Supplementary docking results

As for compound **7**, the molecular docking results indicate that **7** engages in diverse types of interactions, including hydrogen bonding, metal-acceptor interactions, electrostatics, and various hydrophobic contacts with specific amino acid residues. Conventional hydrogen bonds with amino acid residues Asp224 (OD2), Glu223 (OE1), and Asp363, characterized by short distances and strong binding affinities, are crucial for ligand recognition and orientation within the urease binding pocket. The hydroxyl at *para* position can form a hydrogen bond interaction with the side chain (OD2) of Asp363 (1.89 Å). The hydroxyl group on the benzimidazole moiety can form a hydrogen bonding with the side chain (OE1) of Glu223 (1.97 Å) while its amino group forms another hydrogen bonding with the side chain (OD2) of Asp224 (2.44 Å). In addition to the hydrogen bonds, compound **7** establishes carbon hydrogen bonds with the sidechain (NE2) of His323 and backbone (O) of Glu223, further enhancing its stability and specificity for the binding site. Moreover, compound **7** can possibly participate in metal-acceptor interactions with metal ions Ni798 and Ni799 within the binding site. These interactions are characterized by short distances, potentially influencing the ligand's binding orientation and selectivity, offering promising avenues for developing metal-binding ligands with specific biological activities. Electrostatic π-anion interaction observed between compound **7** and the side chain (OD2) of Asp224 (4.74 Å) further strengthens ligand binding within the active site. Additionally, hydrophobic interactions, including π-π stacking with His323 and π-π T-shaped interactions with His249 and His324, can possibly contribute to compound **7** stabilization in hydrophobic regions of the binding site. Furthermore, an amide-π stacked interaction involving Lys169, CYS322, and His323 observed could also further enhance the stability of compound **7** within urease active site.

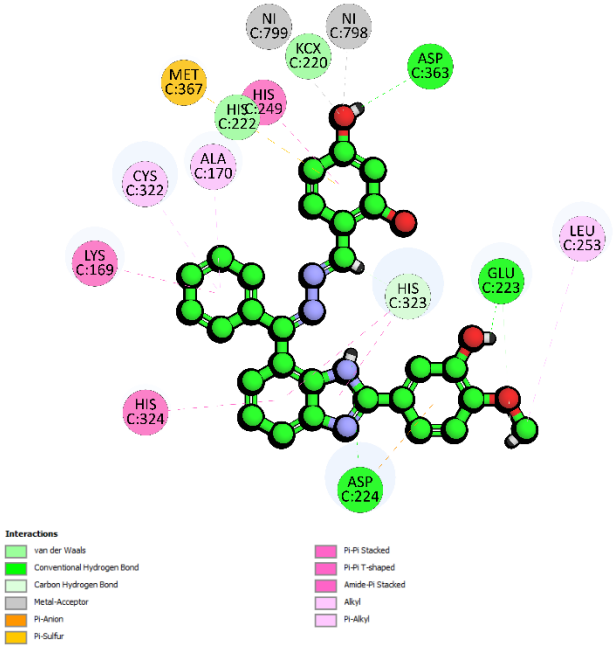


Figure 2. The 2D-interaction diagram of compound **7** interacting with active site of urease enzyme.

Compound **9** engages in multiple hydrogen bond interactions with crucial residues in the urease enzyme. Notably, a stable hydrogen bond is formed with GLU223 (OE1) at a distance of 2.89 Å, likely playing a significant role in ligand specificity and recognition within the active site (**Figure 3**). Compound **9**, through its hydroxyl group at *meta* position was observed to form conventional hydrogen bonds with the side chain (HD1) of His249 (2.98 Å), the backbone (O) of Ala170 (2.11 Å), and the side chain (NE2) of His222 (2.15 Å). On the other hand, the amino group of benzimidazole forms hydrogen bonding with side chain (OD2) of ASP224 (2.61 Å), further enhancing the stability and binding orientation of compound **9**. In addition to hydrogen bonds, compound **9** establishes carbon hydrogen bond interactions with the side chain (OD2) of Asp224 (2.78 Å) and side chain (HD2) of Glu223 (2.53 Å). The docking study also reveals possibility of metal-binding interactions between compound **9** and metal ions Ni in the urease enzyme. A metal-acceptor interaction is observed with NI798 at 2.31 Å and NI799 (OE2) at 3.10 Å, suggesting intriguing possibilities for metal-binding ligands with specific biological activities. Beyond hydrogen and metal interactions, compound **9** engages in other types of interactions within the active site. An electrostatic π-anion interaction is observed with ASP224 (O) at 4.65 Å, contributing to compound **9** stabilization within the

active site. Moreover, hydrophobic interactions, including π-sulfur with Met367 (NE3) at 5.03 Å, π-π T-shaped with His324 (OD3) at 5.18 Å, amide-π stacked with Lys169 (HD3) and Ala170 (HD3) at 4.42 Å, and π-alkyl with Lys169 (OD4) at 5.21 Å, Ala170 (OE3) at 4.89 Å, and Ala170 (O) at 5.34 Å, significantly influence the compound's overall binding mode and orientation within the binding pocket.

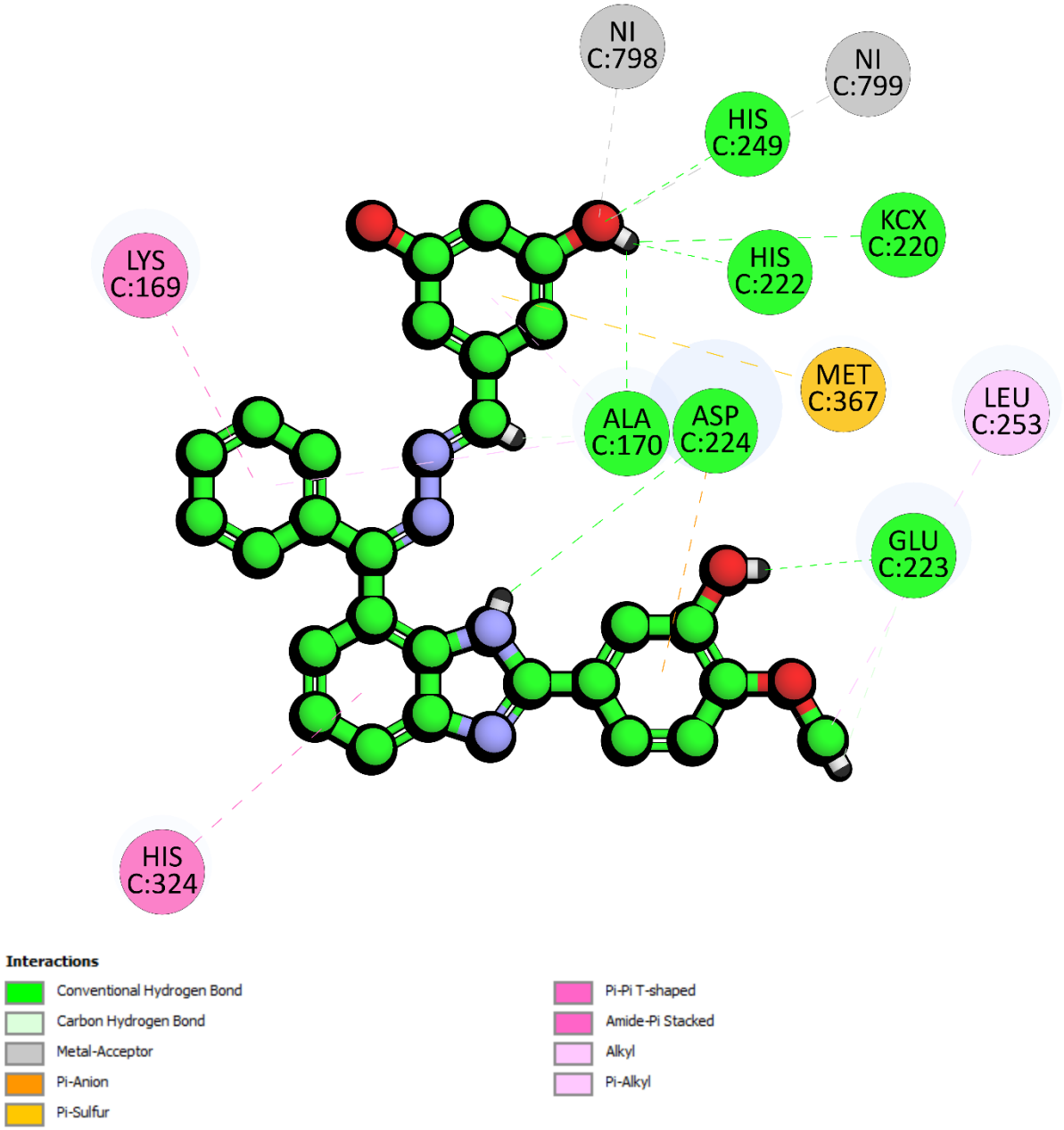


Figure 3. The 2D-interaction diagram of compound **9** interacting with active site of urease enzyme.

The molecular docking study of compound **12** with the urease enzyme has provided a comprehensive understanding of the various interactions occurring within the protein's active

site. Each interaction type plays a critical role in stabilizing the ligand and influencing its binding affinity and specificity for the target enzyme (**Figure 4**). Compound **12** establishes multiple hydrogen bond interactions with key residues. Notably, it forms a conventional hydrogen bond with Lys169 (HZ2) at a distance of 2.03 Å, enhancing the ligand's stability within the active site and contributing to ligand-protein recognition. Additionally, hydrogen bond interactions with Arg339 (HH11, HH22) at distances of 2.30 Å and 2.16 Å, respectively, involve halogen (fluorine) atoms, adding complexity to the binding mode and potentially influencing the ligand's conformational preferences. Furthermore, compound **12** forms hydrogen bond interactions with Cys322 (HZ3) at a distance of 2.58 Å and with His249 (HH33) as a carbon hydrogen bond at a distance of 2.46 Å. These interactions further stabilize the ligand within the active site and contribute to its binding orientation. In addition to hydrogen bonds, halogen interactions are observed. Compound **12** forms halogen interactions with His323 (HH44) at a distance of 3.48 Å and with Arg339 (HZ4) at a distance of 3.77 Å. These interactions may influence the ligand's orientation and conformational preferences within the binding pocket. Moreover, a notable electrostatic interaction in the form of a π-anion bond is observed between compound **12** and Asp224 (HH55) at a distance of 3.38 Å, stabilizing the ligand within the active site. Compound **12** also establishes a unique π-sulfur interaction with Cys322 (HH66) at a distance of 4.81 Å. This specific interaction may have implications for the ligand's binding mode and conformational preferences, potentially influencing its biological activity. Furthermore, several hydrophobic interactions are crucial for determining the ligand's binding mode within the hydrophobic pocket of the binding site. Compound **12** forms π-alkyl interactions with Ala170 (HZ5, HH77, HZ6), Lys169 (HH88), and Ala366 (HH99) at distances ranging from 4.28 Å to 4.90 Å, enhancing its stability and binding affinity.

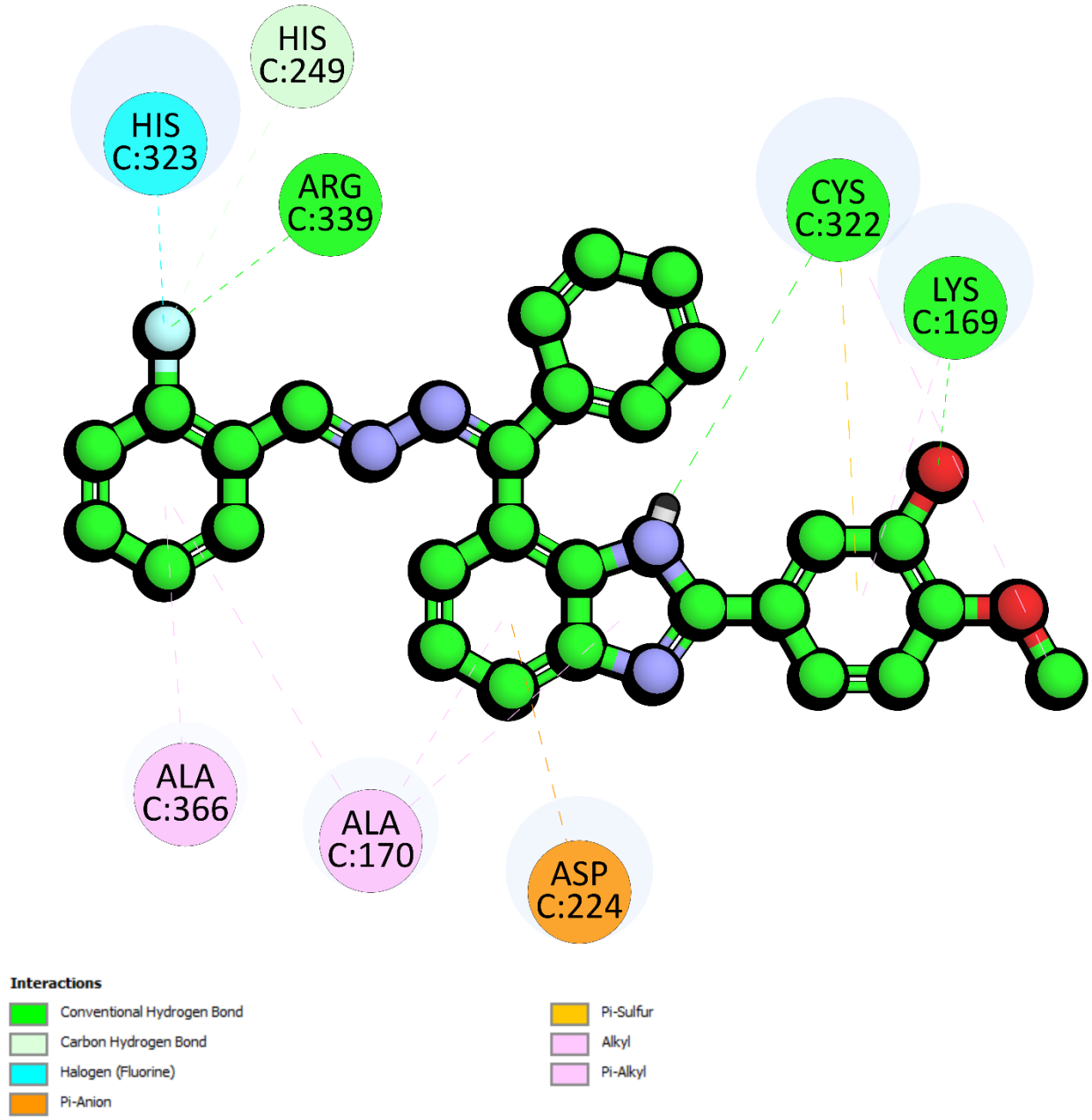
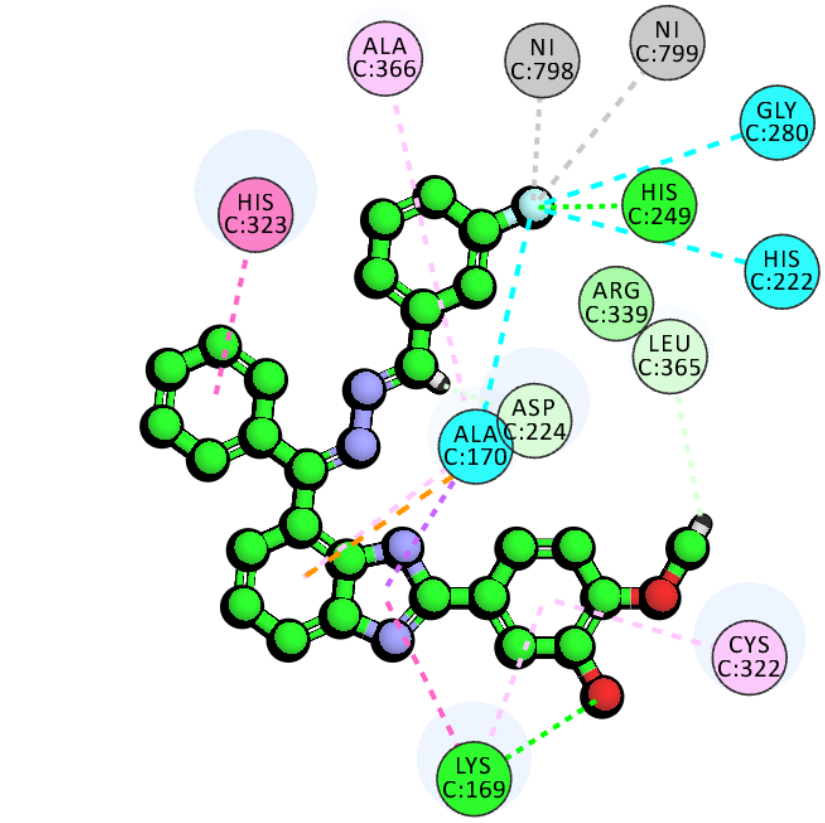


Figure 4. The 2D-interaction diagram of compound **12** interacting with active site of urease enzyme.

The molecular docking study of compound **13** with the urease enzyme reveals a variety of interactions, each playing a significant role in understanding the ligand's potential binding mode and affinity within the protein's active site (**Figure 5**). Firstly, compound **13** forms a conventional hydrogen bond with the residue Lys169 (HZ2) at a distance of 2.21 Å. This interaction is crucial for stabilizing the ligand within the active site and may contribute to ligand-protein recognition and specificity. In addition, another important hydrogen bond is observed between **13** and the residue His249 (HH11) at a distance of 2.34 Å. This hydrogen

bond involves a halogen (fluorine) atom, adding complexity to the binding mode and potentially influencing the ligand's conformational preferences. Furthermore, compound **13** establishes a hydrogen bond with the residue Asp224 (OD1) at a distance of 3.18 Å, further contributing to the ligand's stability within the active site. Moreover, compound **13** forms a hydrogen bond with the residue Asp224 (HZ3) at a distance of 2.59 Å, and this likely enhances the ligand's binding affinity and fine-tunes its binding orientation within the active site. Additionally, a carbon hydrogen bond interaction is observed between **13** and the residue Leu365 (HD2) at a distance of 2.51 Å. This interaction represents a less common type of hydrogen bond and may play a role in determining the ligand's conformational preferences and binding mode. Compound **13** also establishes unique interactions involving halogen (fluorine) atoms and metal acceptors with the residues Ni798 (OD2) and Ni799 (HZ4) at distances of 3.09 Å and 3.48 Å, respectively. These specific interactions may have implications for the ligand's binding mode and its potential interactions with metal ions in the active site. Furthermore, compound **13** forms halogen (fluorine) interactions with the residues Ala170 (HD3), His222 (OD3), and Gly280 (HZ5) at distances of 3.28 Å, 3.20 Å, and 3.38 Å, respectively. These halogen interactions may contribute to the ligand's orientation within the binding pocket and influence its binding affinity. A notable electrostatic interaction in the form of a π-anion bond is observed between compound **13** and Asp224 (HD4) at a distance of 3.33 Å. This interaction likely enhances the ligand's stabilization within the active site. Compound **13** also exhibits hydrophobic interactions, including a π-sigma interaction with Ala170 (OD4) at a distance of 2.75 Å and a π-π stacked interaction with His323 (HZ6) at a distance of 4.05 Å. These interactions contribute to the ligand's stability within the active site. Moreover, **13** forms an amide-π stacked interaction with both Lys169 and Ala170 (HD5) at a distance of 5.09 Å. This interaction involving multiple residues may play a role in enhancing the ligand's binding affinity and specificity. Additionally, compound **13** establishes hydrophobic π-alkyl interactions with Ala170 (OD5) at a distance of 3.93 Å and with Lys169 (HZ7) at a distance of 4.16 Å. These interactions further contribute to the ligand's stability and binding affinity within the hydrophobic region of the active site, while a hydrophobic π-alkyl interaction with Cys322 (HD6) at a distance of 5.24 Å seems to add to the ligand's overall stability and potential conformational preferences. Lastly, compound **13** establishes another hydrophobic π-alkyl interaction with Ala170 (OD6) at a distance of 5.09 Å. This interaction further enhances the ligand's stability within the binding site.



Interactions

- van der Waals
- Conventional Hydrogen Bond
- Carbon Hydrogen Bond
- Metal-Acceptor
- Halogen (Fluorine)
- Pi-Anion
- Pi-Sigma
- Pi-Pi Stacked
- Amide-Pi Stacked
- Pi-Alkyl

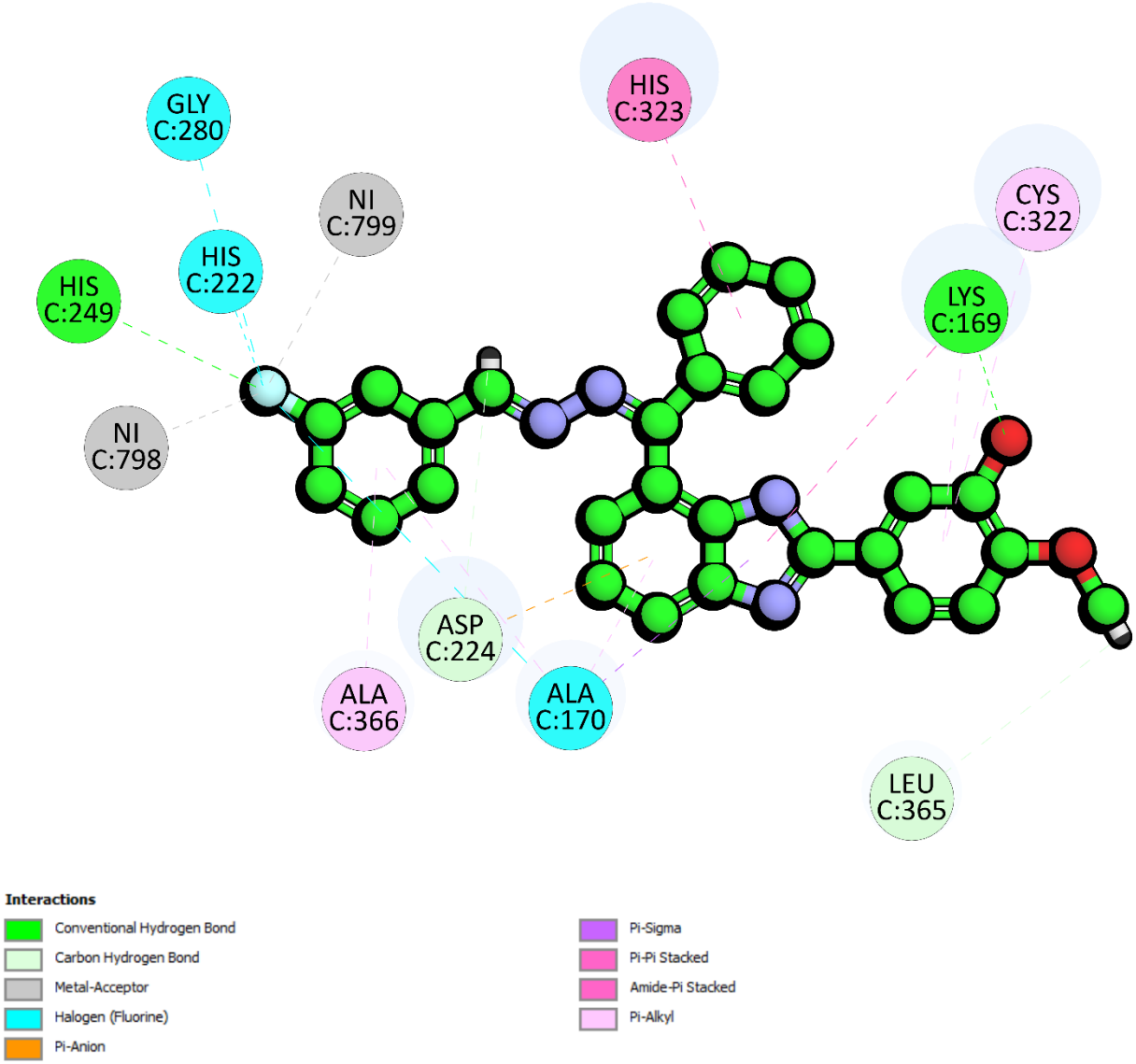


Figure 5. The 2D-interaction diagram of compound **13** interacting with active site of urease enzyme.