# Mechanistic Insight Antimicrobial Potential of Plant Phenolic: Grid Based Docking Approach

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## Background

**Objective:** Antibiotic resistance among bacteria is currently on a phenomenal rise, and this poses a real threat to global health. Particularly concerning are contaminations caused by methicillin-safe Penicillin-safe Staphylococcus aureus (MRSA) Vancomycin-safe strain of Streptococcus pneumoniae Enterococcus and Mycobacterium tuberculosis because significant populations of these organisms are resistant to some kinds of established anti-microbials. The search for novel antibacterial drugs that inhibit essential bacterial targets and are unaffected by mechanisms of current chemotherapeutic agent resistance is being driven by this circumstance. The aminoacyl-tRNA synthetase (AaRS) enzymes have been the focus of recent study for the development of antibacterial drugs in this area. In this project, natural plant phenolics and flavonoids known as rosmarinic acid, chlorogenic acid, rutin, and quercetin have been identified as ligands, and their inhibitory activity against the aminoacyl-tRNA synthetase (AaRS) enzyme has been tested in-silico with the aid of docking technique.

**Methods:** In the current study, a molecular docking technique was used to try and identify aminoacyl-tRNA synthetase (AaRS) enzyme inhibitors. The binding using Auto Dock software has been determined using a grid-based docking approach. Compounds' 2D structures were constructed, transformed to 3D, and then energetically reduced using Merck Molecular Force Field up to an rms gradient of 0.01. (MMFF).

**Results:** The molecular docking result revealed that chlorogenic acid, rosmarinic acid, quercertin and rutin showed encouraging docking score. The docking score found to be -4.18, 4.74, -6.89 &-5.28 kcal $mol^{-1}$ respectively. $mol^{-1}$.

**Conclusion:** The interaction of ligand hits to targeted site and docking score finding it can be predicted that phenolics and flavonoids found in the plants extract exhibited good inhibitor of IleRS enzyme.

## Introduction

Because it can predict the conformation of small-molecule ligands within the appropriate target binding site with a high degree of precision, molecular docking is one of the most frequently utilised methods in SBDD. Molecular docking became a crucial tool in drug discovery in the 1980s after the creation of the first algorithms. Additionally, molecular docking algorithms provide out quantitative estimates of the energetics of binding, presenting rankings of docked molecules based on the affinities of ligand-receptor complexes. These tasks are carried out using molecular docking programmes in a cyclical manner, with the ligand conformation being assessed by particular scoring functions. Recursively, this procedure is carried out up until it converges on a minimum energy solution (1). Today's dramatic increase in the prevalence of antibiotic resistance among bacteria poses a real threat to global health. Specifically, contaminations caused by methicillin-safe Penicillin-safe MRSA Staphylococcus aureus Safe for vancomycin in streptococcus pneumoniae Since many of these organisms are resistant to a few kinds of established anti-microbials, Enterococcus and Mycobacterium tuberculosis are the two most common ones. This circumstance is motivating researchers to search for innovative antibacterial drugs that inhibit crucial bacterial targets and are unaffected by mechanisms of resistance to chemotherapeutic medicines now in use. In this context, current studies for the development of antibacterial drugs have focused on the aminoacyl-tRNA synthetase (AaRS) enzymes. By catalysing the creation of aminoacyl-tRNAs, these enzymes serve crucial roles in protein biosynthesis (2). With this endeavor, natural plant Phenolics & flavonoids found in extracts of the plants known as, rosmarinic acid , chlorogenic acid rutin and quercetin has been identified as ligand and their aminoacyl-tRNA synthetase (AaRS) enzymes inhibitory activity has been checked *in-silico* with the help of docking approach.

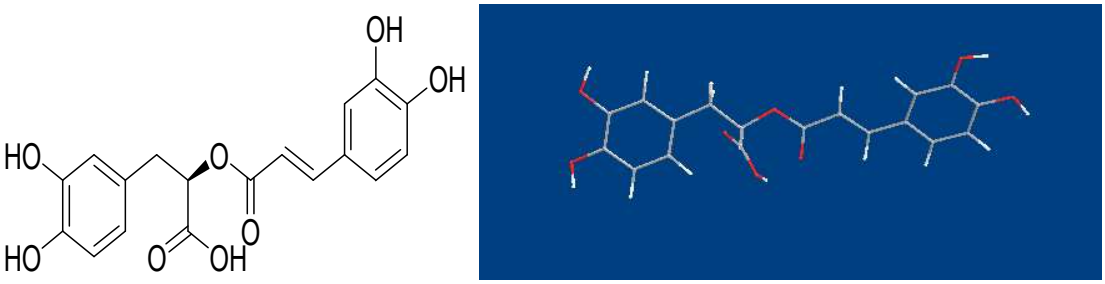
## Experimental works

### Molecular docking studies

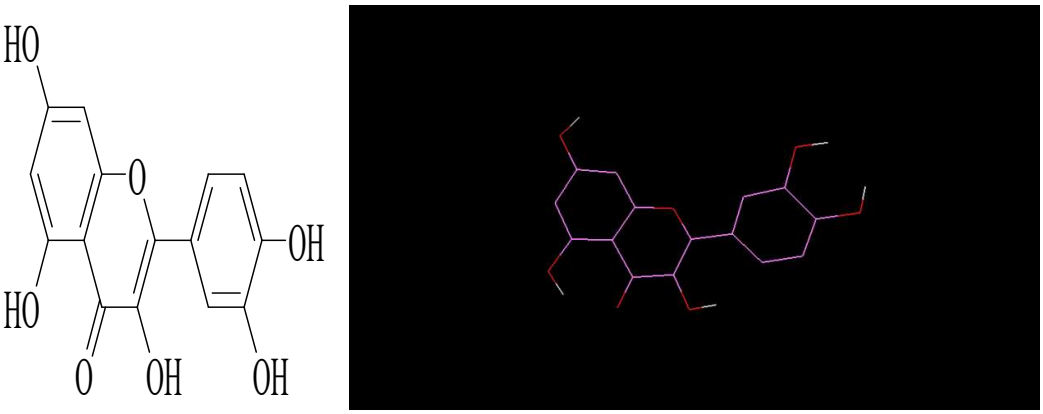
#### *Ligand Preparation:*

2D Structure of ligand (chlorogenic acid) was drawn using Chem Sketch (3), the two-dimensional structure of was converted into 3-D structure and optimized with 3D geometry. The optimized structure was saved in PDB format for AutoDock compatibility. The basic structure of ligand (chlorogenic acid) is given below:

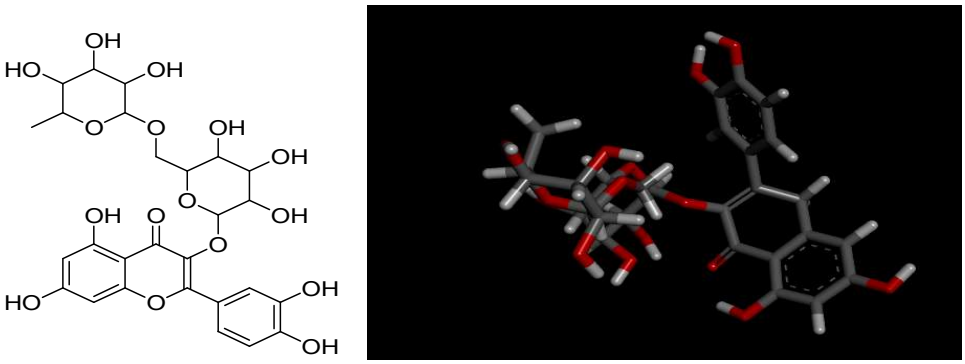
2D and 3D structure of chlorogenic acid



2D & 3D structure of rosmarinic acid



2D & 3D structure of quercetin



2D & 3D structure of rutin

**Figure 1: 2D and 3D conformer of chlorogenic acid**

***Preparation of the grid file***

The regions of interest used by Autodock were defined by considering grid area by making a grid box around the active sites. Grid box plays a central role in process of docking as it is made to cover all the amino acids present in active sites necessary for binding other than those present in receptor. Grid box has 3 thumbwheel widgets which let us change the number of points in the x, y and z dimensions. The spacing between grid points can be adjusted with another thumbwheel, the value in the study taken is 0.503 Å and No. of points considered are 52, 50 and 50 points in the x, y, and z dimensions and 29.845, 79.614 and 80.43 as x, y, z centers (4).

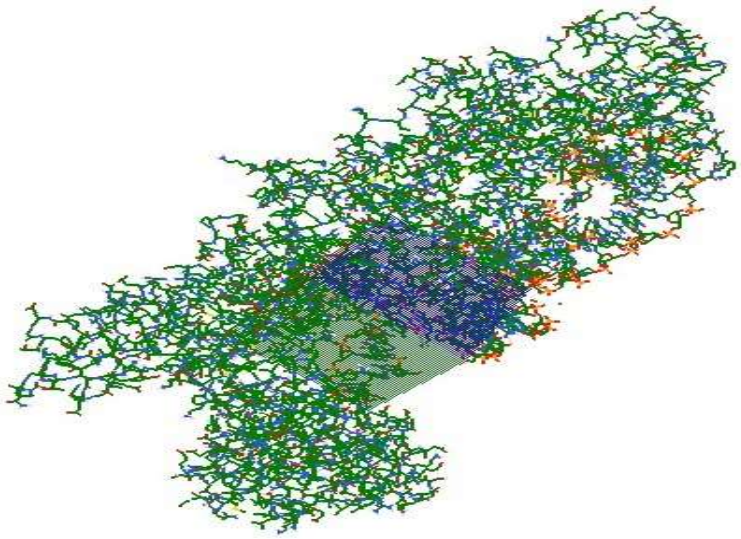


**Figure 2 : Grid box covering all active sites in receptor**

***Preparation of the docking file***

All the calculations were carried out by using Autodock4.2 as docking tool. The visualization and other programs necessary for docking studies were performed out by means of Pymol, Chimera, DS visualizer, MMP Plus (5).

**Docking of Isoleucyl-transfer RNA (tRNA) synthetase (IleRS) with Chlorogenic Acid**

***Crystal structure***

The crystal structure of the protein consisting of receptor associated with bound ligand is downloaded from the Protein Data Bank portal. All the primary information regarding receptor

and structure (1FFY.pdb) registered in the Protein data bank was used. The bound ligand mupirocin is found within the receptor (6).

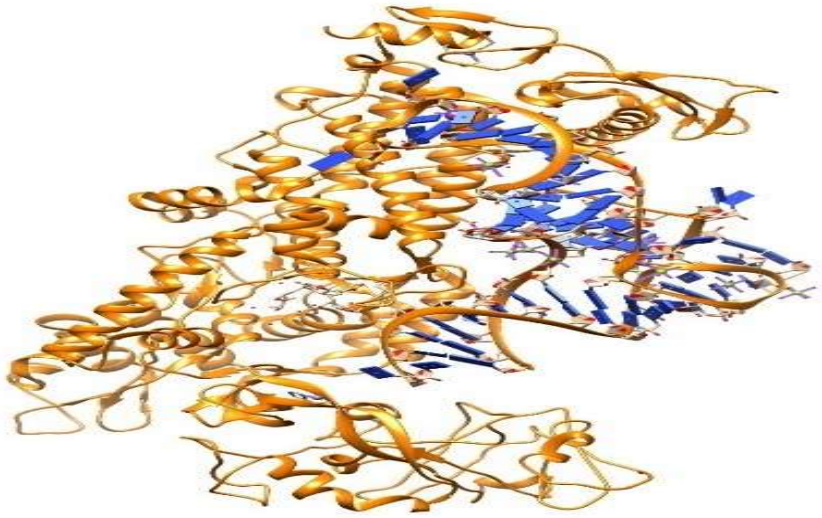


**Figure 3: Crystal structure of IleRS enzyme with bound mupirocin(PDB ID-1FFY)**

***Processing of Protein***

The downloaded receptor protein is having a single chain A, which has been selected for the experimental purpose. The bound ligand mupirocin was separated from the macromolecular complex by using software Chimera (7).

**Molecular Docking Simulation Studies**

Docking of chlorogenic acid ligand on IleRS enzyme was performed by Autodock. All the bonds of ligand were kept flexible, while no residues in receptor were made flexible (8).

**Figure 4: Binding mode of chlorogenic acid within the active site of IleRS receptor**

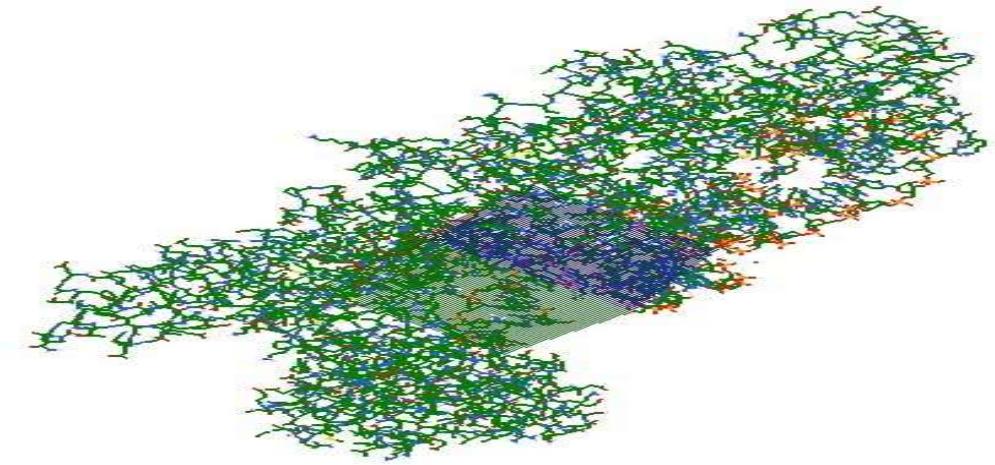


**Figure 5 : Grid box covering all active sites in receptor**

**Docking of Isoleucyl-transfer RNA (tRNA) synthetase (IleRS) with Rosmarinic Acid**

***Crystal structure***

The crystal structure of the protein consisting of receptor associated with bound ligand is downloaded from the Protein Data Bank portal. All the primary information regarding receptor

and structure (1FFY.pdb) registered in the Protein data bank was used. The bound ligand mupirocin is found within the receptor (9).

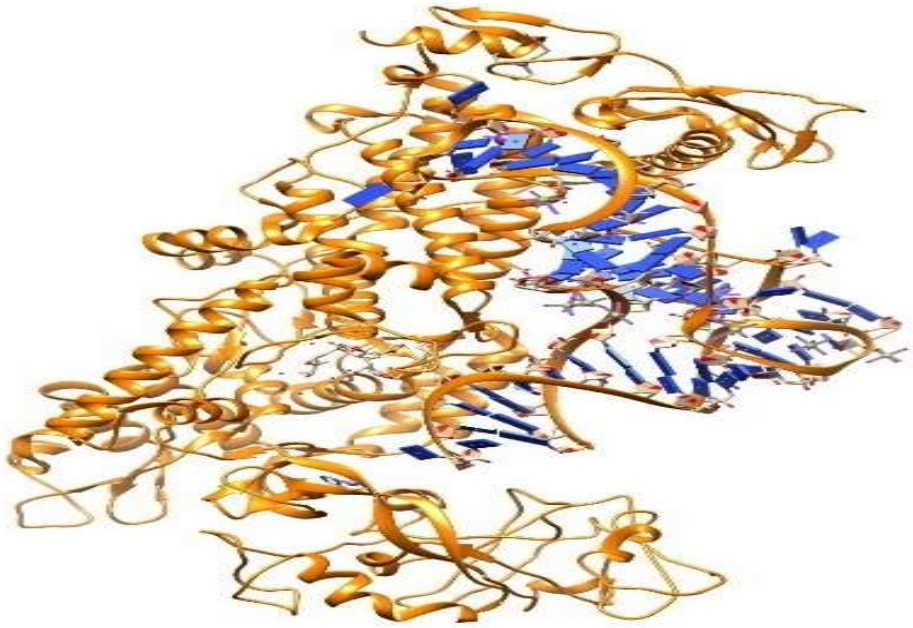


**Figure 6: Crystal structure of IleRS enzyme with bound mupirocin(PDB ID-1FFY)**

***Processing of Protein***

The downloaded receptor protein is having a single chain A, which has been selected for the experimental purpose. The bound ligand mupirocin was separated from the macromolecular complex by using software Chimera (10).

**Molecular Docking Simulation Studies**

Docking of rosmarinic acid ligand on IleRS enzyme was performed by Autodock. All the bonds of ligand were kept flexible, while no residues in receptor were made flexible (11).

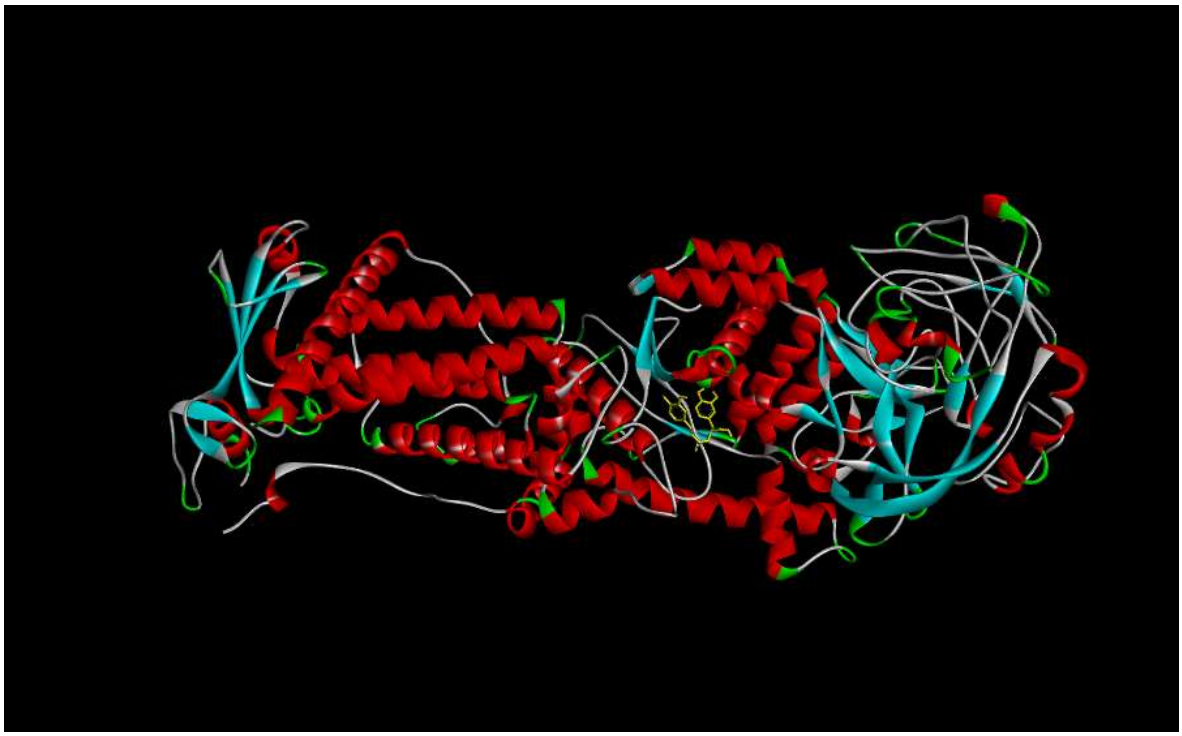


**Figure 7: Binding mode of rosmarinic acid within the active site of IleRS receptor**

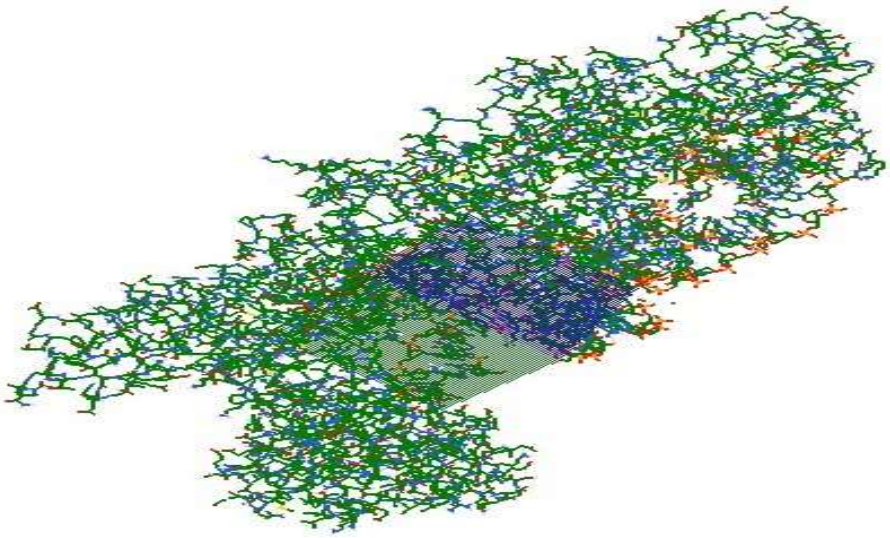


**Figure 8: Grid box covering all active sites in receptor**

**Docking of Isoleucyl-transfer RNA (tRNA) synthetase (IleRS) with Quercetin**

***Crystal structure***

The crystal structure of the protein consisting of receptor associated with bound ligand is downloaded from the Protein Data Bank portal. All the primary information regarding receptor

and structure (1FFY.pdb) registered in the Protein data bank was used. The bound ligand mupirocin is found within the receptor (12).

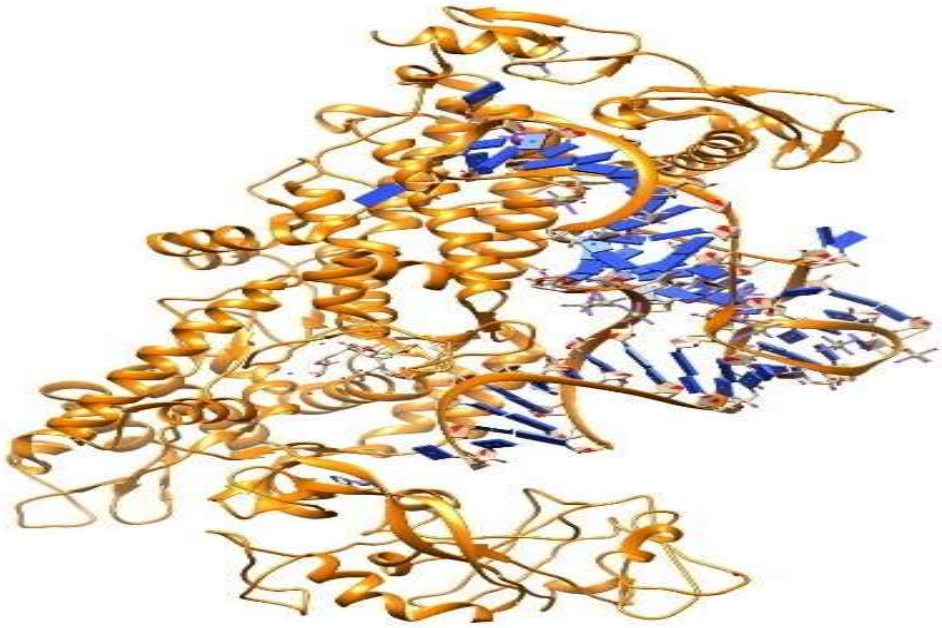


**Figure 9: Crystal structure of IleRS enzyme with bound mupirocin(PDB ID-1FFY)**

***Processing of Protein***

The downloaded receptor protein is having a single chain A, which has been selected for the experimental purpose. The bound ligand mupirocin was separated from the macromolecular complex by using software Chimera (13).

**Molecular Docking Simulation Studies**

Docking of quercetin ligand on IleRS enzyme was performed by Autodock. All the bonds of ligand were kept flexible, while no residues in receptor were made flexible (13).

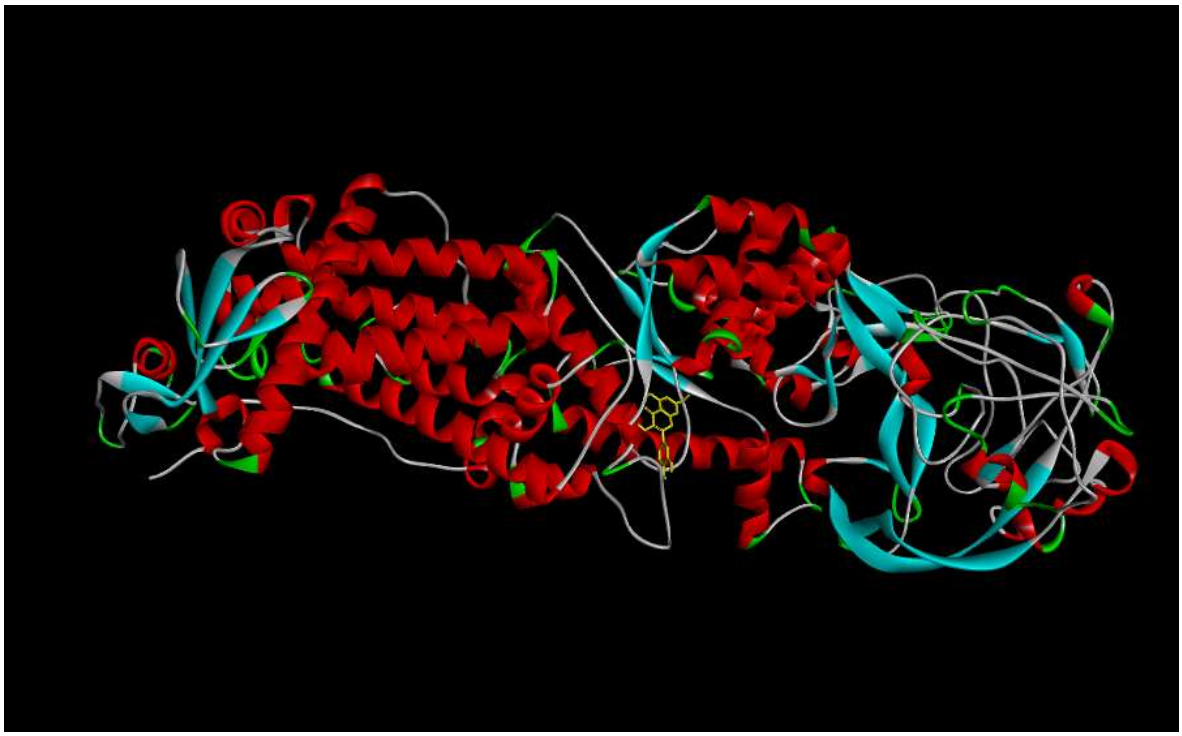


**Figure10: Binding mode of quercetin within the active site of IleRS receptor**

**Molecular docking studies of Rutin**

***Preparation of the grid file***

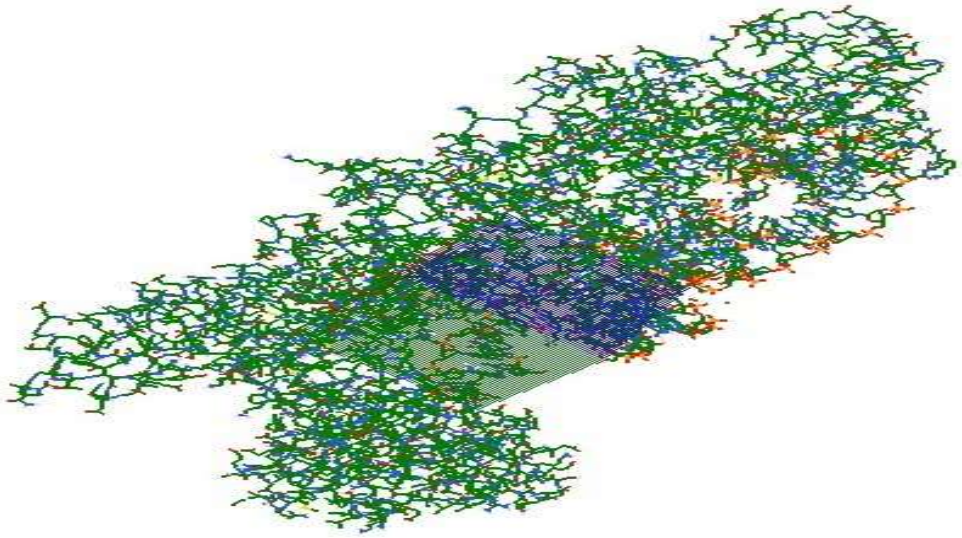


**Figure 11: Grid box covering all active sites in receptor**

**Docking of Isoleucyl-transfer RNA (tRNA) synthetase (IleRS) with Rutin**

***Crystal structure***

The crystal structure of the protein consisting of receptor associated with bound ligand is downloaded from the Protein Data Bank portal. All the primary information regarding receptor and structure (1FFY.pdb) registered in the Protein data bank was used. The bound ligand mupirocin is found within the receptor (13).

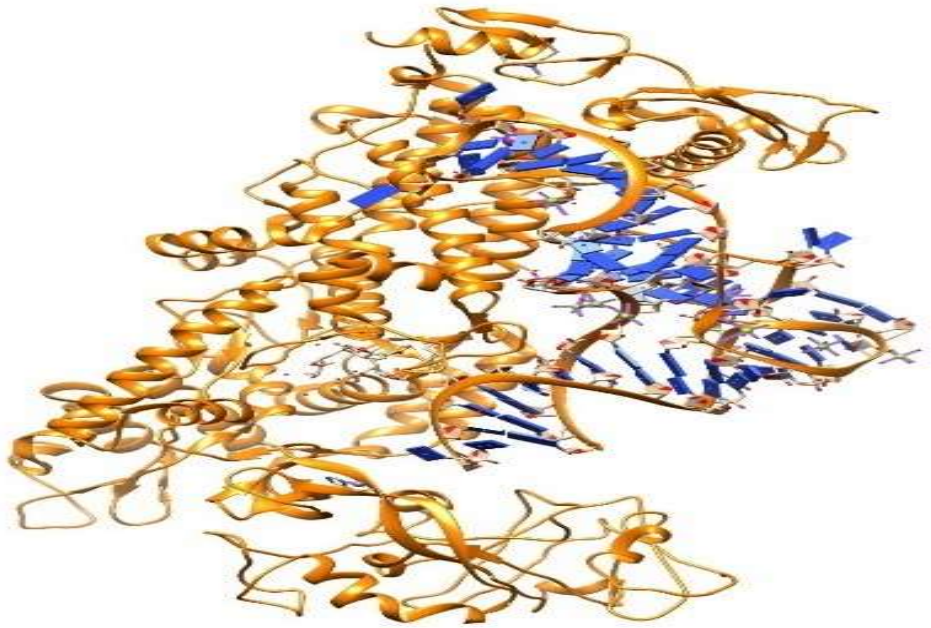


**Figure12: Crystal structure of IleRS enzyme with bound mupirocin(PDB ID-1FFY)**

***Processing of Protein***

The downloaded receptor protein is having a single chain A, which has been selected for the experimental purpose. The bound ligand mupirocin was separated from the macromolecular complex by using software Chimera (14).

**Molecular Docking Simulation Studies**

Docking of rutin ligand on IleRS enzyme was performed by Autodock. All the bonds of ligand were kept flexible, while no residues in receptor were made flexible (15).

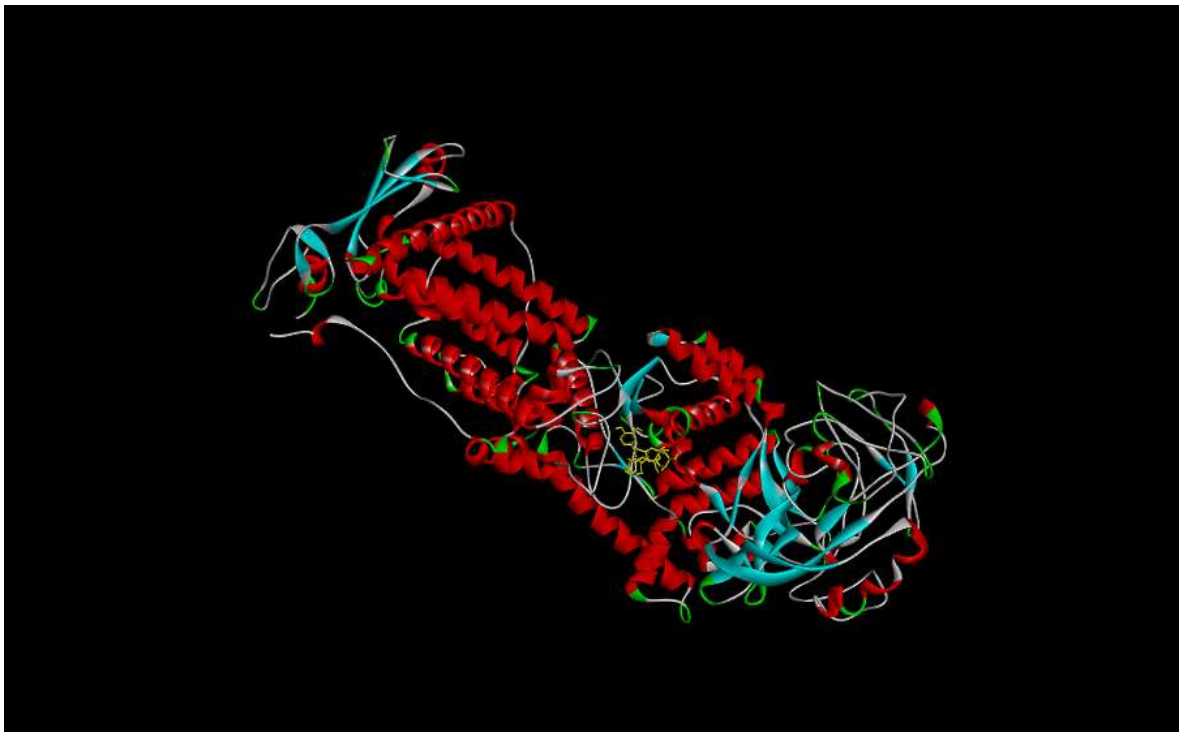


**Figure 13: Binding mode of rutin within the active site of IleRS receptor**

**Result and Discussion**

The search was conducted for novel antibacterial agents that inhibit targets that are essential to bacteria but are unaffected by resistance mechanisms to existing chemotherapeutics. The aminoacyl-tRNA synthetase (AaRS) enzymes have been the focus of recent antibacterial drug discovery research in this regard. By catalysing the synthesis of aminoacyl-tRNAs (aa-rRNA), these enzymes are essential to protein biosynthesis. Protein biosynthesis is stopped when these enzymes are suppressed, which results in the restriction of bacterial development in both in vitro and infectious conditions. These enzymes are fascinating targets for antibiotics. Natural plant phenolics and flavonoids known as rosmarinic acid, chlorogenic acid, rutin, and quercetin have been identified as ligands in this endeavor, and the inhibitory activity of their aminoacyl-tRNA synthetase (AaRS) enzymes has been checked in-silico using the docking method. Chlorogenic acid, rosmarinic acid, quercertin, and rutin all received favourable scores in the molecular docking analysis.The observed docking scores were -4.18, 4.74, -6.89 and -5.28 kcal $mol^{-1}.mol^{-1}$ (Table 1). Table 2 displays the interaction of ligand hits with the targeted site. As a result, it is reasonable to assume that the plant extract's phenolics and flavonoids are effective inhibitors of the IleRS enzyme.

**Table 1: Results of docking of active constituent against against IleRS enzyme**

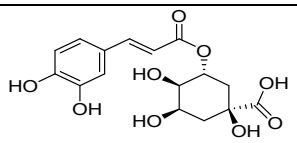
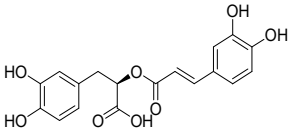
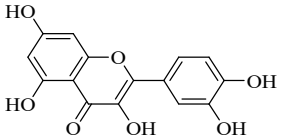
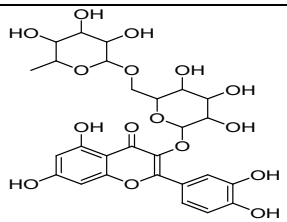
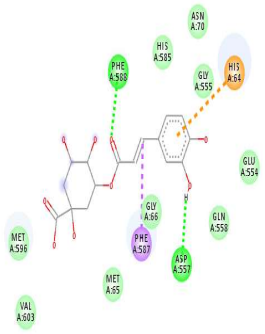
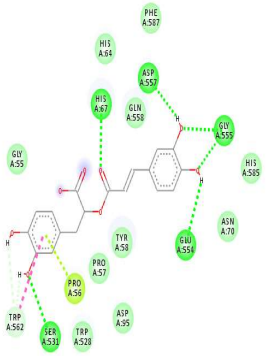
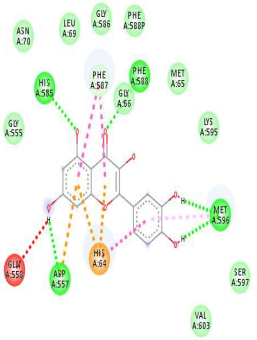
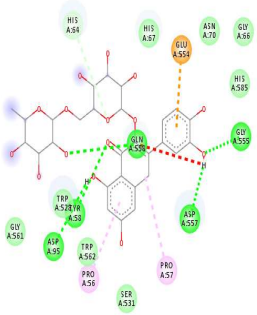
| S. No | Compound | Structure | Binding Energy(Kcal/mole) | Ki (µM) |
|---|---|---|---|---|
| 1. | Chlorogenic acid |  | -4.18 | 158.7 |
| 2. | Rosmarinic acid |  | -4.74 | 335.08 |
| 3. | Quercetin |  | -6.89 | 8.96 |
| 4. | Rutin |  | -5.28 | 134.12 |

**Table 2: Result of Interaction of active constituent against IleRS enzyme**

| S.No. | Compound | Interaction | Inference |
|---|---|---|---|
| 1. | Chlorogenic acid |  | The chlorogenic acid interacts with the Phe587, Phe588, Asp557 and His64 residues of IleRS to form a complex structure. |

| | | | |
|---|---|---|---|
| 2. | Rosmarinic acid |  | The rosmarinic acid interacts with the Trp562, Ser531, Pro56, Glu554, Gly555, Asp557 and His67 residues of IleRS to form a complex structure. |
| 3. | Quercetin |  | The quercetin interacts with the His585, Phe588, Met596, His64 and Asp557 residues of IleRS to form a complex structure. |
| 4. | Rutin |  | The rutin interacts with the His64, Glu554, Gly555, Asp557, Gln558, Pro57, Asp95, and Tyr58 residues of IleRS to form a complex structure. |

**Conclusion**

In the existing research, we aimed to discover novel and powerful inhibitor molecules to conquer the microbial resistance control. Selective plant phenolics molecule turned into

molecular docked with IIeRS. The final results of the existing research found out that the chosen molecule highly bounded with IIeRS thereby inhibiting the bacterial protein synthesis.

**Reference**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