Microbicidal Studies of AMIC Acids Based Novel Ligands and Their Metal Complexes

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Abstract

The increasing resistance of microorganisms to conventional antimicrobial agents has encouraged the search for new chemotherapeutic compounds with enhanced biological activity. *Amic acids*, which are mono-amide derivatives of dicarboxylic acids containing both amide (–CONH–) and carboxylic (–COOH) functional groups, are well known for their strong chelating ability and versatile coordination behavior. Owing to the presence of mixed donor atoms (O and N), amic acid–based ligands can form stable complexes with transition metal ions and exhibit significant biological relevance. In the present study, a series of newly synthesized amic acids based novel ligands (L-1 to L-8) and their transition metal complexes with Cu(II), Zn(II), Co(II), Ni(II), and Mn(II) were evaluated for their antifungal and antibacterial properties. Antifungal activity was assessed against five plant pathogenic fungi using the poisoned food technique at 1000 ppm concentration, while antibacterial activity was determined by the disc diffusion method against selected Gram-positive and Gram-negative bacterial strains. The results revealed that metal chelation significantly enhances antimicrobial activity compared to free ligands, which may be attributed to increased lipophilicity and improved penetration of the metal complexes into microbial cell membranes. Among the metal ions studied, copper complexes exhibited the highest antimicrobial efficacy. The overall activity trend followed the order Cu > Zn > Co > Ni > Mn. These findings suggest that amic acids based metal complexes possess promising potential as effective antimicrobial agents for agricultural and pharmaceutical applications.

Keywords : *Metal complexes; Antifungal activity; Antibacterial activity; Chelation effect and Transition metals etc.*

1. Introduction

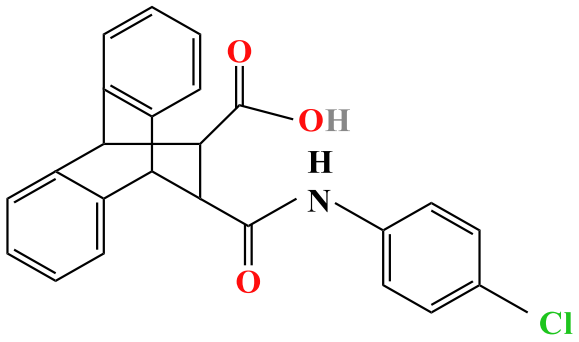
Infectious diseases constitute a major category of human and plant disorders, making the effective management of antimicrobial agents a matter of paramount importance. Chemotherapy refers to the treatment of parasitic infections in which pathogenic organisms such as bacteria, fungi, protozoa, viruses, and worms are destroyed or inhibited without causing damage to the host. The concept of selective toxicity was introduced by Paul Ehrlich, who emphasized the need for chemical agents that selectively target parasites while minimizing harm to host tissues.

Historically, several natural substances were employed for therapeutic purposes. Ancient civilizations used plant-derived compounds such as male fern, chenopodium, and chaulmoogra oil for the treatment of parasitic diseases. However, the scientific foundation of modern chemotherapy began in the early twentieth century when Ehrlich observed that certain dyes selectively stained and killed microorganisms. His work laid the groundwork for rational drug design.

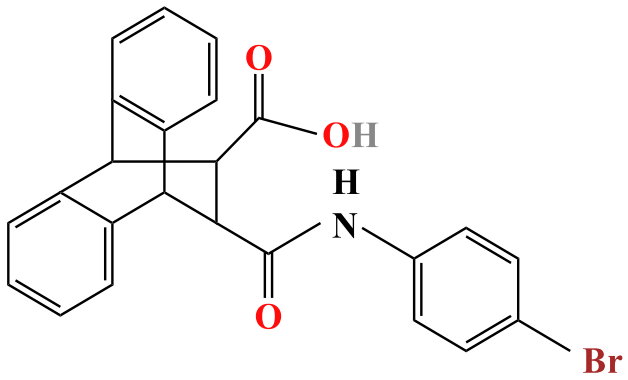
The discovery of penicillin by Alexander Fleming in 1928 and its subsequent development by Florey and Chain marked a revolutionary advancement in antimicrobial therapy. Since then, extensive research has focused on heterocyclic compounds and metal-based drugs due to their broad spectrum of biological activities. Quinoline derivatives, salicylic acid, and 8-hydroxyquinoline are well-known for their antimicrobial properties.

It has been observed that coordination of organic ligands with metal ions often enhances biological activity. This enhancement is attributed to the chelation effect, which reduces the polarity of the metal ion and increases the lipophilicity of the complex, facilitating penetration through microbial cell membranes. In view of these considerations, the present work investigates the antifungal and antibacterial activities of novel ligands and their metal complexes.

The ligands L-2 and L-6 are *amic acids–based ligands* derived from dicarboxylic acid frameworks and contain both amide (–CONH–) and carboxylic acid (–COOH) functional groups within the same molecular skeleton. The presence of these groups provides mixed donor sites (N and O atoms), enabling the ligands to coordinate with transition metal ions in a bidentate manner through the amide nitrogen and the carboxylate oxygen.



(Structure L-2)



(Structure L-6)

The structural arrangement of L-2 and L-6 favors the formation of stable five- or six-membered chelate rings upon complexation. Such chelation enhances the rigidity and stability of the metal complexes and plays a crucial role in improving their biological activity. Minor structural variations between L-2 and L-6 influence their electron-donating ability and steric environment, which in turn affect metal–ligand interaction and microbicidal efficiency.

In the present study, only the amic acids based ligands L-2 and L-6 were selected for microbicidal evaluation owing to their better chelating ability and favorable structural features. Their transition metal complexes exhibited enhanced antifungal and antibacterial activity compared to the free ligands.

2. Materials and Methods

2.1. Antifungal Activity

Test organisms

The antifungal activity was evaluated against the following plant pathogenic fungi:

- *Penicillium expansum*
- *Botrydepladia thiobromine*
- *Nigrospora* sp.
- *Trichothesium* sp.
- *Aspergillus niger*

Experimental procedure

Antifungal screening was carried out in vitro at 1000 ppm concentration using the poisoned food technique. Potato dextrose agar (PDA) medium was prepared using potato (200 g), dextrose (20 g), agar (20 g), and distilled water (1 L). Five-day-old fungal cultures were used for inoculation.

The test compounds were suspended in PDA medium, autoclaved at 120°C for 15 minutes under 15 atm pressure, poured into sterile Petri plates, and allowed to solidify. The fungal cultures were then inoculated, and the plates were incubated for five days. The percentage inhibition of fungal growth was calculated using the formula:

$$\% \text{ Inhibition} = \frac{X - Y}{X} \times 100$$

where
X = Area of fungal colony in control plate
Y = Area of fungal colony in test plate

2.2. Antibacterial Activity

Antibacterial activity was evaluated using the disc diffusion method on Mueller–Hinton agar (MHA) plates. Sterile discs impregnated with test compounds (100 µg/ml) were placed on inoculated plates and incubated at 35 °C for 18–20 hours. The zones of inhibition were measured in millimeters.

3. Results and Discussion

Table 1. Representative Antifungal Activity of Ligands and their Metal Complexes at 1000 ppm

Sample	Penicillium Expansum	Botrydepladia Thiobromine	Nigrospora Sp.	Trichothesium Sp.	A. Niger
L-2	72	71	73	70	71
$(L\text{-}2)_2\,Cu^{+2}$	78	81	79	78	78
$(L\text{-}2)_2\,Mn^{+2}$	74	75	74	73	
$(L\text{-}2)_2\,Co^{+2}$	73	78	76	75	75
$(L\text{-}2)_2\,Zn^{+2}$	75	79	77	76	77
$(L\text{-}2)_2\,Ni^{+2}$	76	78	74	75	75

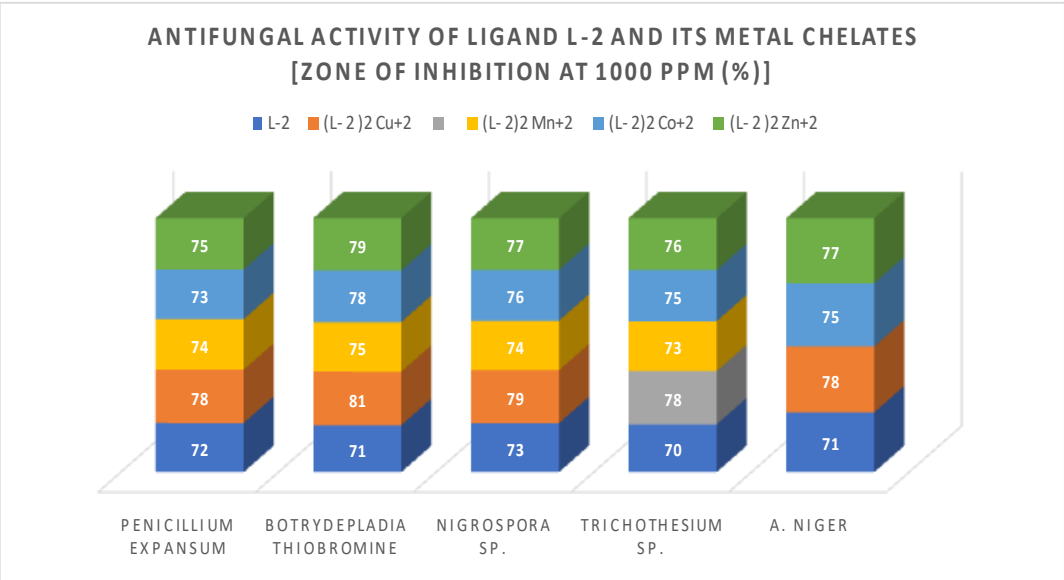


Figure 1. Antifungal activity of ligand L-2 and its metal chelates [Zone of inhibition at 1000 ppm (%)]

Table 2. Representative Antibacterial Activity of Ligand L-6 and Its Metal Complexes

Sample	Zone of inhibition (in mm)			
	Gram + Ve		*Gram - Ve*	
	Bacillus megaterium	Staphylococcus aureus	P. Aeruginosa	E-Coli
L-6	21	21	19	20
$(L\text{-}6)_2\ Cu^{+2}$	22	22	21	22
$(L\text{-}6)_2\ Mn^{+2}$	20	21	20	20
$(L\text{-}6)_2\ Co^{+2}$	21	21	21	22
$(L\text{-}6)_2\ Zn^{+2}$	22	21	21	22
$(L\text{-}6)_2\ Ni^{+2}$	21	21	21	21

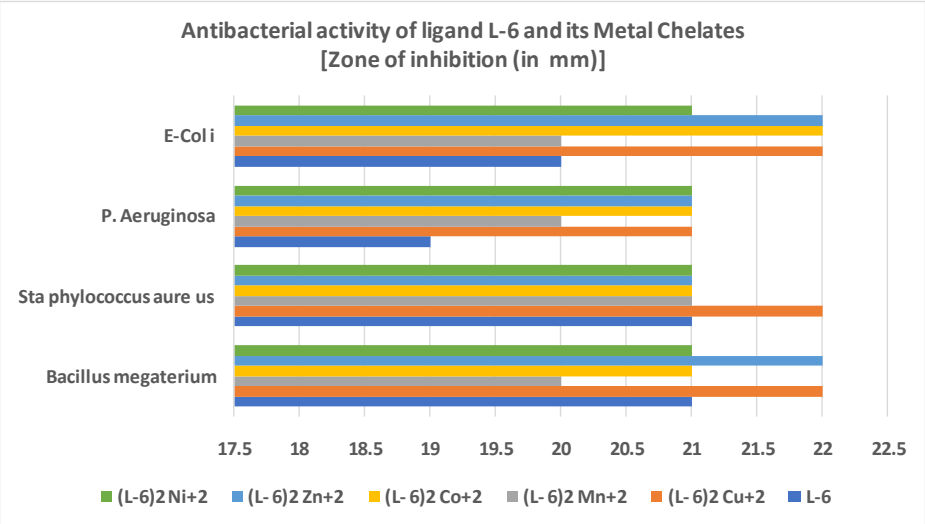


Figure 2. Antibacterial activity of ligand L-6 and its metal chelates [Zone of inhibition (in mm)]

The antifungal data indicate that all metal complexes exhibit higher toxicity against fungal strains compared to their parent ligands. This enhancement can be attributed to chelation, which increases the lipophilicity of the complexes and facilitates their diffusion through fungal cell membranes.

Ligands containing halogen substituents displayed greater fungicidal activity than those containing methyl, methoxy or ethoxy groups. Among the metal ions, copper complexes showed the highest antifungal activity, consistent with the well-established fungicidal nature of copper salts.

The overall antifungal activity followed the trend:
Cu > Zn > Co > Ni > Mn

The relative activity of ligands was observed in the order:
L-2 > L-6 > L-1 > L-7 > L-8 > L-3 > L-4 > L-5

These findings suggest that such metal chelates may serve as effective agricultural fungicides.

3.1. Antibacterial Activity

Test organisms

- Gram-positive: *Bacillus megaterium*, *Staphylococcus aureus*
- Gram-negative: *Pseudomonas aeruginosa*, *Escherichia coli*

3.2. Study of antibacterial activity

The antibacterial results demonstrate that metal complexes show enhanced activity compared to free ligands against both Gram-positive and Gram-negative bacteria. Gram-positive bacteria were generally more susceptible, possibly due to differences in cell wall structure.

Copper and zinc complexes exhibited superior antibacterial activity, while manganese complexes showed comparatively lower inhibition. The increased activity of metal complexes may be attributed to improved membrane permeability and interaction with bacterial enzymes.

4. Conclusion

The present investigation confirms that coordination of organic ligands with transition metal ions significantly enhances their antifungal and antibacterial activities. Copper complexes emerged as the most potent antimicrobial agents, followed by zinc and cobalt complexes. The observed structure–activity relationship highlights the importance of metal chelation and ligand substituents in determining biological efficacy. These findings suggest that such metal complexes have considerable potential for use as antimicrobial agents in agriculture and pharmaceutical applications.

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