYN was measured after a period of 18hrs. The suspensions were then centrifuged at 3500rpm for 30min and the supernatant solutions were decanted for subsequent quantitative estimation, leaving the adsorption adduct as residue and the pH of the solution was again noted which was found to be almost unaffected. Adducts were separated, washed and then dried under vacuum at ambient temperature and used for further analytical investigations. The complete procedure was triplicated and the formation of degradation product after adsorption (if any) was checked by chromatographic techniques (PC and HPLC) [28].

2.4. Analysis and characterization

2.4.1. Properties of adsorbent
The colour of MMT K10 used as adsorbent was found 10R8/1 (White) on Munsell Notations with water holding capacity (W.H.C.) 95%, pH 3.6 and cation exchange capacity (CEC) 200.17 meq/100g.

2.4.2. Absorbance measurements
The different concentrations of CYN in supernatant were determined by recording their absorbances at $\lambda_{\text{max}} = 263$ nm by using Systronics double beam UV/Vis spectrophotometer 2203smart and quartz cuvette with 1cm path length. pH was recorded on Hanna pH meter (model pHep) with an accuracy of $\pm0.1$ pH unit.

The amount of CYN adsorbed under optimized conditions was calculated from the difference between initial concentration and concentration after adsorption in each case. The equilibrium concentration of CYN and the amount adsorbed were used to obtain adsorption isotherms [28] (Figure 2).
2.4.3. Morphological observation

Scanning electron microscopy (SEM) was used to observe the surface morphology of adsorbate, adsorbents with or without divalent metal ions and their respective adducts using a Hitachi S-3700N instrument with an acceleration voltage 15.0kV, take-off angle 44.1° and ZEISS EHT 20kV.

2.4.4. Relative intensity and inter-planar spacing identification

X-ray diffraction (XRD) studies were performed to check the crystalline nature as well as to confirm purity and to know the relative intensity and inter-planar spacing. A Thermal X-TRA ARL diffractometer equipped with a ‘Peltier’ detector, using Cu Kα radiation (wavelength=1.5406Å) operated at 30mA and 40kV with graphite monochromator detection between the range 10-80° 2θ angle was used for powder diffraction analysis of adsorbents and adsorption adducts.

2.4.5. Vibrational studies

A PerkinElmer DX-II FT-IR spectrophotometer equipped with DTGS detector was used to determine the IR adsorption frequencies. The data were collected at 4cm⁻¹ resolution and 100 scan with frequency range 400-4000cm⁻¹.

3. Results and Discussion

This Surface interaction studies of CYN on MMT and on MMT-M²⁺ [M²⁺= Mg²⁺, Ca²⁺, Fe²⁺, Cu²⁺] has been carried out under varied conditions of pH (4.0-9.3) and concentration (10μM-90μM) at room temperature (298K) to figure out the possible affinity of the clay and its biologically active metal entrapped nanocomposites in abiotic formation of complex biomolecules and their stabilization and protection through biomineralization (Figure 3) in relevance to origin of life.

Figure 3. Surface interaction of biomaterial CYN on the nanoclay MMT and biogenic metal entrapped MMT (i.e., MMT-M²⁺) at optimized conditions of pH and temperature.

CYN can exist in two tautomeric forms (Figure 4), in which the amino form facilitate normal biological traits in living systems, while the imino form causes mutation (i.e., the precursors to many molecular-based diseases, including cancer) via forming a stable hydrogen bond, pairing with adenine instead of guanine. Therefore, amino being stable and less energetic state predominates in solution phase [63].
Protonation is known to increase the number of possible pairing modes leading to structural diversity [64]. In CYN molecule protonation takes place at N3 position because of the maximum electron density, while the amino group is electron deficient. The amino group of CYN is hydrogen donor, and the ring nitrogen atom N3 is hydrogen acceptor. CYN also has a hydrogen acceptor group at C2 position, thus can form three hydrogen bonds with guanine. The hydrogen-bonding patterns of bases have important consequences for the three-dimensional structure of nucleic acids. Further, deamination of CYN can lead to the formation of uracil, which pairs with A and implement RNA replication or 5-methyl cytosine can be converted directly to T. The functional role of the exocyclic amino group of CYN is aminating pyruvate (an α-keto acid) to the corresponding amino acid alanine, in a hydrophobic, reducing microenvironment, without the need for extreme pH [65].

### 3.1. Adsorption (steady state observations)

The standard concentration curve plotted between optical absorbance versus molar concentration of various CYN solutions monitored at their absorption maxima ($\lambda_{max}$=263nm) showed the amount of CYN adsorbed during adsorption (Figure S1). Linear relationship between absorbance and concentration of CYN has been observed showing $r^2 = 0.99$. Preliminary studies showed that the amount of nucleic acid base adsorbed is dependent on pH of the solution and was found maximum at pH 6.6 (Figure 5). Subsequent studies were therefore carried out at pH 6.6 and at room temperature.

Figure 5. Quantity (mg g$^{-1}$) of CYN adsorbed on MMT at different pH.
The maximum surface interaction of CYN at this pH may be attributed to the ionic interaction, as at this pH, CYN is in cationic form, while the surface of adsorbents (MMT/MMT-M²⁺) is negatively charged, so that, the specificity of CYN towards adsorption may be traced out. For adsorption study, 3 ml solution of adsorbate has been treated with 30 mg of adsorbent using 100 μl buffer solution. The influence of buffer solution on the optical density (OD) of different CYN solution has been investigated and no effect of buffer on the OD has been observed. Results are recorded in Tables (Table 1 and 2) and illustrated in Figures (Fig. 2, 3, 5-8).

3.2. Adsorption isotherm

The adsorption isotherm indicate the amount of CYN adsorbed on MMT/MMT-M²⁺ increases as their equilibrium concentration increases in solution up to saturation point, above which the adsorption become independent of adsorbate concentration (Figure 6). Initially, plots show linear relationship where the coefficient of correlation, i.e., r² and the value 1.0 was taken as limit of significance.

\[
\frac{C_{eq}}{X_e} = \frac{1}{K_L X_m} + \frac{C_{eq}}{X_m}
\]

(1)

Where, \( C_{eq} \) is equilibrium concentration of the nucleic acid base, \( K_L \) is a constant related to enthalpy (AH) of adsorption (\( K_L \propto e^{-\Delta H/RT} \)), \( X_e \) the amount (mg) of solute adsorbed per gram weight of adsorbent and \( X_m \) the amount (mg) of solute required per gram weight of adsorbent for complete monolayer formation.

As shown in Figure 7 the Langmuir adsorption parameters \( X_m \) and \( K_L \) were calculated from the slope and the intercept obtained from the graph between \( C_{eq}/X_e \) vs \( C_{eq} \). \( X_e \) can be obtained asymptotically from Figure 6 on extrapolating the adsorption curve towards Y-axis when saturation phenomenon occurs. It was observed that the adsorption trend (% binding and Langmuir constants) of adsorbate adsorbed on MMT with or without entrapped dications largely depends on the nature of adsorbate as well as adsorbent.
The percent binding was calculated by the following equation (Equation 2):

\[
\text{% binding} = \left(\frac{C_{b2} - C_{af}}{C_{b2}}\right) \times 100.
\]

Where, \(C_{b2}\) and \(C_{af}\) are concentration before and after adsorption respectively. Table 1 shows the Langmuir constants and % binding for adsorption of CYN on MMT and different MMT nanocomposites at pH 6.6 and temperature 298K.

Table 1. Langmuir adsorption parameters and (%) binding of CYN on the surface of various adsorbents.

<table>
<thead>
<tr>
<th>Adsorbents</th>
<th>pH</th>
<th>% Binding</th>
<th>Langmuir Isotherm Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>(X_m) (mgg(^{-1}))</td>
</tr>
<tr>
<td>MMT-Fe(^{2+})</td>
<td>6.6±0.1</td>
<td>67</td>
<td>(0.47\times10^{-3})</td>
</tr>
<tr>
<td>MMT-Cu(^{2+})</td>
<td>6.6±0.1</td>
<td>60</td>
<td>(0.42\times10^{-3})</td>
</tr>
<tr>
<td>MMT</td>
<td>6.6±0.1</td>
<td>31</td>
<td>(0.22\times10^{-3})</td>
</tr>
<tr>
<td>MMT-Ca(^{2+})</td>
<td>6.6±0.1</td>
<td>20</td>
<td>(0.14\times10^{-3})</td>
</tr>
<tr>
<td>MMT-Mg(^{2+})</td>
<td>6.6±0.1</td>
<td>04</td>
<td>(0.29\times10^{-6})</td>
</tr>
</tbody>
</table>

In terms of % binding and \(X_m\) data, the effectiveness of various adsorbents in adsorption of CYN on MMT/MMT-M\(^{2+}\) was found in order (Figure 8):

\[\text{MMT-Fe}^{2+} > \text{MMT-Cu}^{2+} > \text{MMT} > \text{MMT-Ca}^{2+} > \text{MMT-Mg}^{2+}\]

The above results embarks better adsorption capability of transition metal ion incorporated MMT than the clay without metal ion and with alkaline Earth dications. It may be due to better coordination properties of transition metal ions involving their d-orbitals for stable complex formation with negative sites of cytosine molecules as ligands. With montmorillonite, adsorption is also influenced by the nature of the exchangeable cations [14]. The interactive forces, alone, cannot
solubly be responsible for this adsorption, as size of the metal ions should also be considered. MMT-Mg$^{2+}$ has shown the least affinity with CYN due to smaller size of the Mg$^{2+}$ ions, intercalated into the aluminosilicates. The adsorbate which are not intercalate between the clay layers, have the higher rate of desorption when treated with the water [42] and the same observation has also been found for MMT-Mg$^{2+}$ adsorption adduct of CYN.

Figure 8. Affinity of CYN towards different adsorbents.

Since metal ions are key components of various enzymes which can catalyse a number of biochemical reactions in the living systems [29-31, 33, 41, 59], the present study give better understanding regarding the pathways of aggregation of biomonomers on solid surfaces through surface interaction which themselves might have catalysed the formation of complex biomolecules via polymerization and condensation in enzyme free environment of early Earth which may influences implant biocompatibility [54].

Minerals may have a pivotal role in the prebiotic evolution of complex chemical systems including metals, which are known to be involved in many enzymatic reactions, where metal-binding proteins carry out at least one step in almost all biological pathways [68], so that, metal ions are deemed essential because without them biological function is impaired. Due to the abundance of iron on Earth and its role in biological systems, the transition and mineralogical stages of iron have played a key role in Earth surface systems and in marine geochemistry [69, 70].

Moreover, the characterization of the spectroscopic features of biomolecule-mineral complexes also provides a support in remote sensing spectroscopy for detecting organic compounds on planetary surfaces or cometary grains and asteroid surfaces [71, 72]. It is believed that nucleobases might have played a critical role at the dawn of life due to their photoprotective properties which indeed suggest that their structure could have been naturally selected for the ability to dissipate electronic energy through ultrafast photophysical decay [73, 74].

3.3. Physico-chemical characterization of adduct

3.3.1. SEM observations

The surface morphology of adsorbate (Figure 9a), adsorbents (Figure 9b&c) and adsorption adduct (Figure 9d) was characterized by scanning electron microscopy (SEM). The morphology depict that surface of adduct was finely covered after adsorption.
3.3.2. XRD pattern

The crystalline nature of clay was confirmed by X-ray diffraction analysis. The size of the intercalated metal ions (viz. K⁺, Na⁺, Mg²⁺, Ca²⁺, Fe²⁺, Cu²⁺) in the aluminosilicates influence the basal spacing of the structure. Further, water molecules are also allocated in the interlayer sheets and coordinated by these exchangeable cations, which in turn, also increases the basal spacing to a greater extent [75]. Percentage adsorption data have also shown that adsorption affinity increases in direct proportion with the increase of the size of the exchangeable metal ion (Table 1 and Fig. 8).

Apart from the size of the divalent metal ions, water coordination affinity of metal ions is also significant for this enhanced surface interaction of MMT with CYN [76]. Following, XRD data of the adsorbate, adsorbent and adsorption adduct also suggest the increase in the basal spacing of the adsorbents (Table 2 and Fig. 10).

Table 2. XRD data showing 2θ and basal spacing (Å) for adsorbent and adsorption adduct.

<table>
<thead>
<tr>
<th>S/N</th>
<th>Probe</th>
<th>2θ</th>
<th>d (Å)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>CYN</td>
<td>20.22</td>
<td>4.4577</td>
</tr>
<tr>
<td>2.</td>
<td>MMT</td>
<td>20.2</td>
<td>4.4629</td>
</tr>
<tr>
<td>3.</td>
<td>MMT-Fe²⁺</td>
<td>20.08</td>
<td>4.4876</td>
</tr>
<tr>
<td>4.</td>
<td>MMT-Fe²⁺-CYN</td>
<td>20.06</td>
<td>4.4915</td>
</tr>
</tbody>
</table>

CYN interact with different sites of the clay surface, i.e., the interlayer region and the faces of the lamella showing expansion of the interlayer was greater due to the inclusion of CYN. Results demonstrate the higher affinity of MMT-Fe²⁺ interlayer for CYN.
Figure 10. XRD graph of CYN, MMT, MMT-Fe$^{2+}$ and CYN+MMT-Fe$^{2+}$ adduct after adsorption.

Another reason for the maximum affinity of CYN with MMT-M$^{2+}$ is the affinity of M$^{2+}$ to form complexes (CYN-M$^{2+}$) with the inorganic divalent metal ions. Zaia et al. have shown that the interaction of NBs, cannot be regarded as simple physio-adsorption as NBs penetrate into the interlayer of the clays and oxidise Fe$^{2+}$ to Fe$^{3+}$ [77].

3.3.3. FTIR spectra

The above results were complemented by vibrational data where the possible interactive sites of CYN on MMT-Fe$^{2+}$ were investigated by FTIR spectra (Fig. 11). The absorption frequencies lying in between 1600-1800cm$^{-1}$ are important for sensitivity of molecular interactions between clays and nucleobases, as pyrimidines absorb strongly in this region [76]. The spectra of CYN set forth a large number of vibrational frequencies (Fig. 11a), where the IR bands at 3377.4cm$^{-1}$ and 1651.9cm$^{-1}$ corresponds to N-H stretching and C=O stretching respectively [45, 78], while band at 1537.4cm$^{-1}$ points out N-H bending. Bands at 792.2cm$^{-1}$ and 532.6cm$^{-1}$ could be attributed to C-H and OCN deformation.

Figure 11. FT-IR spectra of (a) CYN, (b) MMT, (c) MMT-Fe$^{2+}$ and (d) adsorption adduct, CYN + MMT-Fe$^{2+}$ after adsorption.
In the IR spectrum of MMT the band at 3623.1 cm\(^{-1}\) coincides –OH stretching and bands at 3444.9 cm\(^{-1}\) and 1634.8 cm\(^{-1}\) could be due to stretching and bending vibrations of water adsorbed in MMT, whereas the band at 1063.4 cm\(^{-1}\) adapts Si-O-Si stretching vibration [28] (Fig. 11b). After encapsulation of Fe\(^{2+}\) ions in MMT a shift displayed in frequencies from 3444.9 cm\(^{-1}\) to 3450.1 cm\(^{-1}\) and 1063.4 cm\(^{-1}\) to 1053.3 cm\(^{-1}\) respectively, while the band at 3623.1 cm\(^{-1}\) disappeared, revealing interaction of Fe\(^{2+}\) with MMT (Fig. 11c). This interaction may be due to the attractive forces between opposite charges of Fe\(^{2+}\) and the lone pair of electrons on oxygen or negative charges on silica layers binding alumina layer in between.

The IR spectrum manifests a complete sweep out of number of vibrational frequencies in the finger print region of CYN and a remarkable shift in various absorption frequencies from 3377.4 cm\(^{-1}\) to 3461.4 cm\(^{-1}\), 1651.9 cm\(^{-1}\) to 1633.6 cm\(^{-1}\), 792.2 cm\(^{-1}\) to 798.6 cm\(^{-1}\), 532.6 cm\(^{-1}\) to 526.9 cm\(^{-1}\) and 440.9 cm\(^{-1}\) to 466.6 cm\(^{-1}\) after adsorption of CYN on MMT-Fe\(^{2+}\) (Fig. 11d) explicitly indicate the interaction of CYN to MMT-Fe\(^{2+}\) particularly via N-H and C=O bonds. These consequences are in perfect accordance with the findings reported earlier [76, 79].

4. Conclusions

The present contribution credibly evinces the illusion of adsorption of CYN onto the siliceous material MMT along with its biogenic metal dications \([M^{2+} = Mg^{2+}, Ca^{2+}, Fe^{2+}, Cu^{2+}]\) entrapped nanocomposites (MMT-M\(^{2+}\)), which was potentially found optimum at pH 6.6 and temperature 298K. Adsorption parameters and percent binding data endorse that CYN was adsorbed appreciably on MMT-Fe\(^{2+}\) followed by MMT-Cu\(^{2+}\) as compared to MMT alone and with Ca\(^{2+}\) and Mg\(^{2+}\). As metal ions in the form of metalloenzymes are very good reflectors of biogenic activities in living systems, the results substantiate the efficacy of biogenic divalent metal ions particularly transition metal ions in prebiological adsorption processes through which concentration and condensation of biomonomers might have facilitated the formation of complex biopolymers. Moreover, during the formation of planet Earth iron was the most abundant element which might have contributed a lot in chemical evolution and origin of life [69, 70]. Our present results are also in good agreement with this scenario.

Iron is also the most abundant multivalent element in planetary reservoirs like Mars, which may record signatures of processes, occurred during the formation and subsequent differentiation of the terrestrial planets [80-83], focuses on extinct or extant life on Mars [84, 85]. Biological informational polymers such as nucleic acids have the potential to provide unambiguous evidence of life beyond Earth [86]. To summarize the most recent achievements in the field of astrobiology [87] reviews on experimental studies of space [88, 89] and on the Earth [90], as a tool for astrobiology, have also been compiled. One of the key goals of astrobiology is to explore planetary bodies of our solar system in order to determine their potential for habitability and extra-terrestrial life.

FTIR spectra emphasize that CYN interacted with Fe\(^{2+}\) encapsulated MMT through electrostatic forces via N-H and C=O bonds. XRD data insinuated that CYN interact with different sites of the clay surface, i.e., the interlayer region and the faces of the lamella showing expansion of the interlayer was greater due to the inclusion of CYN. Results demonstrate the higher affinity of MMT-Fe\(^{2+}\) interlayer for CYN. SEM observations showed the alignment of biomolecule on nanocrystalline solid and surface morphology accordingly. Henceforth, upon adsorption on mineral matrices CYN could be stabilized and protected from hazardous electromagnetic radiations directly coming from Sun in ozone free primitive environment and thus become available for successive molecular evolution.

In a wider perspective the interplay between minerals and organic molecules not only could have provided support necessary for prebiological chemical evolution, but also could modify structural and physico-chemical properties of the matrix itself, thus changing the micro environment where evolution occurs. Mineral matrices could in turn provide a new level of support in the evolutionary dynamics of prebiological systems [76]. The physico-chemical parameters of the study also deliberate several aspects of MMT nanoclay as drug carrier system with multifarious applications in biological assay.
The work provides insight into the prebiotic biogeochemical as well as biological processes such as biomineralization, biomedical implants, primigenial chemical evolution implicated to origin of first life [91, 76, 50]. The interstellar bodies also evidently endue protobiomolecules viz. amino acids, nucleic acid bases, peptides, nucleotides, lipids and extracellular polymeric substances, firmly allied with inorganic tempeltes [92-94]. Therefore, surface interaction studies of such precious molecules on clay minerals are interesting and motivating to explore the extraterrestrial genesis and plausibility of life beyond Earth.

**Supplementary Materials:** The following are available online at www.mdpi.com/link, Figure S1: Standard concentration curve (optical absorbance v/s molar concentration) of cytosine.

Figure S1. Standard concentration curve (optical absorbance v/s molar concentration) of cytosine.

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