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Stability constants, characterization, and Theoretical Modelling Studies of the Complexes of L- Ascorbic Acid with Transition Metal Ions

[1]Fatma M. El – Feky, [2]Eman A. Mohamed, [1]Sahar M. Ibrahiml, and [1]M. E. Moustafa
[1]Chemistry Department, Faculty of Science, Benha University, Benha, Egypt
[2] International Academy for Engineering and Media Science (IAEMS)

Abstract

Potentiometric measurements of L-ascorbic acid's ionization constant as a chelating drug and the stability constants of its Mn^{2+}, Fe^{3+}, Co^{2+}, Cu^{2+}, and Zn^{2+} metal complexes have been made in an ethanol-water (25% ethanol) and 1.0 molar NaCl medium. The complexes that were generated in solution had stoichiometry values of 1:1 and 2:1 (Asc: M), and the order of stability was Fe^{3+}> Cu^{2+}> Mn^{2+}> Co^{2+}> Zn^{2+}. Different physico-chemical techniques were used to confirm the structures of the solid complexes, where a satisfactory agreement between the proposed and theoretical formulae was found. The antibacterial activity toward some Gram-positive, Gram-negative, and fungi was studied, taking Ciprofloxacin and Ampicillin as standards. The outcomes show that the metal complexes have a marginally higher level of activity than ascorbic acid in its free form. The DMOL3 tool, designed for wide-scale density function theory (DFT), was used to calculate a few quantum chemical and energetic characteristics of the free drug and its metal complexes.

Keywords: L-ascorbic acid, potentiometric studies, metal complexes, biological activity, molecular modeling.

1. Introduction

A large number of enzymes' biological functions depend on metal ions. Enzyme-, ligand-, and metal-bridge complexes are among the different ways that metals and proteins can interact [1]. Metals can function as Lewis acids, structural regulators, electron donors, or acceptors [2]. Numerous biomolecules, such as carboxylic acids, peptides, and amino acids, can form metal complexes with

varying stabilities that are significant in the field of biomedicine. Certain medications have specific therapeutic effects (e.g., antibacterial, diuretic, and antidepressant) as a result of the metallic ion (Cu^{2+}, Zn^{2+}, Fe^{2+}, Mg^{2+}, etc.) complexing with them [3]. Applications for chelating drug complexes with transition metals can be found in industrial, analytical, and medical procedures [4-9]. These medications combine with metal ions to produce stable five- or six-membered rings, either at the beginning of the body's metabolic processes or afterwards. Studies on metal complexes using chelating medications as ligands have received a lot of interest during the past few decades [10–15]. The most well-known instance is cisplatin, also known as cis-$[PtCl_2(NH_3)_2]$ chemically, which has anticancer properties because it forms a ternary complex in which two chloride ions are swapped out for adjacent N-7 guanines on the cancer cell's DNA [16]. In comparison to free ligands, the interface between metal ions and antibiotics can boost the activity of the complexes [17]. L-ascorbic acid facilitates several metabolic processes, including the transformation of cholesterol to bile acids, the activation of vitamin B and folic acid, and the conversion of the amino acid tryptophan to the neurotransmitter serotonin. It is an antioxidant that lowers the severity of allergic reactions, protects the immune system from damage caused by free radicals, and aids in the body's defense against infections [18].

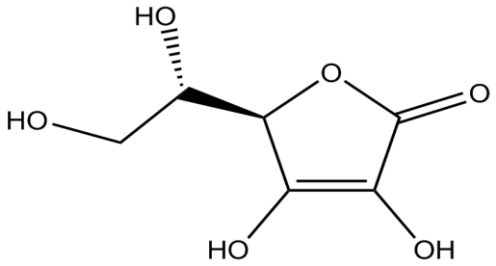
First, the potentiometric approach is used to examine the equilibrium of L-ascorbic acid ionization and its reaction with Mn (II), Fe (III), Co (II), Cu (II), and Zn (II) ions in solution at ambient temperature and ionic strength of 1.0 M NaCl. Second, the solid-state complexes are synthesized and characterized, and their biological activities are studied against a variety of Gram-positive and Gram-negative bacteria and fungi. Using the DMOL3 tool, which is designed for wide-scale density function theory (DFT), some quantum chemical and energetic properties of the free drug and its metal complexes were estimated as part of a theoretical investigation.

While many studies concentrate on well-established metal complexes like cisplatin, my emphasis on L-ascorbic acid and its role as a chelating agent in forming stable metal complexes is less common. This could lead to new insights into the antioxidant properties and metabolic roles of L-ascorbic acid in biomedicine.

2. Experimental

All chemicals used in this study are of highest quality and were used as received.

L-Ascorbic acid has the following structure:



Investigations on Solvable Metal Complexes

Conductometric titration

50 mL of L-ascorbic acid solution (10^{-3} M) was titrated with 10^{-2} M solution of metal ions using a microburette with mechanical stirring. After each increment of titrant, the conductance is measured by a conductance meter type YSI Model 32.

Potentiometric titration

Potentiometric titration was carried out in accordance with Bjerrum's[19] instructions, which were, in short, to prepare the following three mixtures:

5.0 mL of 0.1 M HCl and 5.0 mL of 1.0 M NaCl make up the mixture (A).

Blend (A); Blend (B); plus 25 milliliters of ascorbic acid (10-3 M).

Blend (C): 2.0 mL of a 2.5x10-3 M metal ion solution plus mixture (B)

To maintain the components' solubility, the volume of each mixture is increased to 50.0 mL of an aqueous solution containing 25% (v/v) ethanol. After that, the mixtures were potentiometrically titrated using an automated microburette and a 0.12 M NaOH solution (which included 25% ethanol) while being continuously stirred.
Getting the solid complexes ready The solid complexes were made by heating an aqueous solution containing 2.00 mmol of L-ascorbic acid for approximately 6 hours and then allowing it to cool at room temperature for the entire night. The mixture contained 50 mL of 1.00 mmol aqueous solutions of Mn (II), Fe (III), Co (II), Cu (II), and Zn (II) chlorides. The precipitated solids were removed by filtering, cleaned with distilled water, and then dried in a vacuum.

Measurements in physical terms

As stated in our earlier work [20], the synthesized solid complexes were characterized by FTIR spectroscopy, thermal analysis, absorption spectra in the solid state (Nujol mull), magnetic susceptibility, and molar conductivity tests.

Antimicrobial Activity

The disk diffusion technique, which was developed by Bauer et al. [21] and Moustafa et al. [22], was used to test the antimicrobial and antifungal susceptibilities against two Gram-positive bacteria (Streptococcus faecalis and Staph.

Aureus), two Gram-negative bacteria (Pseudomonas aeruginosa and Escherichia coli), and two fungi (Asper. niger and Asper. ocherationus). The substances were examined at a concentration of 0.420 mg/mL in DMF and contrasted with positive controls such as ampicillin and ciprofloxacin.

Molecular modeling

Using the Materials Studio package [23] and the DMOL3 program [20], quantum mechanical calculations were carried out for the large-scale density functional theory (DFT). The key energy and quantum chemical parameters were identified and addressed.

3. Results and discussion

Study of the complexes in solution

i- Conductometric titration:

By using conductometric titration, the stoichiometry of the complexes produced between L-ascorbic acid and Mn^{2+}, Fe^{3+}, Co^{2+}, Cu^{2+}, and Zn^{2+} was examined. Ten-2 M metal ion solution was introduced from the microburette to the drug solution (50 mL of 10-3 M) and stirred mechanically. Following each increment, the conductance of the resultant solution was measured and plotted against the mole ratio (L/M). The latter produced growing straight lines that intersected at mole ratios of 1:1 and 2:1 (Asc:M), showing that the removal of the H+ ion from the ascorbic acid molecule led to the development of stable complex species.

ii- **Potentiometric titration:**

The potentiometric technique, as suggested by Bjerrum [19], was used to determine the ionization constant of L-ascorbic acid (H-Asc → H^+ + Asc-) and the metal-lligand stability constants (M^{2+} +H-Asc →H^++Asc-M). Three mixtures, A, B, and C (discussed above), were separately titrated with 0.12 M NaOH freed from carbonate containing 25% (v/v) ethanol. The potentiometric titration curves for the free ligand and its metal complexes (refer to Fig. 1) are S-shaped, exhibiting a significant jump in mixtures (A) and (B). However, mixtures of C_n that contain the metal ions yield comparatively smooth curves. The order in which the end points for mixture titration grow is A< B< C_n.

The proton-ligand stability constant (pKH) of L-ascorbic acid is determined by titrating the ligand molecule in its protonated form, which is the cause of the mixture titration curve (B). Through curve (B) analysis, the average number of protons connected to the ascorbic acid molecule (n'_A) at various pH values is determined using the relation:

$$n'_A = Y + \frac{(V_1 - V_2)(N^0 + E^0)}{(V^0 + V_1)(TC_{L0})} \qquad (1)$$

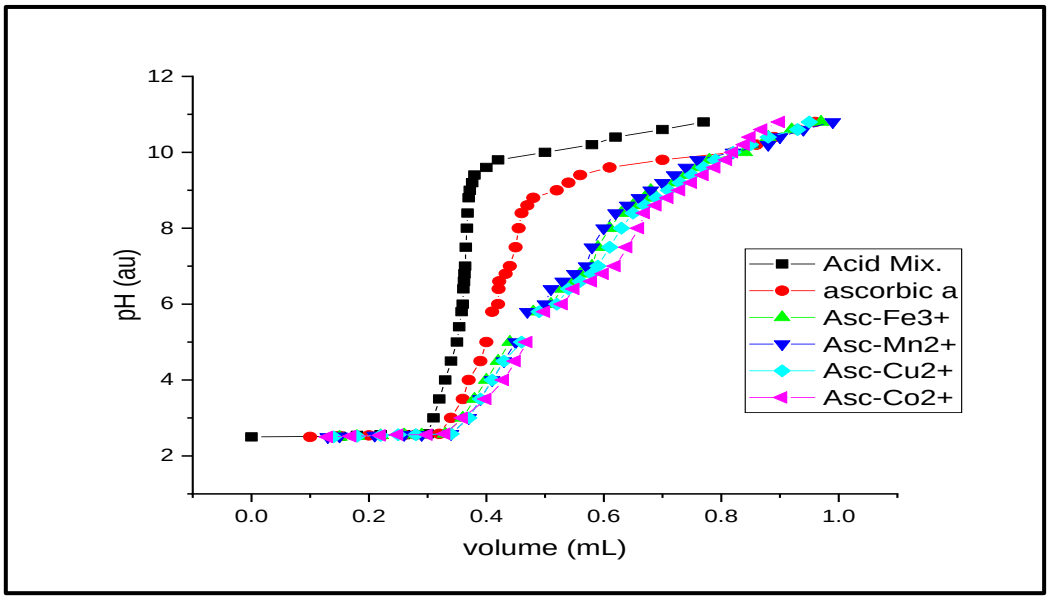


Fig.1: Potentiometric titration for Ascorbic acid and its Mn^{2+}, Fe^{3+}, Co^{2+} and Cu^{2+} complexes.

Where V^0 is the initial volume (50 mL), TC_{L0} is the total concentration of the ligand, Y is the total number of dissociable protons attached to the drug (Y = 2 for ascorbic acid), No is the normalcy of the NaOH solution, and E^0 is the initial concentration of the free acid. These values correspond to the volumes of NaOH consumed to reach the same pH in the titration curves of the acid mixture (A) and the L-ascorbic acid (B), respectively. The proton-ligand formation curve depicted in Fig. (2) is obtained by plotting n'_A against the pH of the solution. By interpolating at half n'_A integrals, the proton-ligand stability constants K_1H and K_2H are determined from this curve (c.f. Table 1).

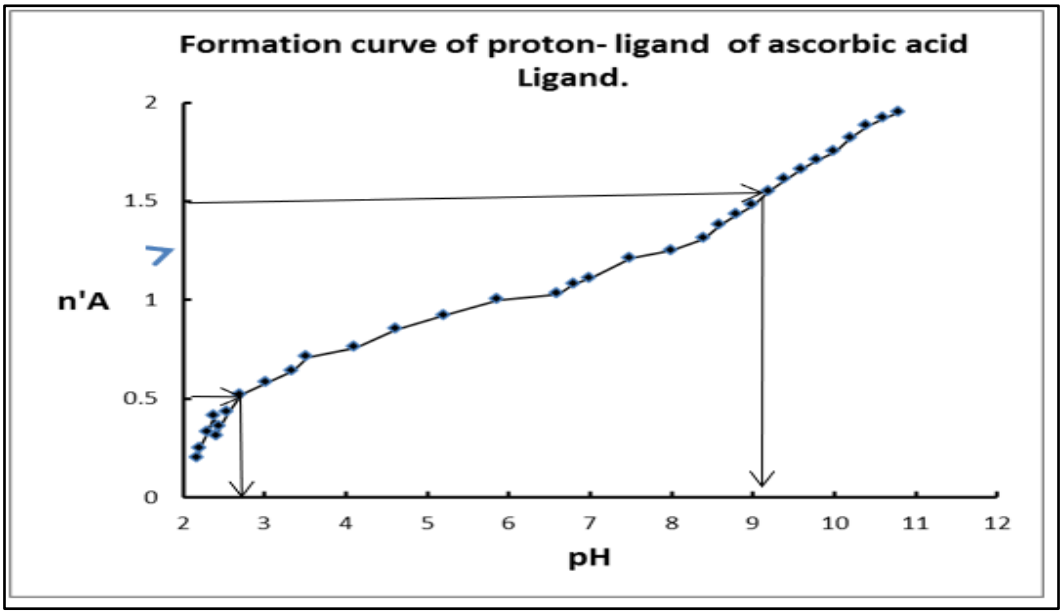
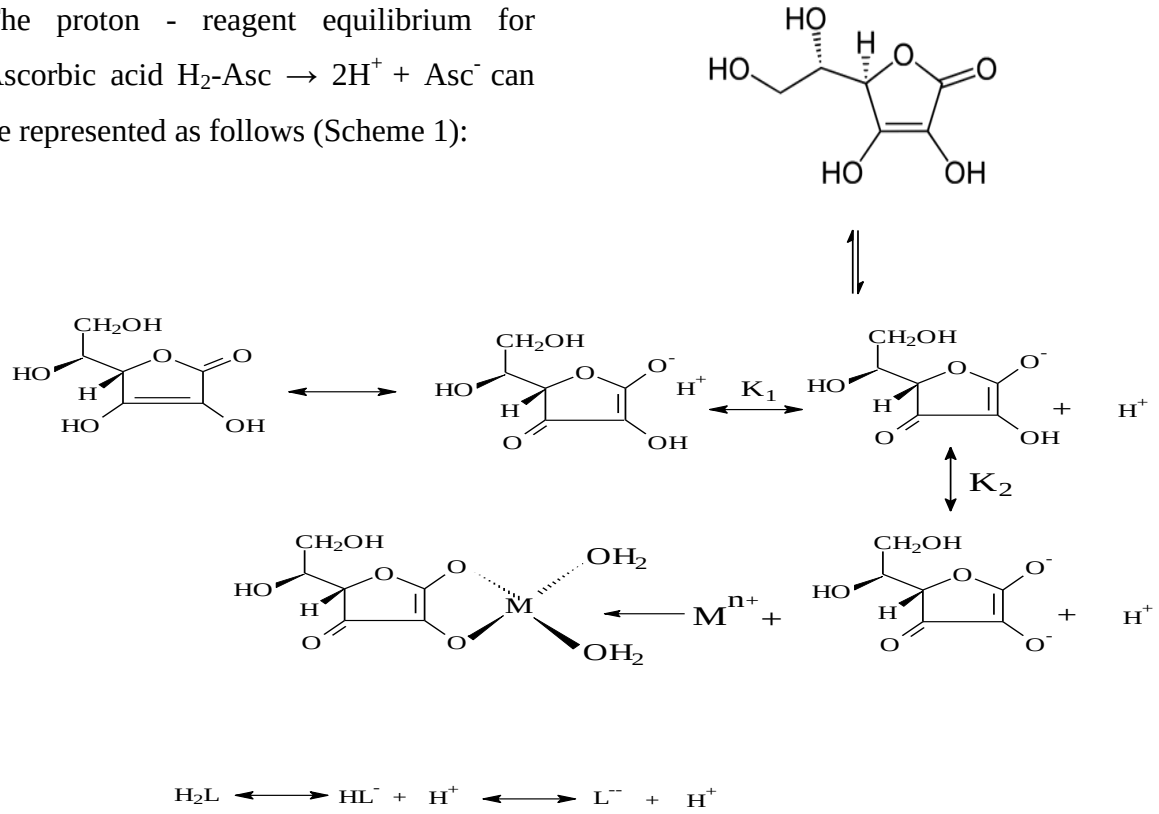


Fig. (2): Formation curve of proton - ligand stability constant of Ascorbic acid

The proton - reagent equilibrium for Ascorbic acid H_2-Asc → $2H^+$ + Asc^- can be represented as follows (Scheme 1):



Scheme 1: The proton - reagent equilibrium for Ascorbic acid

Using the following formulas, one may calculate the average number of ligand molecules attached per metal ion (n') and the free ligand exponent (PL) to find the stepwise formation constants of the complexes formed between L-Ascorbic acid and Mn^{2+}, Fe^{3+}, Co^{2+}, Cu^{2+}, and Zn^{2+} in solution:

$$n' = \frac{(V_2 - V_3)(N^0 + E^0)}{(V^0 + V_1)n'_A(TC_{M0})} \qquad (2)$$

$$pl = \log \frac{\sum_{n=0}^{n=i} \beta_n^H \left(\frac{1}{antilog\ pH}\right)^n}{(TC_{L0} - n'^{TC_{M0}})} \times \frac{V^0 + V_3}{V^0} \qquad (3)$$

where ϵnH is the overall stability constant and TC_{M0} is the total concentration of metal ions in solution. The values of n' for each metal ion can be plotted against PL, and the half-integrals can be interpolated to get the stepwise stability constants of the complexes, as seen in Fig. 3. The metal-ligand stability constants are computed from these curves, and the results are reported in Table 1.

The following is a general mechanism that describes how the ligands and their metal mixes react when alkali is added:

HJL + Mn^+ → [MH(J-1)L]Plus (n-1) + H^+ + OH^- → [MH(J-1)L]HJL + Mn^+ or (n-1) $^+$ + H_2O → (n-J)+[ML] + JH^+ + JOH^- → JH2O + [ML](n-1)+, where J = 2, 3, or 4

Ascorbic acid forms complexes with Mn^{2+}, Fe^{3+}, Co^{2+}, Cu^{2+}, and Zn^{2+} ions, according to titration results shown in Tables (1). The values of the overall stability

constants rise in the following order:

$Fe^{3+}>Cu^{2+}>Mn^{2+}>Co^{2+}>Zn^{2+}$. (3)

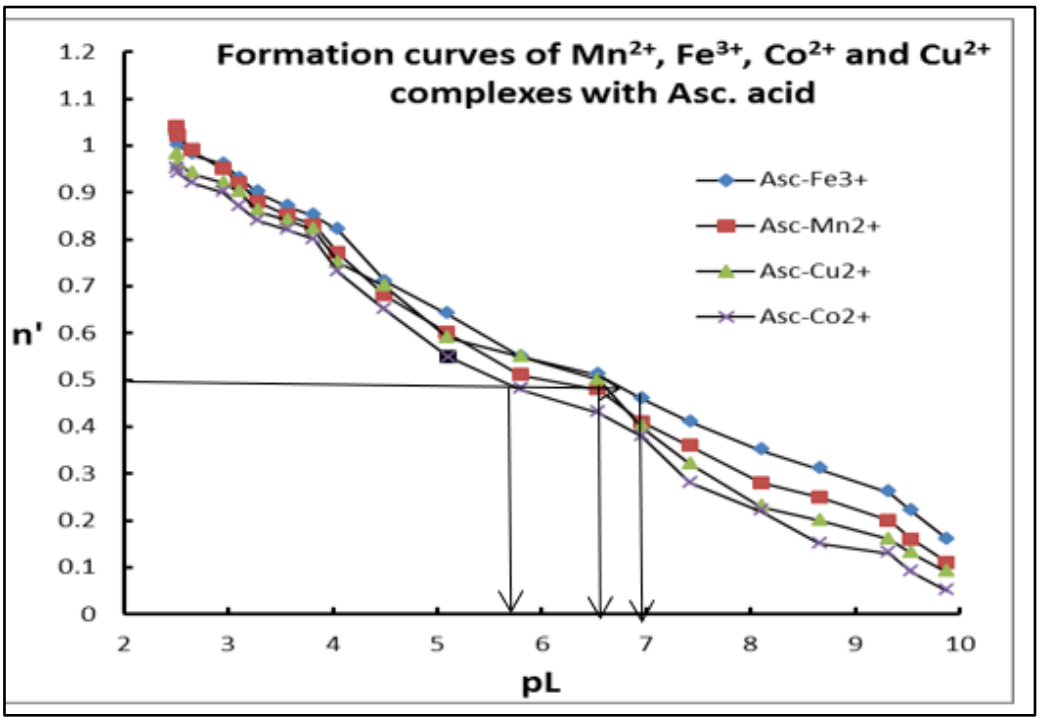


Fig.(3): Formation curves of Mn^{2+}, Fe^{3+}, Co^{2+} and Cu^{2+} complexes with Ascorbic acid.

Table 1: Ionization constants of L-Ascorbic acid and stepwise formation constants of its metal complexes from potentiometric titration.

Comp.	Stoichiometry (Asc:M)8	Ionization constant		Stability constant		
		pK_H1	pK_H2	pK_b1	pK_b2	Logβn
Ascorbic acid	------	2.81	8.89	-----	----	---
Asc-Mn^{2+}	(1:1) & (2:1)	---	---	4.55	6.62	11.17
Asc-Fe^{3+}	(1:1) & (2:1)	---	---	4.63	6.91	11.54
Asc-Co^{2+}	(1:1) & (2:1)	---	---	4.08	5.80	9.88
Asc-Cu^{2+}	(1:1) & (2:1)	---	---	4.62	6.61	11.23
Asc-Zn^{2+}	(1:1) & (2:1)	---	---	4.04	5.77	9.81

Part B: Studies on metal complexes in solid state.

Structure elucidation.

Using the stoichiometric ratio of 2:1 (Asc:M), solid complexes of Mn^{2+}, Fe^{3+}, Co^{2+}, Cu^{2+}, and Zn^{2+} with ascorbic acid were formed. The molecular structure was characterized by elemental analysis, molar conductivity, thermal analysis, IR, and UV-Vis spectra. With the exception of the Fe (III) ion, the percents of C, H, and M (Table 2) demonstrate a satisfactory degree of consistency between the calculated and observed values. In contrast, the molar conductivity values indicate the non-ionic character of the complexes.

Table (2): C,H,N and N analysis and molar conductivity of Mn^{2+}, Fe^{3+}, Co^{2+} Cu^{2+} and Zn^{2}complexes with L-Ascorbic acid (2:1) (L: M)

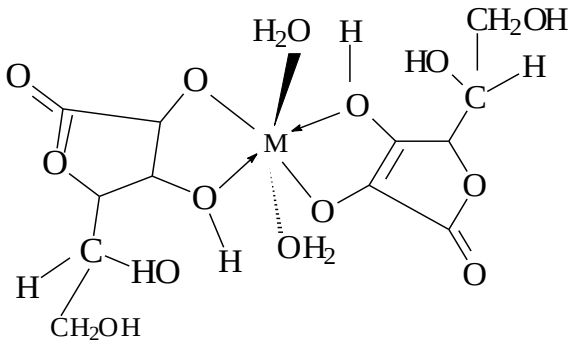
Compllex;	Tentative formula	M.Wt.	Elemental analysiis*			
			%C	%H	%M	Λ_m**
Asc-Mn^{2+}	$[C_{12}H_{18}O_{14}Mn]$	441.20	32.67	4.11	12.45	8.92
Asc-Fe^{3+}	$[C_{12}H_{18}O_{14}Fe]^{+}Cl$	447.57	30.19 (30.64)	3.80 (4.04)	11.73 (11.22)	22.68
Asc-Co^{2+}	$[C_{12}H_{18}O14Co]$	445.20	32.36	4.08	13.24	9.46
Asc-Cu^{2+}	$[C_{12}H_{18}O14Cu]$	449.81	32.07 (32.70)	4.04 (4.22)	14.02 (13.87)	9.85
Asc-Zn^{2+}	$[C_{12}H_{18}O_{14}Zn]$	451.65	31.91 (32.08)	4.02 (4.24)	14.48 (13.91)	8.28

** $ohm^{-1}cm^{2}mol^{-1}$ *values between parentheses are found values

i- ***Thermal analysis*** of the prepared complexes shows that they have high melting points and are stable at ambient Thermogravimetric analysis shows that they lose the physically adsorbed water molecules at about 85–100 ^{o}C, while the coordinated molecules dehydrate within the temperature range of 135–150 ^{o}C. The anhydrous metal complexes showed thermal stability up to ≈ 350 ^{o}C, then decomposed, leading to the corresponding metal oxides as final products.

ii- ***The IR spectrum*** of the free Ascorbic acid is compared to those of the metal complexes. This comparison revealed that the two prominent bands, which correspond to the stretching and bending vibrations of the two nearby hydroxyl groups, respectively, at 3450 cm^{-1} and 1121 cm^{-1}, are moved to lower frequencies, suggesting their role in the creation of coordinate bonds. However, as a result of the carbonyl group in ascorbic acid stretching vibration, the strong band at 1627 cm^{-1} experiences only a minor change upon complex formation. The stretching vibrations of the free OH groups in the ascorbic acid moiety and the

hydroxyl groups of coordinated water molecules are what cause the broad band that is found within the wavenumber range of 2465 to 2448 cm^{-1}. The stretching vibrations of the M–O bond cause a new band to appear in the far infrared portion of the metal chelates' spectra in the 455–525 cm^{-1} range. The solid complexes' molar conductance in DMF measurements revealed that they are non-ionic (with the exception of Fe3+), meaning that only one of the two nearby hydroxyl groups is deprotonated. As a result, ascorbic acid functions as a monobasic bidentate in an OO manner, and the bonding mode is shown as follows:



Scheme 2: mode of bonding of Ascorbic acid.

In Vitro Biological Activity

i- Biological activity

The disk diffusion technique developed by Bauer et al. [21] and as described in our previous work [22] was used to test the antimicrobial and antifungal susceptibilities of ascorbic acid and its metal complexes against two Gram-positive bacteria (Streptococcus faecalis and Staph. aureus), two Gram-negative bacteria (Pseudomonas aeruginosa and Escherichia coli), and two fungi (Asper. niger and Asper. ocheratious). Tested at 0.420 mg/mL in DMF, the compounds were contrasted with ampicillin and ciprofloxacine as positive controls. According to the findings shown in Tables (4a, b), the metal complexes exhibit marginally higher levels of activity than ascorbic acid in its free form. By obstructing the bacteria's active sites, the lipophilic character of the metal ions in complexes is responsible for this increased activity [23]. When it comes to bacteria, the examined species are more effective than fungi. The results are displayed graphically in Fig. 4a, b) and numerically in terms of inhibition zone and activity index in tables 4a, b.

Table (3): IR vibrational frequencies (cm^{-1}) off some function groups off tthe Ascorbic acid and its Mn^{2+}, Fe^{3+}, Co^{2+} Cu^{2+}, and Zn^{2+} metal compllexes

Compound	ν_{OH}	$\nu_{H2\,O}$	$\nu_{C=O}$	$\nu_{C=C}$	δ_{OH}	νM-O
Ascorbic acid	3450	3465	1627	1415	1121	---
Asc.- Mn^{2+}	3419	3457	1623	1402	1101	455
Asc.- Fe^{3+}	3435	3462	1624	1493	1115	505
Asc.- Co^{2+}	3418	3465	1622	1403	1126	500
Asc.- Cu^{2+}	3433	3455	1621	1414	1105	504
Asc.- Zn^{2+}	3437	3448	1620	1439	1107	525

Table (4-a): Tthe inhibition zone diameter (mm) and % activity index for antibacterial activities off Ascorbic acid and some off its metal compllexes.

Compound	Grram positive bacteria				Grram negative bacteria			
	Streptococcus faecaliis		*Staph.aureus*		*Pseudomonas aeruginosa*		*Escherichia coli*	
	Inh.zone, mm	% Ac.Ind	Inh.zone, mm	% Ac.Ind	Inh.zone, mm	% Ac.Ind	Inh.zone, mm	% Ac.Ind
Ciproffloxacin	15	100	15	100	22	100	23	100
Ascorbic acid	14.6	97.33	13.4	89.33	19.5	88.64	20.0	86.96
Asc- Mn^{2+}	18.8	125.33	13.4	89.33	20.3	92.27	19.3	83.91
Asc- Fe^{3+}	19.8	132.0	14.8	98.67	22.6	102.7	21.2	92.17
Asc- Co^{2+}	13.7	91.33	19.4	129.3	21.0	95.45	19.5	84.78
Asc- Cu^{2+}	13.4	89.33	18.9	126.0	19.8	90.00	22.3	96.96
Asc- Zn^{2+}	17.5	116.67	12.6	84.0	22.8	104.1	22.2	96.52

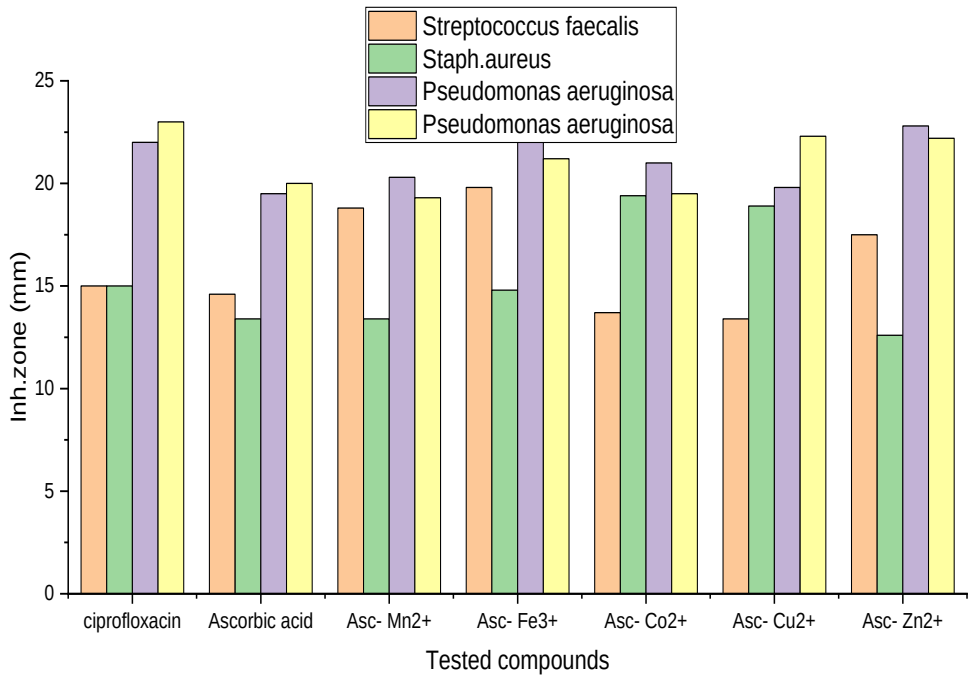


Fig. (4-a): Antibacterial activities of Ascorbic acid and its metal complexes in terms of inhibition zone diameter (mm).

Table (4-b): The inhibition zone diameter (mm) and % activity index for antifungal activities of its complexes:

Organiism	Fungi			
	Asper.niger		*Asper.ocheratious*	
	Inh.zone, mm	% Ac.Ind	Inh.zone, mm	% Ac.Ind
Ampicillin*	20	100	20	100
Ascorbic acid	19.5	97.5	19.0	95
Asc- Mn^{2+}	19.3	96.5	18.3	91.5
Asc- Fe^{3+}	19.6	98.0	19.2	96.0
Asc- Co^{2+}	19.0	95.0	19.5	97.5
Asc- Cu^{2+}	19.8	99.0	19.3	96.5
Asc- Zn^{2+}	18.8	94.0	16.2	81.0

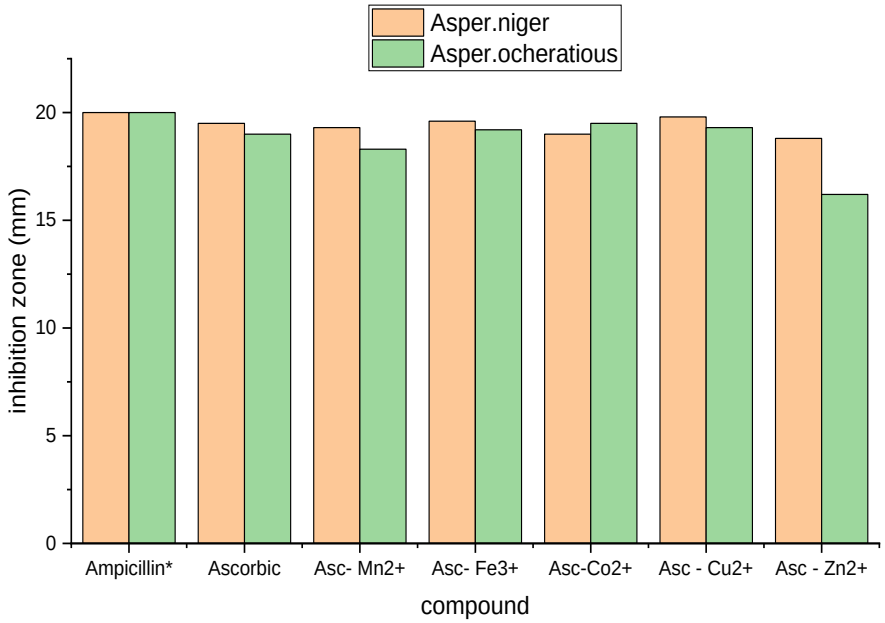


Fig. (4-b): Antifungal activity of Ascorbic acid and its metal complexes in terms of inhibition zone diameter (mm).

4. Theoretical studies

Molecular modelling

For the purpose of computing the molecular modeling, total density function, deformation density function, and frontier orbital energy (the HOMOs and LUMOs) for ascorbic acid and some of its complexes, quantum mechanical calculations using the DMOL3 program in the Materials Studio package were carried out. Table 5 provides the quantum chemical properties of the complexes under study.

The HOMOs and LUMOs, which are Frontier molecular orbitals (FMOs), are crucial for assessing the molecular chemical stability as well as the hardness-softness of the molecule [24]. Compound

chemical reactivity is represented by the energy gap, ΔE (EHOMO − ELOMO), where a system with a smaller value of ΔE is more reactive. As Table 5 illustrates, the energy gap of ascorbic acid is the lowest (2.284 eV), and it increases for complex species, suggesting their stability. The ability of a system to take electrons is described by the electrophilicity index (ω), where higher values of (ω) indicate a greater aptitude to do so. The increase in electron-accepting capacities is indicated by this value, which rises in the following order: Zn^{2+} > Cu^{2+} > Asc. > Fe^{3+}. The direction of charge transfer within a molecule is dictated by the electronic chemical potential (μ) value. Because an electrophile is a chemical entity that can take in electrons from its surroundings and must lose energy in order to receive electronic charge, this value is lower than that of the free ligand. Table 5 displays the computed values of electronegativity (χ), chemical potential (μ), global softness (σ), global hardness (η), additional electronic charge (ΔNmax), and global electrophilicity index (ω) [25]. Table 6 presents some of the energy characteristics of ascorbic acid and its compounds that were determined using the DFT approach.

Geometry Optimization with DFT Method

Fig. (5) displays the atomic numbers and molecular structures of the Fe^{3+}, Cu^{2+}, and Zn^{2+} complexes with ascorbic acid. Table (7) lists the significant bond lengths and bond angles. The data analysis shows that upon coordination, the free ligand's bond lengths and bond angles are somewhat changed.

Table (5): The calculated quantum chemical parameters of ascorbic acid and its complexes

Comp.	HOMO	LUMO	ΔE	ω	Σ	χ	M	H	ΔN_{max}
Ascorbic acid	-5.171	-2.887	2.284	1.142	0.8757	-4.029	4.029	7.1072	3.5280
Asc.- Cu^{2+}	-5.632	-3.344	2.288	1.144	0.8741	-4.488	4.488	8.8034	3.9231
Asc - Fe^{3+}	-4.974	-3.49	1.484	0.742	1.3477	-4.232	4.232	12.0686	5.7035
Asc - Zn^{2+}	-5.181	-1.687	3.494	1.747	0.5724	-3.434	3.434	3.3750	1.9657

Table(6): Some energetic properties of L-Ascorbic acid and its complexes calculated by DFT-method

Comp.	Eenergy components (Kcal/mol)						Binding energy (Kcal/mol)	Dipole moment (debye)
	Sum of atomic energies	Kinetic energy	Electrostatic energy	Exchange-correlation energy	Spin polarization energy	Total energy		
Asorbic	-4.28 $X10^6$	$-3.28X10^3$	$-1.44X10^3$	$6.95\ X10^3$	$7.28\ X10^3$	$-4.3X10^5$	$-2.03\ X10^3$	6.4846
Asc - Cu^{2+}	-1.09 $X10^6$	$-8.96X10^3$	$1.17X10^3$	$1.57X10^3$	$1.45\ X10^3$	$-1.1X10^5$	$-4.76\ X10^3$	8.9105
Asc - Fe^{3+}	-1.04 $X10^6$	$-5.78X10^3$	$-1.86X10^3$	$1.69X10^3$	$1.52\ X10^3$	$-1.1X10^5$	$-4.44\ X10^3$	2.8602
Asc - Zn^{2+}	-1.11 $X10^6$	$-7.13X10^3$	$-9.75X10^3$	$1.99X10^3$	$1.45\ X10^3$	$-1.1X10^5$	$-4.65\ X10^3$	12.4812

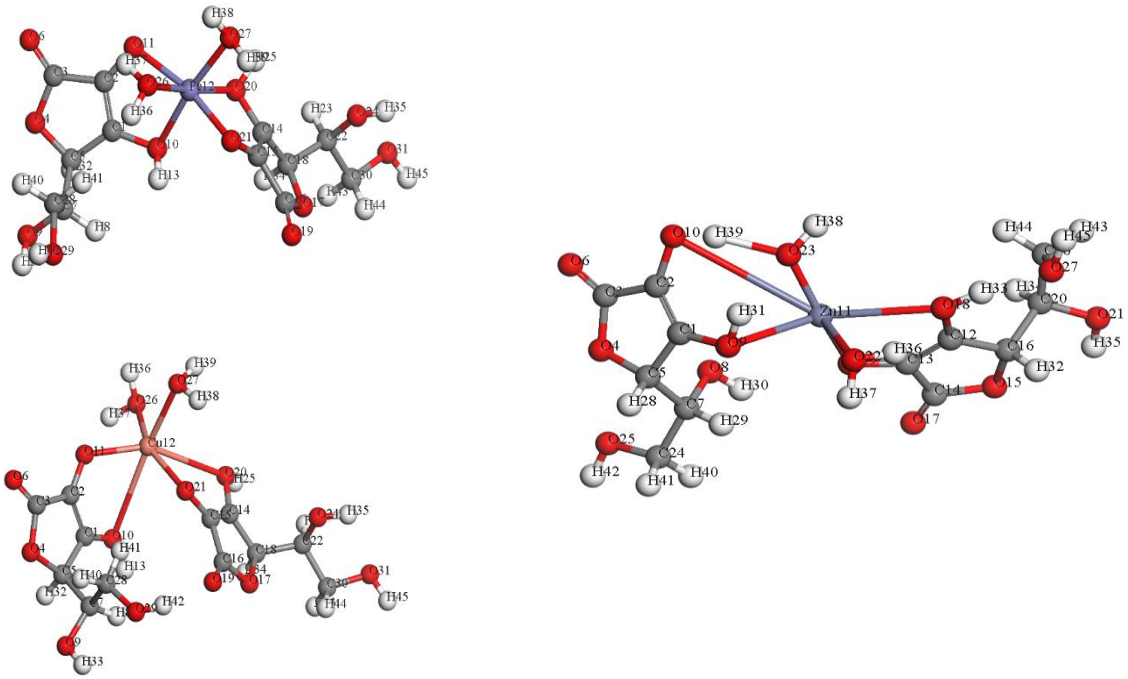
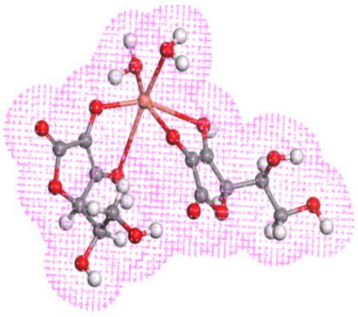


Fig. (5): The molecular structures and atomic numbering of the Fe^{3+}, Cu^{2+} and Zn^{2+} complexes with Ascorbic acid.

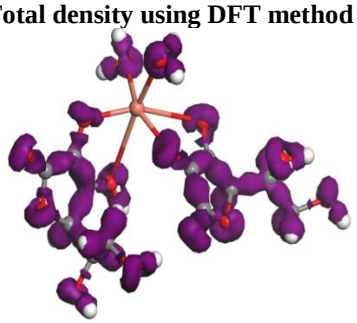
Table (7): Important bond lengths and bond angles of Fe^{3+}, Cu^{2+} and Zn^{2+} complexes

Bond length (Å)				Bond angle (o)	
For Asc-Cu^{2+}					
Cu(12)-O(27)	2.411	Cu(12)-O(27)	2.411	O(20)-Cu(12)-O(11)	146.348
Cu(12)-O(26)	2.487	Cu(12)-O(26)	2.487	O(20)-Cu(12)-O(10)	83.134
O(21)-Cu(12)	2.108	O(21)-Cu(12)	2.108	O(20)-Cu(12)-O(10)	83.134
O(20)-Cu(12)	3.102	O(20)-Cu(12)	3.102	O(11)-Cu(12)-O(10)	65.182
O(11)-Cu(12)	2.092	O(11)-Cu(12)	2.092	Cu(12)-O(11)-C(2)	118.699
For Asc-Fe^{3+}					
Fe(12)-O(27)	2.084	O(20)-Fe(12)	2.153	O(21)-Fe(12)-O(20)	85.886
Fe(12)-O(26)	2.121	O(11)-Fe(12)	2.107	O(21)-Fe(12)-O(11)	166.074
O(21)-Fe(12)	2.006	Fe(12)-O(27)	2.084	O(21)-Fe(12)-O(20)	85.886
				O(21)-Fe(12)-O(11)	166.074
				O(21)-Fe(12)-O(10)	89.693
For Asc-Zn^{2+}					
Zn(11)-O(23)	1.862	O(9)-H(31)	0.981	O(10)-Zn(11)-O(9)	51.518
Zn(11)-O(22)	2.348	O(9)-Zn(11)	2.594	Zn(11)-O(10)-C(2)	75.146
O(19)-Zn(11)	1.883	O(18)-Zn(11)	2.898	O(18)-Zn(11)-O(9)	141.564
O(10)-Zn(11)	4.024	O(9)-Zn(11)	2.594	O(22)-Zn(11)-O(18)	66.295

The total density, spin density, and 3D plots of frontier orbital energies using the DFT method of the complex were studied. An example for the Asc-Cu^{2+} and Asc-Fe^{3+} complex is shown in Figs. 6 and 7.



Total density using DFT method for ligand Asc. - Cu^{2+}



Deformation density using DFT method for Asc. – Cu^{2+}

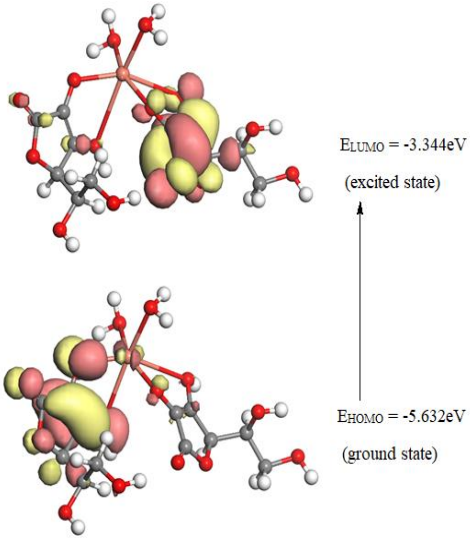
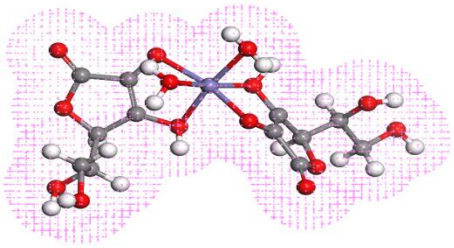
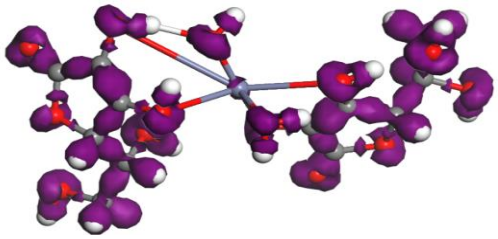


Fig. (6): Total density, Deformation density and 3D plots frontier orbital energies for Asc,- Cu^{2+} complex using DFT method.



Total density using DFT method for ligand Asc.- Fe^{3+}



Deformation density using DFT method for Asc. - Fe^{3+}

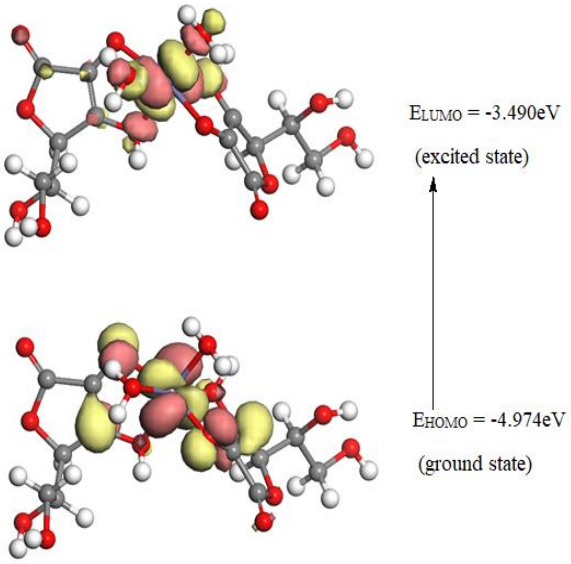


Fig. (7): Total density, Deformation density and 3D plots frontier orbital energies for Asc. - Fe^{3+} complex using DFT method.

5. Conclusion

Ascorbic acid, as a chelating drug, and its complexes with Mn^{2+}, Fe^{3+}, Co^{2+}, Cu^{2+}, and Zn^{2+} have had their proton—ligand ionization constants and metal—ligand stability constants measured potentiometrically in 1.0 M NaCl and ethanol—water (25% ethanol). The complexes that were generated in solution had a stoichiometry of 2:1 (Asc.: M), with Fe^{3+} being more stable than Cu^{2+}, Mn^{2+}, Co^{2+}, and Zn^{2+}. The solid complexes underwent preparation, characterization, and testing for their antibacterial activity against gram positive, gram negative, and fungal microorganisms using various physical and chemical procedures. When compared to ampicillin and ciprofloxacin as standards, there was increased antibacterial and antifungal activity. A set of measurements using the materials studio package's DMOL3 program were applied in order to recognize the wide-scale density function theory (DFT). The present investigation yielded the quantum chemical. Previous studies have established that metal ions play crucial roles in biological systems, influencing enzyme activity and the stability of biomolecules. Your potentiometric analysis of L-ascorbic acid with Mn(II), Fe(III), Co(II), Cu(II), and Zn(II) contributes to a deeper understanding of how these interactions can alter the ionization equilibrium of ascorbic acid. Comparing your ionization constants and stability constants with those reported in earlier studies could highlight any novel findings or discrepancies.parameters and certain energy characteristics of the medication in its free form as well as its

complexes. The synthesis and characterization of solid-state complexes formed between L-ascorbic acid and metal ions allow for the evaluation of their biological activities. Previous literature has shown that metal complexes can exhibit enhanced antibacterial and antifungal properties compared to their free ligands. Utilizing the DMOL3 tool for DFT calculations provides a theoretical framework for understanding the electronic properties of your complexes. The synthesis and characterization of solid-state complexes adds a practical dimension to this study that is often overlooked in literature. By providing empirical data on these complexes, my research bridges the gap between theoretical predictions and real-world applications, enhancing the relevance of my findings. The increased efficacy of metal complexes suggests that incorporating metal ions can enhance the interaction of the compound with microbial cells. This enhanced activity may lead to more effective treatments against resistant strains of bacteria and fungi, which is a growing concern in public health. Understanding how the lipophilic character of metal ions allows for better penetration and interaction with bacterial membranes can inform future designs of antimicrobial agents. This insight could lead to the development of new compounds that specifically exploit these mechanisms to improve their efficacy. If metal complexes prove to be significantly more effective, it may be possible to achieve the desired antimicrobial effects at lower concentrations. This could reduce side effects and minimize the risk of developing resistance, as lower doses may exert less selective pressure on microbial populations. The ability to enhance the activity of known compounds like ascorbic acid through metal coordination could lead to novel formulations that combine the antioxidant properties of ascorbic acid with the antimicrobial properties of metal complexes. This dual action could be particularly useful in food preservation, wound healing, and the development of new disinfectants.

6. Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

7. References

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