Article

a special issue for the scientific conference held by the Department of Chemistry- College of Education for Girls/University of Kufa and in cooperation with Hilla University College, under the title (**5'th Postgraduate Students Annual Conference**) **(PSAC2024)**, which held for Wednesday, **24/4/2024.**

Synthesis, characterization and docking studies of Sliver(I)-N-heterocyclic carbene ligated Schiff base complexes

Mustafa Naji Mohammed Saleh and Mohammed Z. Ghadhyeb,

Department of Chemistry, Faculty of Science, University of Kufa, Najaf, Iraq.

Email: mohammed.alhallaqi@uokufa.edu.iq

Abstract

This study presents the synthesis, characterization, and molecular docking studies of a novel Schiff base functionalized N-heterocyclic carbene. The synthesis involved the coordination of Schiff Base/NHC ligand precursor (1M) with silver(I) ions, yielding Silver(I)- Schiff base /NHC Complex (1N). Characterization of (**1M**) and(**1N**) were performed using various spectroscopic techniques, including NMR, IR and UV-Vis, supplemented by elemental analysis. Molecular docking studies were conducted to explore potential biological activities of the synthesized complex. The results demonstrate successful synthesis of the Schiff base functionalized NHC complex of silver(I) and provide valuable insights into its structural properties and potential biological applications.

Keywords: Ag(I)-NHC, Carbene, S, Heterocyclic, Imidazolium salts**,** Schiff Base,

1.Introduction

NHC-silver complexes exhibit anticancer properties against various cancer cell lines in the realm of organometallic chemistry. The initial discovery of the anticancer potential of Ag(I)-NHC complexes, particularly those derived from 4,5-dichlorimidazole, was reported by Young and colleagues. Their research showcased the effectiveness of these complexes against OVCAR-3 (ovarian), MB157 (breast), and hila (cervix) cancer cell lines [1]. Subsequently, Takei et al. synthesized cyanobenzyl NHC silver complexes and investigated their anticancer activity against human kidney cancer cells (Caki-1)[2-4]. Tacke and collaborators also explored the potential of benzyl NHCs as antitumor agents against Caki-1 and MCF-7 cancer cell lines[4-6] The intriguing combination of anticancer and antimicrobial properties in NHCs prompted an investigation into the mechanism of action of silver(I)-NHCs, which was previously shrouded in mystery [7]. Additionally, there has been a shift in the approach to anticancer drug design, with a focus on developing molecules that can target multiple pathways, potentially enhancing effectiveness and reducing toxicity [8, 7] .

Furthermore, against human cancer cells, Schiff ligands and their metal complexes have potent anticancer action [9,10] Schiff-bases are potentially possible anticancer medications because of their capacity to break DNA and insert between DNA base pairs, which gives them their anticancer action [11]. Furthermore, one of the most popular ligand classes in metal coordination chemistry is Schiff-base, which offers adaptable and flexible ligands that may mix different metals to create complexes with shared structures and characteristics [12].Consequently, it is anticipated that Schiff-bases will be employed as a lead ligand in the logical design of new metal complexes with anticancer properties [13-17].

Survivin is a member of the inhibitor of apoptosis protein (IAP) family. This protein adopts a unique dimer structure. In addition to its function as an inhibitor

of apoptosis, survivin is a chromosomal passenger protein that mediates the spindle assembly checkpoint and cytokinesis. In addition to normal proliferating cells, Survivin is also overexpressed in cancer cells where it also likely acts as an inhibitor of apoptosis. As such, Survivin has been proposed as an attractive target for anti-cancer therapeutics[18_20].

In order to create compounds that combine the potency of these two important classes (1a,1b) and test their anticancer efficacy through molecular docking with the active sites of survivin dimer H. sapiens protein (PDB ID: 1E31).

2.Synthesis part

2.1 General information

All chemicals and solvents, were of the highest analytical grade and used as supplied from commercial sources. All reactions were performed using Schleck techniques under an inert atmosphere. Glassware was dried in an oven at 120 °C. Nuclear magnetic resonance (NMR) spectra were recorded using Bruker 400 MHz spectrometers at ambient temperature. 'H NMR peaks are labeled as singlet (s), doublet (d), triplet (t) and multiplet (m), chemical shifts were referenced with respect to solvent signals. The high-resolution mass spectra. The infrared spectra were recorded with FT-IR spectrophotometer (FTIR- 8400s, Bruker). UV-Visible spectrometer double beam was assigned on (Shimadzu UV 1650 PC) and the elemental analysis (CHN) were carried out on (PerkinElmer series II, 2400 microanalyzer). Thin Layer Chromatography (TLC) was carried out on Machery-Nagel polygamist/G/UV254 pre-coated plates. Melting points analysis were recorded using an Electrothermal 9100 melting point apparatus.

2.2 Synthesis of the Schiff Base/NHC ligand precursor 1M

Into a dry Schleck flask with a stirrer bar and nitrogen gas was placed an equimolar amount of both the **Schiff base a** (2.53 g, 1 mmol) and the **1**-methyl imidazole b (0.82 g, 1 mmol). The mixture was refluxed at 80 ^{o}C for 24 hours, with the progress of reaction monitored using TLC. After the completion of

reaction, the solvent was evaporated then recrystallized using methanol, 0.8g (75% yield) as a brown powder (m.p = 286-288 °c). **IR (ATR cm-1)**:3395(NH),3102 (CH, sp^3), 1618 (C=N), 1510, (C=C): **HRMS (ESI)**: [M+ - Br-] calcd for $C_{15}H_{19}BrN_4$, 338.04; found, 337.82. 1H NMR (301 MHz, DMSO) δ 12.16, 9.95, 8.31, 8.12, 8.10, 8.02, 7.54, 7.52, 7.28, 7.28, 7.26, 7.25, 7.23, 3.89, 3.42, 3.38, 3.23, 2.53, 2.52. **13C NMR** (76 MHz, DMSO) δ 154.27, 137.49, 129.14, 127.82, 121.65, 63.12, 50.04, 44.29. **Anal. Calcd** for $C_{15}H_{19}BrN_4$: C, 53.74; H, 5.71; N, 16.71. Found: 53.76; H, 5.08; N, 13.08.

2.1.2. Synthesis of Silver(I)- Schiff base /NHC Complex 1N.

Silver oxide (1.15g, 0.005 mol) was added to a solution of compound **1M** (0.253, 0.01 mol) in 20 mL methanol. The mixture was stirred for 24 hr. in glassware, covered by aluminum foil. After the black suspension was filtered through the celite to remove the excess Ag_2O, the solvent was removed to give the product as a white solid 0.8 g (61 % yield) (m.p = 226-228 °c). **IR (ATR cm-1)**:3393(NH) ,3085 (CH, sp^3), 1660,1616 (C=N), 1509, (C=C): **HRMS (ESI)**: [M+ - Br-] calcd for $C_{15}H_{17}AgBrN_4$, 438.97; found, 441.21. 1H NMR (301 MHz, DMSO) δ 11.58, 8.44, 8.41, 8.38, 8.03, 7.67, 7.64, 7.38, 7.21, 7.11, 7.04, 6.71, 6.68, 6.37, 4.47, 4.12, 3.98, 3.88, 3.64, 3.41, 3.41, 3.21, 3.01, 2.85, 2.59, 2.57, 2.55, 2.54, 1.24, 1.15, 1.06. **13C NMR** (76 MHz, DMSO) δ 191.03, 163.89, 154.27, 134.31, 111.70, 104.93, 96.40, 60.63, 48.18, 44.60, 12.91.**Anal. Calcd** for $C_{14}H_{15}Br_2N_3OAg$: C, 33.13; H, 2.98; N, 8.28. Found: C, 33.16; H, 3.01; N, 8.27.

2.3 Theoretical approach

The molecular indices produced from the compounds provided allowed for accurate predictions regarding their chemical and biological properties. To model the ligand-protein receptor binding for the complexes, the MOE-2015 software package was utilized. In this study, the survivin dimer H. sapiens protein, corresponding to the PDB ID: 1E31, was selected as the protein receptor, with data obtained from the Protein Data Bank [21]. Prior to docking, several essential steps were undertaken, including the removal of solvent molecules, addition of

hydrogen, fixing the protein chain, and identification of active sites. The docking of the tested compounds was optimized using Merck Molecular Force Field calculations, which involved minimizing energy, adjusting potential energy, calculating atomic charge, and determining binding energy [22].

Results and discussion

3. Synthesis and characterization

3.1 NMR study

The ^{1}H NMR for **1** in d_6-DMSO showed the singlet peak at (12.16) ppm was assigned to amine proton (NH). The signal of imidazolium proton H2′ appeared at (9.95) ppm as a singlet peak. The singlet peak at (8.31) ppm was assigned to Schiff base proton (HC=N). In addition, aromatic protons were seen as a multiplet at around 7.23 to 8.12 ppm due to the deshielding effects of the positively charged imidazolium ring system. The -CH_2 proton peaks bonding to the (-N-CH_2-CH_2-N-) has been observed as triplets at (3.87,3.23) ppm respectively. Figure (3.1).

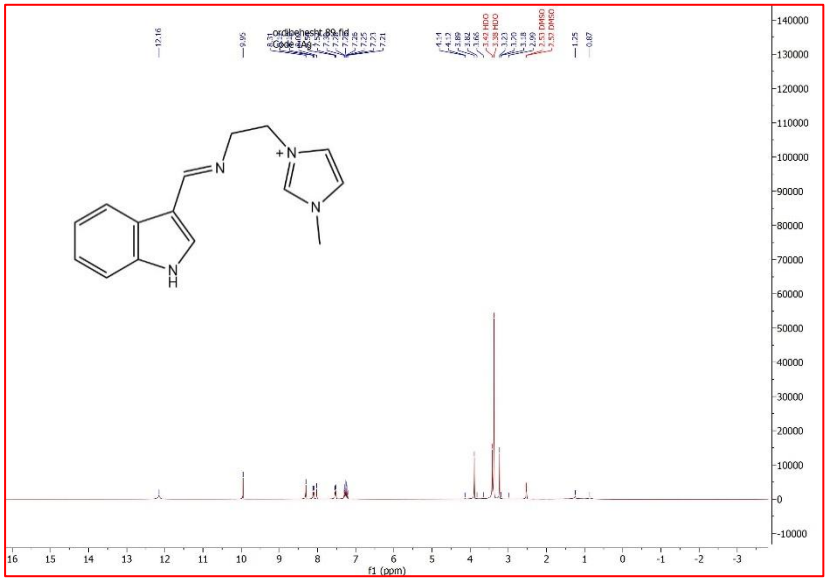


Figure 3.1: 1H NMR Spectrum of 1**M** in d_6-DMSO

The ^{1}H NMR for **1M** in d_6-DMSO showed the singlet peaks at (9.9) ppm were assigned to aldehyde proton (NH). The singlet peak at (8.3) ppm was assigned to Schiff base proton (HC=N). In addition, aromatic protons were seen as a multiplet at around 7.23 to 8.12 ppm. The -CH_2 proton peaks bonding to the (-N-CH_2-CH_2-N-) has been observed as triplets at (3.87, 3.23) ppm respectively. Figure (3.2).

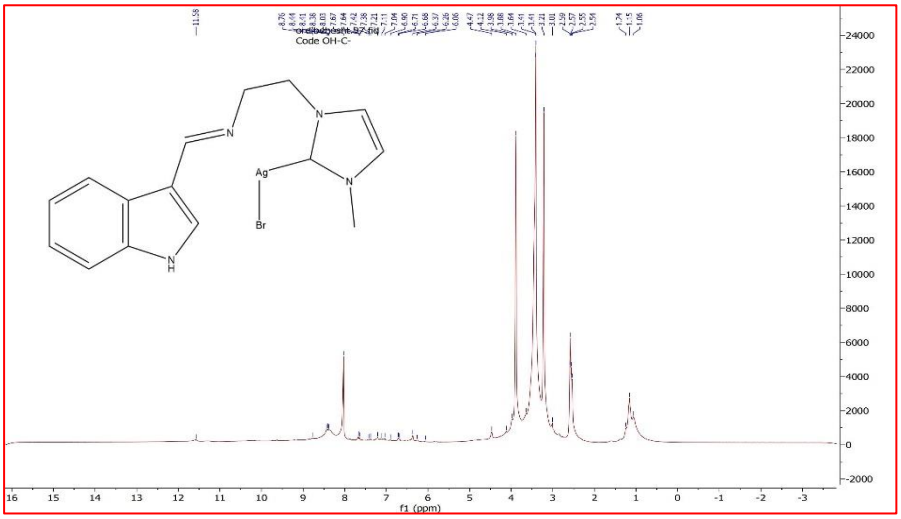


Figure 3.2: 1H NMR Spectrum of 1**N** in d_6-DMSO

The successful coordination of Ag-NHC in 1**s** complex via deprotonation of 1N is the absence of characteristic singlet protons (9.95) ppm compared with (**1M**) ligand spectra[23] Figures (3.2) .

In addition, the ^{13}C NMR spectrum of 1s showed the characteristic imidazolium 1M signal at 137.49 ppm. The carbon alpha to imine (N=C) appears at 154.27 ppm while Figures (3.3)

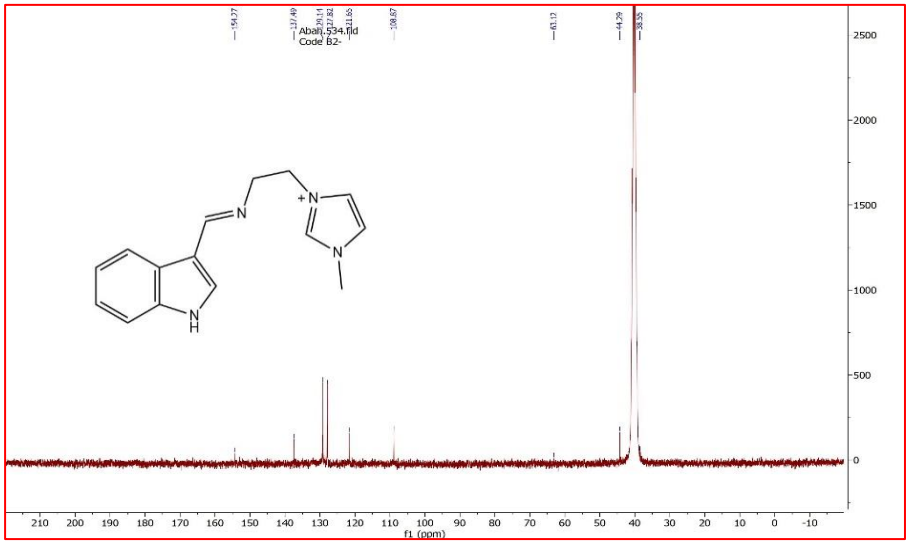


Figure 3.3: ^{13}C NMR Spectrum of 1**M** in d_6-DMSO

The ^{13}C NMR spectra of **1N** showed that C-Ag signal at (191.03) ppm which did not appear compared to the ligand **1M** spectra, indicating the formation of an Ag-C bond. Figures (3.4).

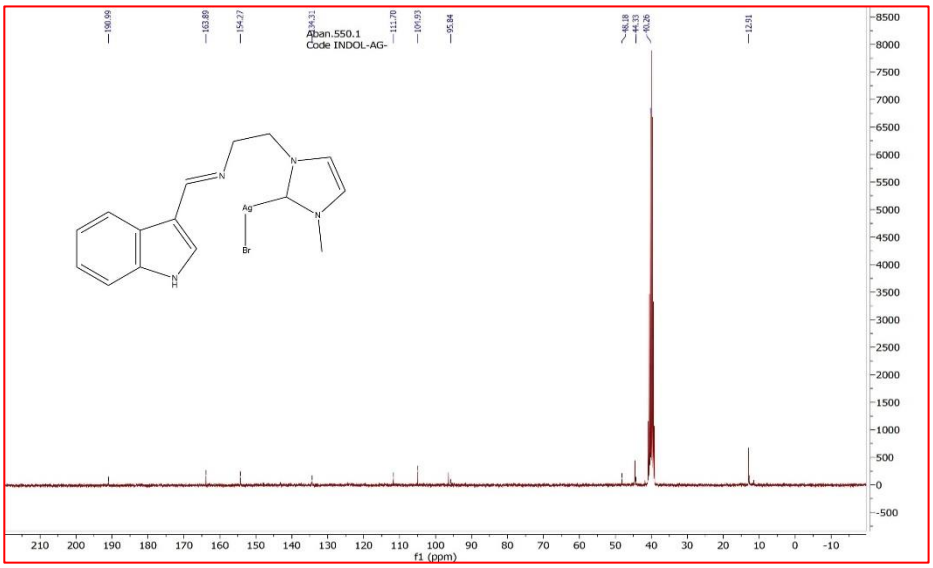


Figure 3.4: ^{13}C NMR Spectrum of 1**N** in d_6-DMS

3.2 UV-Vis Study

The spectra of ligand were recorded in methanol. The **1M** spectrum showed two distinguishable bands at (**275 and 314**) nm these bands can be indicating to π-π* and n-π* for the benzimidazole ring.

In addition to the absorption peaks due to electronic transitions in ligands(241,260), the novel Ag-NHC complex **1N** showed a new absorption peak at (**297**) nm, related to MLCT (metal-ligand charge transfer)

This complex couldn't show **297**any d-d transition due to its d^{10} configuration because of d- d transitions are forbidden by the Laporte selection rule, which confirms the absence of any (d-d) transitions and absence of visible region absorptions in their electronic spectra. The difference in absorption peaks and the appearance of new absorption peaks confirm the complexity process figures (3.5)

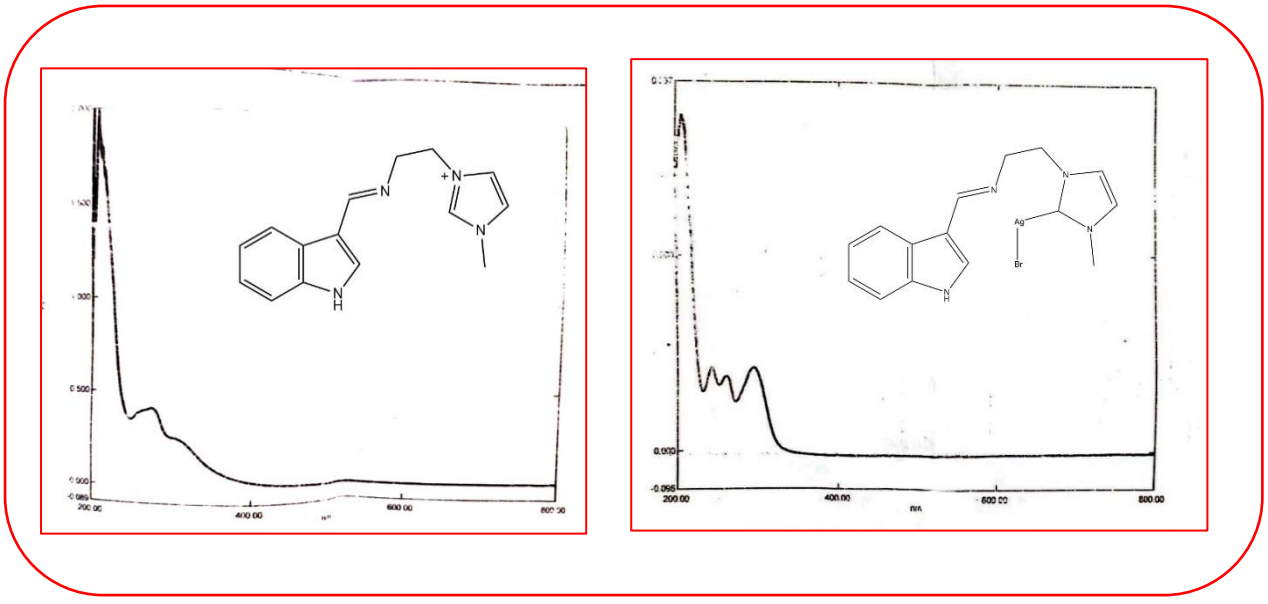


Figure 3.5: UV-Vis Spectrum of 1**M** and 1**N**

3.3 FT-IR Study

The FT-IR spectrum of ligand **1M**, was gave the following peaks which was assigned as following, 1618 cm^{-1} due to (HC=N) group. Figure (3.6).

The FTIR spectra of complex **1N,** Figure (3.7) showed the following characteristic peaks; the peak 1616 cm^{-1} for the (HC=N). The slight shifting at the (HC=N) peak may be evidence of the lack of coordination bond with the silver ion. Figure (3.7).

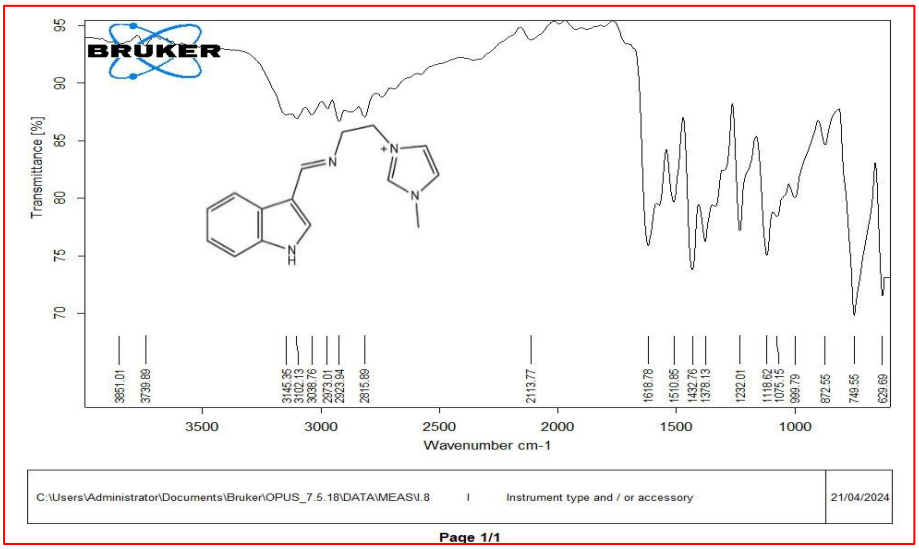


Figure (3. 6) FT-IR of compound 1M

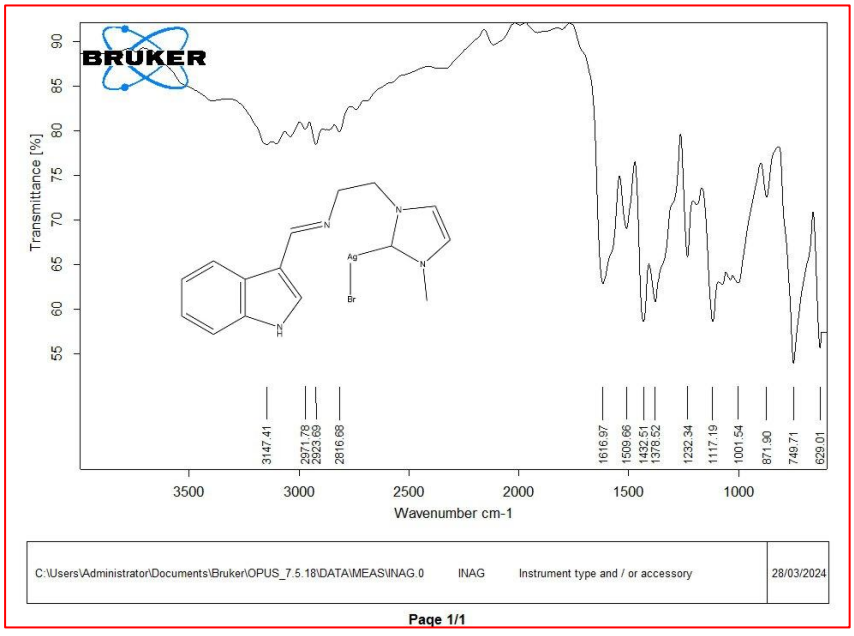


Figure (3.7) FT-IR of compound 1N

3.16. Molecular docking studies

Molecular docking is a valuable tool in structure design that enables the prediction of binding affinities and conformations between ligands and target receptor proteins. This knowledge greatly speeds up the development of medications [24,25]. In order to explore the binding mechanisms, molecular docking experiments were conducted on compounds within the active sites of survivin dimer H. sapiens protein receptors (PDB ID: 1E31) to assess their binding interactions.

3.16.1. Molecular docking against 1E31

Docking the molecules with 1E31 brought them into contact with each other, facilitating interaction . This interaction provides valuable insights, as illustrated in Figure 1. The Ag(I) metal complex showed a calculated score of (S = -5.1989 kcal/mol) with RMSD = 1.5074 Å. Modifying the Ag(I) complex led to an improved binding affinity of the free ligand. In terms of coordination patterns, the free ligand formed H-acceptor and pi-H bonds with specific amino acid residues, namely O 15 with ARG 18 (A), 6-ring with LYS 15 (A), and 6-ring with PHE 93 (A), with bond distances ranging from 3.03 to 4.47 Å. The Ag(I) complex, on the other hand, formed H-donor and two H-acceptor bonds in the 1E31 protein via Br 14, O 1, and O 2 atoms with ASN 111 (A), ALA 85 (A), and ASN 111 (A) residues, respectively, with bond distances ranging from 3.08 to 3.48 Å. Molecular docking serves as a valuable tool in structure design to predict the binding affinities and conformations between ligands and target receptor proteins, thereby accelerating medication development (24,25). Docking experiments were specifically conducted on compounds located in the active sites of survivin dimer H. sapiens protein (PDB ID: 1E31) receptors to gain insights into their binding mechanisms.

S (Kcal/mol)	RMSD (Å)
-5.1989	1.5074

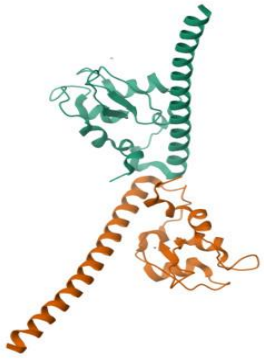
Ligand Interactions Report

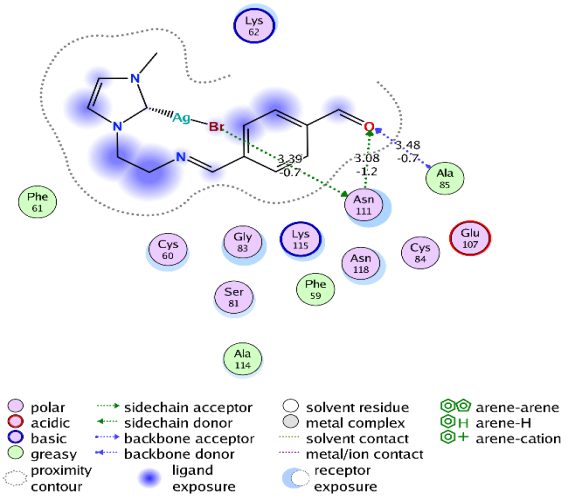
Tue Feb 06 02:07:02 2024 (MOE 2015.10)

1E31: APOPTOSIS INHIBITOR / 1E31

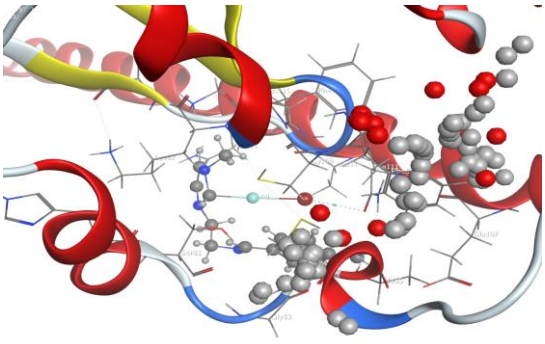
Ligand	Receptor			Interaction	Distance	E (kcal/mol)
BR14 23	OD1 ASN	111	(A)	H-donor	3.39	-0.7
O1 1	N ALA	85	(A)	H-acceptor	3.48	-0.7
O1 1	ND2 ASN	111	(A)	H-acceptor	3.08	-1.2

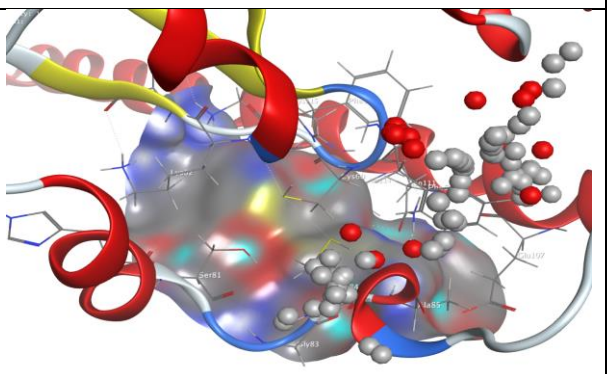
Ligand Interactions





Protein ID (1E31) | **2D**





3D | **Map**

Figure 1. Detailed analysis of the interactions between protein 1E31 and Ag-Complex in 2D, 3D, and map views, (S = the final score is the score of the last step, RMSD_refine = the mean square deviation between the laying before refinement and after refinement pose).

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