Microwave Synthesis, Characterization, Biological Activity of N-(p-Chlorophenyl)-N'-Benzoyl Thiourea and its Complexes

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Abstract

A derivative of thiourea ligand N-(p-chlorophenyl)-N'-Benzoyl thiourea (PCBT) in equimolar ratio 1:1 and its transition metal complexes CoII, NiII , CuII and ZnII were synthesized by microwave (green chemistry). The structure of ligand and its complexes have been characterized by using elemental analysis, mass Spectroscopy, FT-IR, UV-Vis., 1HNMR and 13CNMR. The geometry of the proposed structures of the chelates based on their electronic spectra, electron spin resonance (ESR) and magnetic susceptibility. The stability of complexes was studied by TGA analysis (Thermal studies). The free derivative thiourea ligand (PCBT) and its complexes were studied for antimicrobial and antifungal activity.

Results and Discussion

The structure of the prepared ligand N-(p-chlorophenyl)-N'-Benzoyl thiourea (PCBT) was characterized by melting point, elemental analysis, IR, mass spectra, UV-Vis and NMR studies. This data is compatible with the required product. Analytical and physical properties of prepared compounds tabulated in Table 1.

Compd.	Molecular formula	M.Wt.	Yeild%	M.P. °C	Color	%C	%H	%N	%M
PCBT	C14H11ON2SCl	290.8	86.3	140	Pale yellow	57.83 (57.57)	3.81 (3.96)	9.63 (10.06)	
Co-PCBT	CoC18H25O9N2SCl	539.58	79.8	320	Dark (pink)	40.03 (41.47)	3.79 (4.6)	5.16 (5.2)	
Ni-PCBT	NiC18H25O9N2SCl	539.21	81.3	310	Dark Green	40.06 (43.08)	3.68 (4.6)	5.2 (5.87)	10.88 (11.28)
Cu -PCBT	CuC18H19O6N2SCl	490.1	82.42	330	Greenish	44 (43.21)	3.72 (3.8)	5.72 (5.7)	
Zn-PCBT	ZnC18H21O7N2SCl	510	82.27	305	White	42.35 (43.7)	3.27 (4.01)	5.58 (5.49)	12.81 (14.31)

Table 1. Analytical and physical properties of the prepared compounds (CPds).

IR spectra

The characteristic IR bands of all thiourea ligands showed the expected frequencies of υ (C=O), υ (N-H), υ (C-N) and υ (C=S). The coordinative behavior of present ligand, towards Zn (II) and Ni (II) ions to form complexes is difficult to establish, as this ligand is capable of exhibiting three tautomeric forms as clear from Scheme 2 due to presence of [â NHâ C (=S)] and [â NHâ C (=O)] functional groups.12

Chemical-Sciences-prepared-ligand

Scheme 2. Tautomeric forms of the prepared ligand.

However, the lack of the characteristic vibrations of υ (S–H) around 2500â 2600 cm-1 [10] and presence of a peak at 3251 cm-1 characteristic of υ (N–H) [11] confirmed the absence of tautomeric form (N=Câ SH) and (N=C-OH). Two sharp intense bands observed at 1670 and 1341 cm-1 can be ascribed to the stretching vibration of carbonyl group υ (C=O) and thionyl group υ (C=S) respectively [12,13]. These observations confirmed the ketonicâ thion form of the ligand in the solid-state. Moreover, the υ (C–Cl) stretching frequency was observed at 765

cm-1 , while this band appearing at 678-686 cm-1 assigned to the usual modes of phenyl ring vibration, respectively [14,15]. On the other hand and upon coordination of the metal center to ligand, the characteristic bands of υ (C=O) and υ (C=S) present in the spectrum of the free ligand at 1670 and 1341 cm-1 were found to be shifted to a lower frequency and appear from 1535 to 1605 for C=O and from 1273 to 1250 cm-1 for C=S. This finding may be taken as an evidence for the coordination of the carbonyl oxygen and thionyl Sulphur atoms with the metal ions. **Introduction**

Coordination compounds attracted great attention due to their structural variety, interesting physical & chemical properties and promising applications in many fields. Metal ions play an important role in the structure and function of many bio macromolecules and have important roles in the biological processes of metabolism as well as in pharmaceutical chemistry due to their chemical properties. Compounds bearing carbonyl and thio carbonyl groups are used as potential donor ligand for the preparation of complexes [1,2]. Among these, thiourea and its derivatives are versatile ligands that coordinate to form stable compounds. Thiourea (NH2)2C=S is a compound where the oxygen atom of urea compound replaced by a Sulphur atom. They are able to coordinate with metal either as neutral or mono-anion or dianion ligand [3,4]. Chemotherapy concerned in treatment of disease in which a chemical is specifically targeted for a microbial agent or a specific tissue. These thiourea ligands and their metal complexes were reported to act as antimicrobial, antibacterial, antifungal, antimalarial, anti-tuberculous and anticancer activities [7]. They also form a variety of complexes of different symmetries with various metal ions [7,8]. In view of the importance of thiourea and their derivatives it was worth interesting to synthesize Nsubstituted thiourea ligand and their complexes with transition metal elements because it was observed that this activity was enhanced by complexing with certain transition metal elements [3,9,10]. Complexes were synthesized using microwave-assisted irradiation. Microwave gives shorter reaction times, clean, high yields, and low cost [11].

Materials and Experimental Method

All purchased chemicals were of Annular AR grade and were obtained from Sigma Aldrich and all Metals salts were purchased from ADWIC. The Microwave-assisted synthesis was carried out in a domestic microwave energy output 900 W. Purity of Schiff base ligand and its complexes were detected by using thin-layer chromatography (TLC) technique. Melting points were recorded in open capillaries with Barnstead Thermolyne Mel-temp 1001D Electrothermal Melting Point. Elemental analysis was done on automatic analyzer CHNS Vario El III-Elementar, Germany. The FT-IR spectra samples were ground with (CsBr) powder. Then pressed into a disk and recorded on Shimadzu FTIR spectrometer. Mass spectra were determined by using Mass GC-2010 Shimdazu instrument. Metals content were determined by complexometric titration using xylenol orange (XO) as indicator and hexamine as a buffer (pH = 6). Electronic absorption spectra in DMF were measured using automated UV/Vis-NIR 3101 PC Shimadzu spectrophotometer ranged from 200-900 nm. 1HNMR spectra for Schiff base ligand was recorded in 300MHz Varian-Oxford Mercury in DMSO-d6 as solvent and the chemical shifts were recorded in ppm relative to TMS as an internal standard. Magnetic susceptibility of complexes was measured on powdered samples using the faraday method. Thermal analysis measurements (TGA) were carried out with Shimadzu thermal analyzer model 50 at Microanalytical. The ESR spectra of the powdered CuII complex recorded at room temperature by X-band EMX spectrometer (Bruker, Germany) using a standard rectangular cavity of ER 4102 with 100 KHz frequency.

Schiff base ligand and their metal complexes were screened for in-vitro antibacterial activity against two species of Gram-positive bacteria and two species of Gram-negative bacteria as well as two

species of fungi. All of these were carried out in faculty of Science, Cairo University.

Chemicals

All consumed chemicals were from analytical grade and were used as received without further purifications. Chemicals used are Benzoyl chloride, ammonium thiocyanate, p-chloro analin, acetone, cobalt acetate, nickel acetate, copper acetate, zinc acetate and methanol.

Synthesis of N-(p-chlorophenyl)-N'-Benzoyl thiourea (PCBT)

0.1 m of ammonium thiocyanate (7.6 gm) dissolved in 50 ml of acetone then added drop by drop to 0.1 m of benzoyl chloride (14.06 gm) (11.62 ml) taken in 3 neck flask with continuous stirring. The mixture is refluxed for 1 hour with continuous stirring. After 45 minutes white ppt (ammonium chloride) appeared and then disappeared at 1 hr. The mixture left in room temp until the precipitate appears again completely. Filtration done and precipitate washed by acetone to get all the filtrate (Benzoyl thiocyanate). The filtrate added drop by drop in a 3-neck flask contains 0.1 m (12.75 gm) of para chloro analin dissolved in 25 ml acetone with continuous stirring. The mixture refluxed for 2 hrs with continuous stirring. The mixture transferred to a baker and covered for two days for complete precipitation. Then the Precipitate was filtrated and washed by ethanol and acetone. Purity of ligand achieved by recrystallization using ethanol and dried over anhydrous CaCl2 in a desiccator to give the pure ligand (PCBT).

Synthesis of metal complexes

The prepared ligand and the acetate salts of the metal Co (CH3COO)2.4H2O, Ni (CH3COO)2.4H2O, Cu (CH3COO)2.H2O and Zn (CH3COO)2.2H2O were mixed in (1:1) ratio. The reaction mixtures were then irradiated by the microwave oven by using drops of methanol as a solvent. The reaction was completed in a short time (3-5 min) with higher yields. The resulting product washed by hot methanol and ether and finally dried under reduced pressure over anhydrous CaCl2 in a desiccator. The progress of the reaction and purity of the product was monitored by TLC using silica gel (yield: 78-83 %). The synthetic route of the prepared compounds is illustrated in **Scheme 1.**

Scheme 1. Synthesis of N-(p-chlorophenyl)-N'-Benzoyl thiourea (PCBT) and its metal Complexes.

Biological Activity

Antibacterial and anti-fungal effect

Some chelates exhibited a moderate inhibitory activity of complexes than that of the corresponding free ligands.

The free ligand (PCBT) and its metal complex CoII in addition to the standard drugs were screened separately for their antibacterial activity against Staphylococcus aureus (ATCC:6538), streptococcus mutans (ATCC: 25175) (Gram-positive bacteria), Escherichia Coli (ATCC:9637) and Klebsiella Pneumonia (ATCC:10031) (Gram-negative bacteria) and antifungal activity against Aspergillus Nigar (ATCC:32856) and Candida albicans (ATCC:6538)fungi.

The antimicrobial activity against the growth of various microorganisms were determined by measuring the inhibition zone in millimeters around the well, also the activity index data was calculated [22].

The result is recorded in **Table 5**. As we can observe from the results metal complexes act as more powerful bactericides and fungicides agents and they may serve as a vehicle for activation of ligand where the metal ions being more hypersensitive against the microbial cells.

Table 5. Antibacterial and antifungal assay of ligand and its ZnII complex.

This behaviour of the metal complexes may be a result to the modification in structure

upon coordination and formation of metal organic framework and can be explained on the basis of the overtone concept and chelation theory.

In general, the easy penetration of the metal complexes into lipid membranes, disturbance of the respiration process of the cell and blocking the synthesis of proteins are restrict further growth of the organism and lead to enhance of activity of metal complexes compared with the organic ligand [23-31]. We did different fungi and ligand interactions with proper docking of the compound.

Conclusion

Complexes were synthesized successfully and fully characterized by chemical and spectroscopic methods. Then biological activity was studied and compared between the ligand itself and the ZnII complex. The study showed that the prepared compounds have an appreciable activity and can consider as an effective inhibitor towards the different microbial strains. Generally such activity enhanced upon complexation where metal complexes show better activity than their parent ligand.

References

1. Hakan Arslan, Nevzat Külcü and Ulrich Flörke "Synthesis and characterization of copper (II), nickel (II) and cobalt (II) complexes with novel thiourea derivatives". Journal Transition Metal Chemistry 28 (2003): 816-9.
2. Demet Sezgin Mansuroglu, Hakan Arslan, Ulrich Flörke and Nevzat Külcü "Synthesis and characterization of nickel and copper complexes with 2, 2-diphenyl-N- (alkyl (aryl) carbamothioyl) acetamide: The crystal structures of HL1 and cis-[Ni (L1) 2]." Journal of Coordination Chemistry 61 (2008): 3134-46.
3. Mohd. Shadab and Mohammad Aslam. "Synthesis and characterization of some transition metal complexes with n-phenyl-n'-[substituted phenyl] thiourea." Material Science Research India 11 (2014): 83-9.
4. Hakan Arslan, Ulrich Flörke, Nevzat Külcü and Fatih Emen. "Crystal structure and thermal behaviour of copper (II) and zinc (II) complexes with N-pyrrolidine-N'-(2-chloro-benzoyl) thiourea." Journal of Coordination Chemistry 59 (2006): 223-8.
5. Sevgi Karakus and Sevim Rollas. "Synthesis and antituberculosis activity of new N-phenyl-N'- [4-(5-alkyl/arylamino-1, 3, 4-thiadiazole-2-yl) phenyl] thioureas." Il Farmaco 57 (2002): 577-81.
6. Raja Solomon Viswas, Wahajul Haq, Kumkum Srivastava, and Sunil Kumar Kumar Puri. "4-Aminoquinoline derived antimalarials: synthesis, antiplasmodial activity and heme polymerization inhibition studies." European journal of medicinal chemistry. 45 (2010): 4990-6.
7. Ahuja, Sapna, Ramya Raghunathan, Elango Kumarasamy, Steffen Jockusch, and Jayaraman Sivaguru. "Realizing the Photoene Reaction with Alkenes under Visible Light Irradiation and Bypassing the Favored [2+2]-Photocycloaddition." Journal of the American Chemical Society 140 (2018): 13185-13189.
8. Smith L. Holt and Richard L. Carlin. "Some Transition Metal Complexes of Substituted Thioureas. II. Nickel (II)." Journal of the American Chemical Society 86 (1964): 3017-24.
9. Carmen Puglisi and Robert Levitus. "Some substituted thiourea complexes of nickel (II) thiocyanate." Journal of Inorganic and Nuclear Chemistry 29 (1967): 1069-77.
10. Peter J. Sadler and Zijian Guo." Metal complexes in medicine: design and mechanism of

action." Pure and Applied Chemistry 70 (1998): 863-71.

11. Ali M. Hassan, Osama A. G. Wahba, A. M. Naser and A. Mohy Eldin. "Microwave synthesis and spectroscopic studies of some complex compounds as pigments and their applications in paints." Journal of Coatings Technology and Research 13 (2016):517-25.
12. Shao-Yong Kea and Si-Jia Xue. "Synthesis and herbicidal activity of N- (o-fluorophenoxyacetyl) thioureas derivatives and related fused heterocyclic compounds." Arkivoc 10 (2006):63-8.
13. Amel F. Elhusseiny, Ali Eldissouky, Ahmed M. Al-Hamza, and Hammed H.A.M. Hassan. "Metal complexes of the nanosized ligand N-benzoyl-N'- (p-amino phenyl) thiourea: Synthesis, characterization, antimicrobial activity and the metal uptake capacity of its ligating resin." Journal of Molecular Structure 1100 (2015):530-45.
14. Tuncay Yesilkaynaka, Celal Özpinarb, Fatih Mehmet, and Emenc Burhan, et al. "N- ((5-chloropyridin-2-yl) carbamothioyl) furan-2-carboxamide and its Co (II), Ni (II) and Cu (II) complexes: Synthesis, characterization, DFT computations, thermal decomposition, antioxidant and antitumor activity." Journal of Molecular Structure 1129 (2017):263-70.
15. Tuncay Yesilkaynak, Harun Muslu, Celal Özpinar, and Fatih Mehmet Emen, et al. "Novel thiourea derivative and its complexes: Synthesis, characterization, DFT computations, thermal and electrochemical behavior, antioxidant and antitumor activities." Journal of Molecular Structure 1142 (2017):185-93.
16. Mohammed Fouda, Manal Abd-Elaziz Abd-ElZaher, Mohamad Shakdofa, El-Saied Faoula, et al. "Synthesis and characterization of transition metal complexes of N'- [(1, 5-dimethyl-3-oxo-2-phenyl-2, 3-dihydro-1H-pyrazol-4-yl) methylene] thiophene-2-carbohydrazide." Trans ition Metal Chemistry 33 (2008):219-28.
17. Yusof MS, Khairul WM, Yamin BM. "Synthesis and characterisation a series of N- (3, 4-dichlorophenyl)-N'- (2, 3 and 4-methylbenzoyl) thiourea derivatives." Journal of Molecular Structure 975 (2010):280-4.
18. Omyma A.M. Ali, Samir M. El-Medani, Maha R. Abu Serea, and Abeer S.S. Sayed. "Unsymmetrical Schiff base (ON) ligand on complexation with some transition metal ions: Synthesis, spectral characterization, antibacterial, fluorescence and thermal studies." Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy 136 (2015):651-60.
19. Vinod P. Singh, Singh Sakshi, Divya Pratap Singh, and Pooja Singh. Synthesis, spectral and single crystal X-ray diffraction studies on Co (II), Ni (II), Cu (II) and Zn (II) complexes with o-amino acetophenone benzoyl hydrazone." Polyhedron 56 (2013):71-81.
20. Gülsah Kurt, Fatih Sevgi, Bedrettin Mercimek. "Synthesis, characterization, and antimicrobial activity of new benzoylthiourea ligands." Chemical Papers 63 (2009): 548-53.
21. Gunasekaran Nanjappan, Nattamai Bhuvanesh, Ramasamy Karvembu. "Synthesis, characterization and catalytic oxidation property of copper (I) complexes containing monodentate acylthiourea ligands and triphenylphosphine." Polyhedron 12 2 (2017):39-45.
22. Chernyavskaya AA, Natalia V Loginova, Polozov GI, Shadyro OI, Sheryakov AA, Bondarenko EV. "Synthesis and antimicrobial

activity of silver (I) and copper (II) complexes with 2- (4, 6-di-tert-butyl-2, 3-dihydroxyphenylsulfanyl) acetic acid." Pharmaceutical Chemistry Journal 40 (2006):413-5.

23. Murilo Massonia, Juan C. Tenorio, Clavijob Legna, and Colina-Vegasc et al. "Propyl gallate metal complexes: Circular dichroism, BSA-binding, antioxidant and cytotoxic activity." Polyhedron 129 (2017):214-21.
24. Abd El-Wahab ZH. "Complexation of 4-amino-1, 3 dimethyl-2, 6 pyrimidine-dione derivatives with cobalt (II) and nickel (II) ions: synthesis, spectral, thermal and antimicrobial studies." Journal of Coordination Chemistry 61 (2008):1696-709.
25. Willis B. Person. "A criterion for reliability of formation constants of weak complexes." Journal of the American Chemical Society 87 (1965):167-70.
26. Sharma Rajesh and Ambwani Jai. "Synthetic, Structural and Antimicrobial Studies of some Macrocyclic Ligands and their Copper (II) Complexes." Journal of the Indian Chemical Society 72 (1995):507-9.
27. Bipin Bihari Mahapatraa, Satyanarayan Chaulia, Ashish Kumar Sarangi, Satyanarayan Dehurya, et al.. "Synthesis, characterisation, spectral, thermal, XRD, molecular modelling and potential antibacterial study of metal complexes containing octadentate azodye ligands." Journal of Molecular Structure 1087 (2015):11-25.
28. Farideh Heidari, S. Jamil A. Fatemi, S. Yousef Ebrahimipour, and Hadi Ebrahimnejadb. "Six-coordinate oxo-vanadium (V) dimer complex with methoxy bridging: Synthesis, crystal structure, biological activity and molecular docking." Inorganic Chemistry Communications76 (2017):1-4.
29. Parveez Gull, Manzoor Ahmad Malik, Ovas Ahmad Dar, and Athar Adil Hashmi. "Design, synthesis and spectroscopic characterization of metal (II) complexes derived from a tetradentate macrocyclic ligand: Study on antimicrobial and antioxidant capacity of complexes." Microbial pathogenesis 104 (2017):212-6.
30. Reedijk Jan. "Improved understanding in platinium antitumour chemistry." Chemical Communications (1996):801-6.
31. Lei Qiao, Jie Huang, Wei Hu, Yu Zhang, et al. "Synthesis, characterization, and in vitro evaluation and in silico molecular docking of thiourea derivatives incorporating 4-(trifluoromethyl) phenyl moiety." Journal of Molecular Structure 1139 (2017):149-59.