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Synthesis, Spectroscopic Characterization, and Analytical Applications of Cinnamaldehyde Thiosemicarbazone and Its Fe(III) Complex in Environmental Water Analysis

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Abstract

In this study, a novel ligand, cinnamaldehyde thiosemicarbazone (Cin-TSC), was synthesized and thoroughly characterized for its coordination behaviour with Fe(III) ions. The ligand was prepared by refluxing equimolar quantities of cinnamaldehyde and thiosemicarbazide in ethanol, yielding a yellow crystalline solid with a melting point of 173 °C and a calculated molecular weight of 205.284 g/mol. Elemental analysis confirmed the compound's stoichiometry, while antimicrobial screening against *Klebsiella pneumoniae* indicated moderate bioactivity with a 0.60 cm inhibition zone.

The ligand was analyzed using UV-visible and infrared (IR) spectroscopy. A strong absorption peak at 330 nm was observed in the UV-Vis spectrum ($\varepsilon = 1.250 \times 10^5$ L·mol^{-1}·cm^{-1}), and IR spectral data confirmed the presence of key functional groups such as C=S, C=N, N–H, and aromatic moieties. X-ray diffraction (XRD) studies revealed that Cin-TSC crystallizes in the orthorhombic system with high structural order.

Complexation studies with Fe(III) were conducted at pH 5 (Optimum pH)using spectrophotometric techniques. The complex exhibited a maximum absorbance at 410 nm with a molar absorptivity of 6.847×10^2 L·mol^{-1}·cm^{-1}. Job's method and mole ratio analysis both confirmed a 1:2 metal-to-ligand stoichiometry. The dissociation constant of the complex was calculated to be 4.010×10^{-10}, corresponding to a high stability constant ($\beta = 2.493 \times 10^9$), indicating the strong binding affinity of the ligand toward Fe(III). Beer's law was obeyed in the concentration range of 8.953×10^{-5} to 8.953×10^{-4} M, and Sandell's sensitivity was determined to be 0.0818 µg cm^{-2}. Interference studies demonstrated that most divalent metal ions had negligible impact on complexation, whereas chelating agents such as EDTA and acetate ions significantly interfered. Furthermore, Cin-TSC was successfully applied to the spectrophotometric determination of Fe(III) in Panchganga River water samples. The experimentally observed Fe(III) concentration (0.83 ppm) closely matched the calculated value (0.82 ppm), confirming the reliability and accuracy of the method. These findings suggest that Cin-TSC is a promising ligand for the spectrophotometric determination of iron and offers potential for biological, analytical, and environmental water analysis applications.

Keywords
Cinnamaldehyde thiosemicarbazone, Iron(III) complex, Spectrophotometric determination, Stability constant, UV-Visible spectroscopy, Infrared spectroscopy, Antimicrobial activity.

1. Introduction

Iron is fundamental across science, health, and industry—in metallurgy, physiology, and analytical chemistry—making its precise quantification in food, biological fluids, metals, and alloys essential (1). Its coordination with thiosemicarbazones has gained notable attention for both medicinal and analytical applications (2). Thiosemicarbazone–metal complexes exhibit diverse pharmacological activities including antitumor, antitubercular, antibacterial, antifungal, and anti-inflammatory effects (3), often operating via mitochondrial pathways and reactive oxygen species (ROS) generation, and some are even entering clinical phases (4). Therapeutically, iron–thiosemicarbazone complexes are being explored as photodynamic and theranostic agents, capable of red-light–induced apoptosis and MRI contrast enhancement in cancer cells (5).

In analytical chemistry, their strong chelation and chromophoric ability underpin sensitive spectrophotometric and extractive methods for trace iron detection (6). In the present study, we investigate the iron(III) complex of cinnamaldehyde-thiosemicarbazone (Cin-TSC). Literature reveals that Ghosh showed Fe(II) auto-oxidation dynamics compared to hemoglobin using spin-relaxation measurements, with notable behaviour in EDTA systems (7); Aswar and Bhave detailed structural and thermal characteristics of Fe(II) polychelates (8); Sawalkar et al. confirmed Fe(III) chelates exhibit greater stability than their Cu(II) and Ni(II) analogues (9); Bandyopadhyay and

Banerjee investigated Fe(III) complexation kinetics with hydroxamic acids (10); Ghosh evaluated pH-dependent Fe(III) complexation with BSA, noting hydration changes (11); Prakash synthesized mixed-ligand Fe(II) complexes with analytical and pharmacological potential (12); Balasubramaniam studied thermal behaviour in polypyridine–Fe(II) systems (13); Pandey reported stability constants of ternary Fe(II) complexes (14); Singh described polymeric Fe(II) complexes with quadridentate ligands (15); Swamy introduced novel N,O-donor ligand Fe(II) complexes; and Sarma developed a highly sensitive spectrophotometric Fe(III) assay using a pyridoxal-phenyl-thiosemicarbazone ligand.

Building on this foundation, our work aims to deepen understanding of Cin-TSC's complexation with Fe(III), its stability constants, and its potential pharmaceutical and analytical applications.

2. Experimental

2.1. Synthesis and Characterization of Cinnamaldehyde Thiosemicarbazone (Cin-TSC)

2.1.1 Synthesis of Cin-TSC

Cinnamaldehyde thiosemicarbazone was synthesized by heating equimolar quantities of cinnamaldehyde (C_6H_5–CH=CH–CHO; molar mass: 132.07 g/mol) and thiosemicarbazide in a minimal quantity of ethanol under reflux conditions for approximately one hour. A yellow solid separated upon cooling, which was filtered, washed with cold ethanol, and recrystallized from hot ethanol to yield pure crystalline material. The compound exhibited a melting point of 173 °C, and its molecular weight was calculated as 205.284 g/mol based on its proposed structure.[16]

2.1.2 Elemental Analysis

The elemental composition of the synthesized ligand was determined to verify its purity and validate its molecular formula. The experimental values were found to be in good agreement with theoretical calculations, as shown in Table 1, confirming the successful formation of Cinnamaldehyde-TSC.

2.1.3. Antimicrobial Activity of the Ligand

To evaluate biological activity, the ligand was tested against *Klebsiella pneumoniae*. A 100 mg/mL ethanolic solution of Cin-TSC was prepared and sterile paper discs were immersed in this solution. The solvent was allowed to evaporate before placing the discs onto nutrient agar plates inoculated with a 24-hour-old bacterial culture. After incubation at 37 °C for 24 hours, zones of inhibition were measured. A clear zone of 0.60 cm was recorded, indicating moderate antimicrobial activity.[17]

2.1.4. UV-Visible Absorption Spectral Study

The UV-Visible absorption spectrum of Cin-TSC was recorded using a spectrophotometer in ethanol medium at pH 5, adjusted with acetate buffer. A prominent absorption band was observed at 330 nm for a 2.439×10^{-5} M solution, with a molar extinction coefficient (ε) of 1.250×10^5 $L \cdot mol^{-1} \cdot cm^{-1}$. This indicates that the ligand exhibits strong absorption in the UV region, just at the edge of the visible spectrum.[18]

2.1.5. Infrared Spectral Analysis

The infrared (IR) spectrum of Cinnamaldehyde Thiosemicarbazone (Cin-TSC) was recorded in the range of 4000–200 cm^{-1} using the KBr pellet method on a Perkin Elmer 221 spectrophotometer. The spectrum exhibited a number of prominent absorption bands that support the structural framework of the ligand.[19] A strong absorption band at 950 cm^{-1} is assigned to C=S stretching, while another at 2580 cm^{-1} corresponds to N–C=S stretching, confirming the thiosemicarbazone moiety. The peak at 1500 cm^{-1} is attributed to C=S with NH coupling. The bands at 3000 cm^{-1} and 3330 cm^{-1} correspond to aromatic C–H and hydrogen-bonded C–H stretching, respectively. A broad absorption band around 3520 cm^{-1} indicates $-NH_2$ stretching.

Additional bands at 1560 cm^{-1} and 1510 cm^{-1} are characteristic of C=N (azomethine) and aromatic C=C stretching vibrations. The absorption around 1340 cm^{-1} may be due to overlapping C–N and C=S stretching, while the band near 660 cm^{-1} is tentatively assigned to C–S deformation.

A low-frequency band was also observed, confirming metal–ligand coordination via M–N or M–S bonding. The IR spectral features thus substantiate the proposed structure of the synthesized ligand. The detailed IR frequencies together with their corresponding functional groups are summarized in Table 2

2.1.6. X-ray Diffraction (XRD) Analysis

The XRD pattern of the ligand was recorded and analyzed following the Hesse–Lipson method to determine its crystalline structure. A good match was observed between the calculated $\sin^2\theta$ values and those obtained experimentally for peaks A, B, and C. Interplanar distances (d-spacings) were calculated using Bragg's Law, and excellent agreement was found with observed values. These results suggest that the ligand crystallizes in the orthorhombic system, characterized by distinct axial lengths ($a \neq b \neq c$) and right angles between all axes ($\alpha = \beta = \gamma = 90°$).[20] The crystallinity and symmetry observed in the diffraction pattern confirm the ordered structure of the synthesized thiosemicarbazone ligand.

2.2. Spectrophotometric study of Fe(III)- Cinnamaldehyde-Thiosemicarbazone

2.2.1 Preparation of Standard Fe(III) Solution

A standard stock solution of Fe(III) was prepared by accurately dissolving 0.6050 g of ferric chloride hexahydrate ($FeCl_3 \cdot 6H_2O$) in distilled water and diluting the volume to 250 mL in a volumetric flask. The molar concentration of the resulting solution was 8.953×10^{-3} M. This stock was further diluted as required for subsequent experiments.

2.2.2 Preparation of Standard Cinnamaldehyde-Thiosemicarbazone Solution

A standard solution of Cinnamaldehyde-thiosemicarbazone was prepared by dissolving 0.459 g of the synthesized compound in alcohol and making up the volume to 250 mL. The resulting concentration was also 8.953×10^{-3} M, identical to the Fe(III) solution to maintain stoichiometric conditions during complexation studies.

2.2.3 Preparation of Buffer Solution (pH 5)

An acetate buffer of pH 5 was prepared by mixing 14.75 mL of 0.2 N acetic acid (CH_3COOH) with 35.25 mL of sodium acetate (CH_3COONa) solution. All reagents and solvents used were of analytical reagent (AR) grade to ensure high purity and reproducibility of results.

2.2.4 Recommended Procedure for Complex Formation

To a clean test tube or cuvette, 1.0 mL of the standard iron(III) solution was added, followed by 1.0 mL of the Cin-TSC solution. The pH of the mixture was adjusted to 5.0 by adding 1.0 mL of the acetate buffer. The resulting solution was diluted to a final volume of 10.0 mL using absolute alcohol. This solution was then subjected to further analytical measurements, including spectrophotometric analysis.[21]

2.2.5 Other Experimental Details

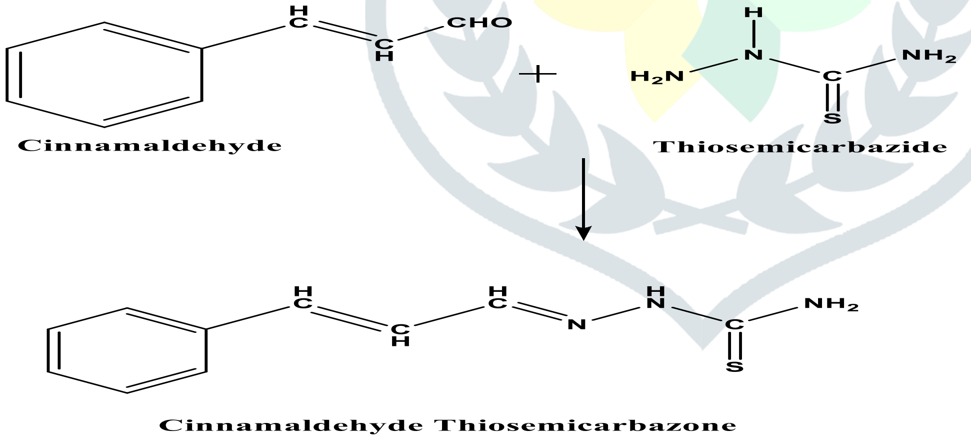
Additional parameters for the Fe(III)–Cin–TSC system were investigated, including the effects of reaction time, pH, and reagent concentration on absorbance, as well as the stability of the complex. Studies were also conducted to determine the validity of Beer's law, the dissociation constant, the stability constant, and the influence of diverse ions on the Fe(III)–Cin–TSC system.

3. Results and Discussion:

3.0 Characterization of Cinnamaldehyde Thiosemicarbazone (Cin-TSC)

3.1.0 Synthesis and Physical Characteristics

Cin-TSC was synthesized by refluxing equimolar quantities of cinnamaldehyde and thiosemicarbazide in ethanol. The reaction yielded a yellow crystalline solid with a melting point of 173 °C, indicating good purity and well-defined crystalline nature. The compound was stable upon recrystallization from ethanol and had a calculated molecular weight of 205.284 g/mol, consistent with its proposed molecular formula.



3.1.1 Elemental Analysis

The purity and stoichiometry of the ligand were confirmed by elemental analysis. The experimentally observed values for carbon (58.22%), hydrogen (5.10%), nitrogen (20.22%), and sulphur (15.30%) were found to be in close agreement with the calculated theoretical values. These findings support the successful synthesis and proper stoichiometric incorporation of each element into the ligand framework. The relatively lower sulphur content observed may be attributed to experimental deviations, but the overall data validate the ligand's composition. Elemental Analysis of Cinnamaldehyde-TSC is shown in Table No.1

Table 1: Elemental Analysis of Cinnamaldehyde-TSC

Sr. No.	Chemical Element	Percentage Found (%)	Expected (%)
1	Carbon	58.22	58.53
2	Hydrogen	5.10	5.36
3	Sulphur	15.30	15.60
4	Nitrogen	20.22	20.48

3.1.2 Antimicrobial Activity

The Cin-TSC ligand was evaluated for antimicrobial activity against *Klebsiella pneumoniae* using the disc diffusion method. A zone of inhibition measuring 0.60 cm was observed after 24 hours of incubation at 37 °C, indicating moderate antimicrobial potential. The result highlights the bioactive nature of the thiosemicarbazone functional group, known for its pharmacological relevance, particularly in metal coordination complexes.

3.1.3 UV–Visible Spectroscopy

The UV–Vis absorption spectrum of the ligand, recorded in ethanol at pH 5, displayed a strong absorption band at 330 nm. The molar extinction coefficient (ε) was calculated as 1.250×10^5 $L \cdot mol^{-1} \cdot cm^{-1}$, signifying intense $\pi \rightarrow \pi^*$

or n→π* transitions typically observed in conjugated systems involving azomethine and C=S functionalities. The absorbance profile supports the conjugated electronic structure of the ligand, and this wavelength served as a reference for future metal complexation studies.

3.1.4 Infrared Spectral Analysis

Infrared spectroscopy provided further insight into the functional groups present in Cin-TSC. The IR spectrum displayed characteristic bands confirming the presence of the thiosemicarbazone moiety. A sharp band at 950 cm^{-1} was attributed to C=S stretching, while 2580 cm^{-1} corresponded to N–C=S stretching vibrations. Additionally, 1500 cm^{-1} was assigned to C=S coupled with NH bending. The IR spectrum of Cin-TSC is presented in Fig. 1 (Table 2)

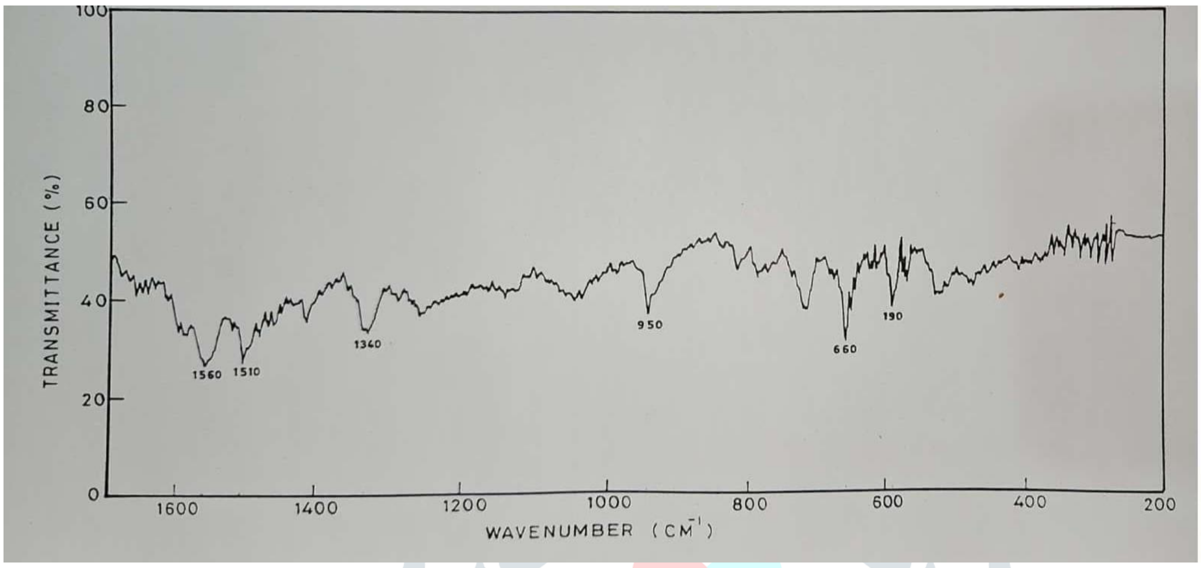


Fig.1 IR Spectra of Cinnamaldehyde -Thiosemicarbazone.

Table 2 IR Spectra and Expected Functional Group

Sr. No.	Frequency (cm^{-1})	Expected Functional Group
1	655	C–S stretching or deformation
2	715	Out-of-plane =C–H bending (aromatic ring)
3	950	C=S stretching (thiocarbonyl group)
4	1335	C–N stretching or CH_2 bending
5	1410	C–N stretching or CH_2 bending
6	1500	C=S coupled with N–H bending
7	1555	C=N stretching (azomethine group)
8	2580	N–C=S stretching
9	3000	C–H stretching of mono-substituted aromatic ring
10	3200	N–H stretching (overlapped with hydrogen bonding)
11	3330	Aromatic C–H stretching; intermolecular hydrogen bonding (possibly O–H/N–H)
12	3520	N–H symmetric and asymmetric stretching of –NH_2 (primary amine group)

Other significant absorptions included:

- 1560 cm^{-1} and 1510 cm^{-1}, representing azomethine (C=N) and aromatic C=C stretching.
- 3000 cm^{-1} and 3330 cm^{-1}, corresponding to aromatic and hydrogen-bonded C–H stretching.
- A broad peak at 3520 cm^{-1} due to –NH_2 group stretching, indicating the presence of primary amines.
- The band at 1340 cm^{-1} was likely due to overlapping C–N and C=S stretching.
- A lower band near 660 cm^{-1} suggested C–S deformation.

These features collectively support the formation of the desired ligand with the expected functionalities required for metal coordination, indicating successful metal–ligand interaction through nitrogen or sulphur atoms.

3.1.5 X-ray Diffraction (XRD) Analysis

The crystalline structure of Cin-TSC was analyzed by XRD, and the pattern matched well with calculated values based on the Hesse–Lipson method. The $\sin^2\theta$ values and d-spacings corresponded to an orthorhombic crystal system, defined by unequal axis lengths ($a \neq b \neq c$) and equal interaxial angles ($\alpha = \beta = \gamma = 90°$). The sharp and well-defined diffraction peaks confirmed a high degree of crystallinity and structural order. These results provide substantial support for the ligand's purity and its potential for forming coordination complexes with metal ions. XRD pattern of Cin-TSC is presented in Fig. 2.

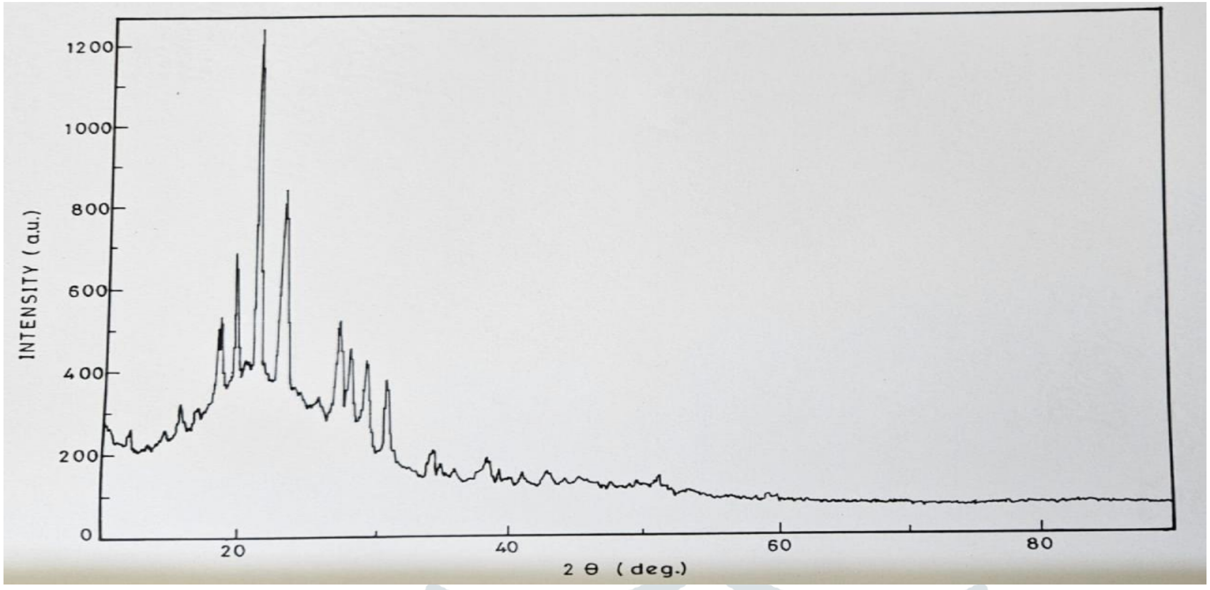


Fig. 2 X-RD of Cinnamaldehyde- Thiosemicarbazone.

3.2. Study of Fe(III)–Cinnamaldehyde-Thiosemicarbazone Complex

3.2.1 Absorption Spectra and Molar Extinction Coefficient of Fe(III)–Cinnamaldehyde-Thiosemicarbazone Complex

The absorption spectrum of the Fe(III)–Cinnamaldehyde–Thiosemicarbazone (Cin-TSC) complex was recorded using a solution containing 1 mL of Fe(III) (8.953×10^{-3} M) and 1 mL of Cin-TSC (8.953×10^{-3} M), adjusted to pH 5 and diluted to 10 mL with absolute alcohol. Spectral data indicated a strong absorption peak at 410 nm, which was selected as the optimum analytical wavelength for further studies. The molar extinction coefficient (ε) of the complex at this wavelength was calculated to be 6.847×10^{2} L mol^{-1} cm^{-1}.

3.2.2 Effect of Time on Absorbance

The time stability of the Fe(III)–Cin-TSC complex was examined at pH 5. Upon mixing the metal and ligand, the complex formed immediately with an intense color, indicating rapid complexation. The absorbance remained essentially unchanged over 24 hours, demonstrating that the complex is stable for at least one day under experimental conditions.

3.2.3 Effect of pH

To determine the influence of pH on complex formation, a series of solutions containing equal concentrations of Fe(III) and Cin-TSC were prepared over the pH range of 1 to 9, using acetate buffer where necessary. Absorbance values were recorded at 410 nm against the reagent blank. Results indicated that the complex formation is highly pH-dependent, with maximum absorbance at pH 5, confirming this as the optimum pH for the reaction

3.2.4 Effect of Reagent Concentration

The effect of ligand concentration was assessed by keeping the Fe(III) concentration constant (8.953×10^{-4} M) and varying the volume of Cin-TSC reagent. All solutions were maintained at pH 5 and diluted to 10 mL with alcohol. Absorbance readings at 410 nm revealed that the absorbance increased with ligand volume until reaching a plateau. A reagent volume of 2.2 mL was found sufficient for complete complexation

3.2.5 Stability of the Complex

The Fe(III)–Cin-TSC complex exhibited excellent stability under the optimized conditions. Absorbance values remained consistent for up to 24 hours, confirming the short-term stability of the complex, which is beneficial for analytical applications.

3.2.6 Validity of Beer's Law

Beer's Law was verified by preparing a series of solutions with varying Fe(III) concentrations ranging from 8.953×10^{-5} to 8.953×10^{-4} M, while keeping the ligand concentration constant at 2.5 mL. The pH was maintained at 5, and all solutions were diluted to 10 mL with alcohol. A linear relationship between absorbance and concentration was observed at 410 nm, confirming that the complex obeys Beer's Law over the studied range

3.2.7 Sandell's Sensitivity

Based on the molar absorptivity and analytical procedure, the mean molar extinction coefficient at 410 nm was found to be 6.829×10^{2} L mol^{-1} cm^{-1}. Using the standard method Sandell's sensitivity for Fe(III) was calculated as 0.0818 µg cm^{-2}, indicating the method's suitability for trace-level determination of iron.

3.2.8 Composition of the Complex

The stoichiometry of the Fe(III)–Cinnamaldehyde–Thiosemicarbazone (Cin-TSC) complex was established using two classical spectrophotometric techniques: Job's method of continuous variation and the mole ratio method. Both methods confirm the formation of a 1:2 metal-to-ligand complex.

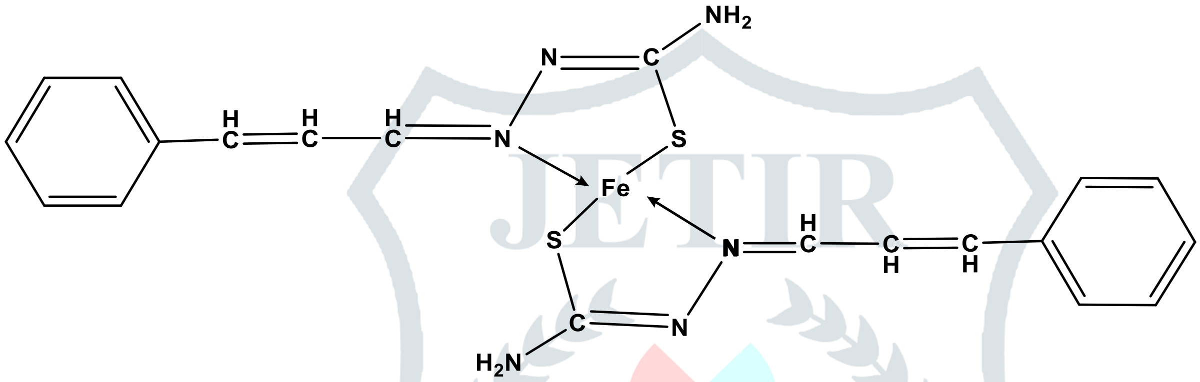
3.2.8.1 Job's Method of Continuous Variation

To determine the combining ratio, a series of equimolar solutions (8.953×10^{-3} M) of Fe(III) and Cinn-TSC were mixed in varying mole fractions while maintaining the total volume and total molarity constant. The pH of each mixture was adjusted to 5.0, and the final volume was made up to 10 mL using absolute alcohol. The absorbance of each solution was measured at 410 nm against a reagent blank. A plot of absorbance versus mole fraction exhibited a maximum at a mole fraction of 0.33, indicating a 1:2 metal-to-ligand stoichiometry in the complex.

3.2.8.2 Mole Ratio Method

In this method, the concentration of Fe(III) was kept constant while the volume of the Cin-TSC reagent was varied. All solutions were prepared at pH 5, diluted to 10 mL with alcohol, and their absorbance measured at 410 nm. The resulting plot of absorbance versus ligand-to-metal mole ratio showed a distinct inflection point at a molar ratio of 1:2 (Fe:Cin-TSC). This supports the result obtained from Job's method, confirming the stoichiometry of the complex as 1:2.

Probable Structure of the Fe(III)–Cinnamaldehyde–TSC Complex



Based on the observed 1:2 stoichiometry and known coordination behaviour of thiosemicarbazones, the probable structure of the Fe(III) complex involves coordination of two Cin-TSC ligands to a central Fe(III) ion. Each ligand likely binds through the azomethine nitrogen (–C=N–) and thione sulfur (–C=S) donor atoms, forming stable five membered chelate rings. The resulting geometry around Fe(III) is expected to be octahedral, commonly observed for such trivalent metal complexes.

3.2.9: Dissociation Constant

Degree of Dissociation (α):

The degree of dissociation (α) was determined using the relation:

$\alpha = (dE - d_0) / dE$

Where:

- *dE* = Optical density when the complex is completely formed = 0.61
- *d₀* = Optical density at stoichiometric molar ratio = 0.58

Substituting the values:

$\alpha = (0.61 - 0.58) / 0.61 = 0.03 / 0.61 = 0.0492$

Dissociation Constant (K):

The dissociation constant (instability constant) of the Fe(III)–Cinnamaldehyde-TSC complex was calculated using the mole ratio method and the following formula:

$K = [(m \times \alpha \times c)^m \times (n \times \alpha \times c)^n] / [(1 - \alpha) \times c]$

Where:

- *m* = Number of moles of Fe(III) = 1
- *n* = Number of moles of reagent = 2
- *α* = 0.0492
- *c* = Final concentration after dilution = 8.953×10^{-4} M

Substituting the values:

$K = [(1 \times 0.0492 \times 8.953 \times 10^{-4})] \times [(2 \times 0.0492 \times 8.953 \times 10^{-4})^2] / [(1 - 0.0492) \times 8.953 \times 10^{-4}]$

$K = 3.414 \times 10^{-13} / 8.512 \times 10^{-4} = 4.010 \times 10^{-10}$

3.2.10 Stability Constant (β):

The stability constant (β) is the reciprocal of the dissociation constant:
$\beta = 1/K = 1/(4.010\times10^{-10}) = 2.493\times10^{9}$

The values of the dissociation constant and stability constant indicate the formation of a highly stable metal–ligand complex at pH 5

3.2.11 – Diverse Ion Effect

The objective of this study was to evaluate the influence of various foreign ions on the complexation of Fe(III) with Cinnamaldehyde-Thiosemicarbazone under controlled experimental conditions at pH 5. The solutions used for this experiment contained equimolar concentrations of Fe(III) and Cinnamaldehyde-TSC, each at 8.953×10^{-3} M. The extent of interference was monitored based on changes in complexation behaviour, and a tolerance limit of less than 2% deviation in absorbance was considered acceptable for minimal interference.

The experimental results revealed that several ions such as Cd^{2+}, Sn^{2+}, Mg^{2+}, Co^{2+}, Ni^{2+}, and Hg^{2+} did not cause significant interference in the formation of the Fe(III)–Cinnamaldehyde-TSC complex. These ions were found to be tolerable within the defined limits, indicating their minimal impact on the complexation process. However, the presence of strongly coordinating or chelating agents like EDTA and acetate ions led to serious interference. These substances significantly disrupted complex formation, likely due to their ability to preferentially bind with Fe(III), thereby inhibiting its interaction with the ligand.

In conclusion, the complexation of Fe(III) with Cinnamaldehyde-TSC is largely unaffected by several divalent metal ions, demonstrating good selectivity. However, complexing agents such as EDTA and acetate ions must be strictly avoided during analytical procedures, as they can hinder accurate complex formation and compromise the reliability of the results.

3.2.12 Analytical Applications of Cinnamaldehyde Thiosemicarbazone

Cinnamaldehyde thiosemicarbazone was effectively employed for the spectrophotometric determination of iron(III) in water samples collected from the Panchganga River. A calibration curve was constructed using standard Fe(III) solutions of known concentrations, following Beer–Lambert's law. The absorbance of the river water samples was measured at the wavelength of maximum absorption (λ_{max} 410 nm), and the corresponding iron concentrations were determined in parts per million using the standard curve.

The experimentally observed value for Fe(III) in the sample was 0.83 ppm, which closely matched the calculated value of 0.82 ppm, confirming the reliability and accuracy of the method. These findings demonstrate that cinnamaldehyde thiosemicarbazone is an efficient and selective analytical reagent for the spectrophotometric estimation of Fe(III) in environmental water samples.

4.0 Conclusion

In this study, cinnamaldehyde thiosemicarbazone was successfully synthesized and structurally characterized through elemental analysis, UV–Visible spectroscopy, FTIR, and X-ray diffraction. The spectral and diffraction data confirmed the presence of functional groups essential for metal coordination and revealed that Cin-TSC crystallizes in the orthorhombic system, indicating a well-ordered crystalline structure.

The complexation behaviour of Cin-TSC with Fe(III) ions was thoroughly investigated. The Fe(III)–Cin-TSC complex exhibited strong absorbance at 410 nm and obeyed Beer–Lambert's law over the concentration range of 8.953×10^{-5} to 8.953×10^{-4} M. Stoichiometric analysis confirmed a 1:2 (metal: ligand) ratio, and the high stability constant ($\beta = 2.493 \times 10^{9}$) along with a low dissociation constant ($K = 4.010 \times 10^{-10}$) indicate strong metal–ligand binding affinity.

The ligand also showed promising analytical utility. It was applied to the spectrophotometric determination of Fe(III) in Panchganga River water samples. The observed Fe(III) concentration (0.83 ppm) closely matched the calculated value (0.82 ppm), validating the accuracy and sensitivity of the method. The Sandell's sensitivity (0.0818 $\mu g\ cm^{-2}$) and minimal interference from common divalent ions further demonstrate its potential as an effective analytical reagent, though interference from EDTA and acetate was noted.

Additionally, the ligand exhibited moderate antimicrobial activity against *Klebsiella pneumoniae*, suggesting its potential biological relevance.

Overall, Cin-TSC emerges as a highly stable, selective, and sensitive ligand for Fe(III) detection, with dual applications in environmental monitoring and bioinorganic chemistry.

Limitations and Future Scope

The present study demonstrates the efficiency of cinnamaldehyde thiosemicarbazone as a selective ligand for Fe(III) determination and its potential antimicrobial activity. While the work establishes a strong foundation, it is currently limited to one test organism (*Klebsiella pneumoniae*) and a single water source (Panchganga River). These focused choices highlight the reliability of the method in controlled conditions but also open opportunities for future exploration. Expanding antimicrobial studies to a wider spectrum of bacterial and fungal strains, as well as validating the analytical method in diverse environmental matrices such as industrial effluents and groundwater, would further enhance the applicability of the ligand. In addition, exploring its coordination with other transition metals and investigating its pharmacological properties, including cytotoxic and antitumor potential, could significantly broaden the scientific and practical impact of this research.

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