



## Molecular Docking Studies of Isorhamnetin from *Corchorus olitorius* with Target Alpha-amylase Related to Type 2 Diabetes

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### ABSTRACT

Type II diabetes (T2DM) is a metabolic disorder arising from impaired insulin action and insulin secretion, which leads to hyperglycemia and also the decimation of the body system. Acarbose, an anti-diabetic drug decreases the postprandial hyperglycemia, by slowing down the absorption of glucose through the inhibition of the carbohydrate-hydrolyzing enzyme ( $\alpha$ -amylase), howbeit with serious side effects. In the present study, with the aid of *in silico* experiment we have identified isorhamnetin from *Corchorus olitorius* plant to have a higher inhibitory potential on alpha-amylase (-8.5Kcal/mol) as compared with acarbose (-7.1Kcal/mol) and other compounds (caffeic acid, -6.5 Kcal/mol and chlorogenic acid, -7.3 Kcal/mol) characterized from the same plant (*Corchorus olitorius*). This study however corroborates the earlier wet experiment on the inhibitory properties of isorhamnetin.

**Keywords:** Type II diabetes, Acarbose, Isorhamnetin, Chlorogenic acid, caffeic acid and *in silico*

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### INTRODUCTION

Diabetes is the 7<sup>th</sup> leading cause of mortality in the United States, with over 68,071 death certificates registering it as a major cause of death [1]. The approximate estimation of diabetes in Africa is 1% in rural areas, and varies from 5% to 7% in urban sub-Saharan [8].

Type II diabetes (T2DM) is a group of metabolic disorders arising from impaired insulin action and insulin secretion, leading to hyperglycemia. T2DM destroys many of the body system, most especially blood vessels [4]. T2DM may be in part as a result of increased hepatic glucose production, reduced insulin secretion, and impairment of insulin action [5, 13]. Patients suffering from type II diabetes are insulin-resistant. There are several mechanisms employed in the treatment of type II diabetes, these vary from changes in life-styles, exercises, insulin injections, and various anti-diabetic drugs. However, the management of this disease without any side effect still poses a great challenge to the medical system [2, 7].

One of the therapeutic approaches for the treatment of diabetes in the early stage is to decrease the postprandial hyperglycemia, by slowing down the absorption of glucose through inhibition of the carbohydrate-hydrolyzing enzymes,  $\alpha$ -amylase and  $\beta$ -glucosidase [11]. Many drugs are currently in use to achieve this, one of such drugs is acarbose which acts by inhibiting the  $\alpha$ -amylase enzyme, this drug has been shown to have serious side effects ranging from severe stomach pain, severe constipation to diarrhea and so on. Hence, the need for a novel drug with little or no side effect, medicinal plants have been reported to have anti-diabetic effects, with less side effects and relatively low cost [12] and such are good replacement for anti-diabetic drugs like acarbose.

In the present study, we carried out *in silico* experiment which corroborates the earlier wet experiment [6], that *Corchorus olitorius* consumed in various part of the world as leafy vegetable has anti-diabetic effect by inhibition of the  $\alpha$ -amylase enzyme. However, molecular docking of these characterized compounds (caffeic acid, chlorogenic acid and isorhamnetin) from *Corchorus olitorius* revealed that, only isorhamnetin possessed a better inhibitory effect on  $\alpha$ -amylase enzyme than the rest of the compounds as compared with the standard anti-diabetic drug acarbose.

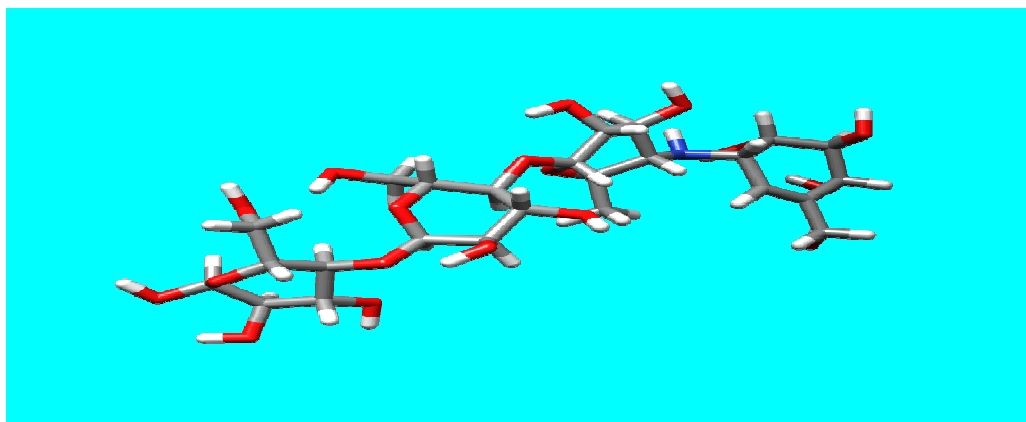


Fig. 1: Showing the 3 D structure of acarbose

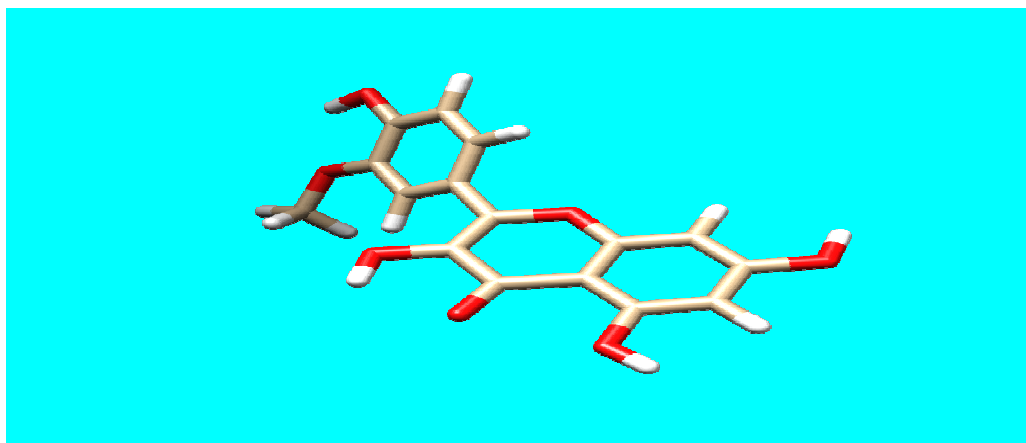


Fig. 2: Showing the 3 D structure of Isorhamnetin

## EXPERIMENTAL SECTION

### *Selection and Preparation of Macromolecule*

The crystal structure of Human Pancreatic alpha-amylase in complex with myricetin (PDB ID: 4GQR) was retrieved from the protein data bank (<http://www.rcsb.org>). Human Pancreatic alpha-amylase (Accession: AAA52280.1 GI: 178567) “Fasta” file was downloaded from [www.ncbi.nlm.nih.gov/pubmed](http://www.ncbi.nlm.nih.gov/pubmed), this was used in modeling the starting structure of Human Pancreatic alpha-amylase, used in this study. The homology modeling was done in the Swiss Model Server (<http://swissmodel.expasy.org>). The 3D structure of the human alpha-amylase was modeled using the co-ordinate file of template from protein data bank (PDB ID: 4GQR). Deletion of water and ligand coordinates was carried out prior to the molecular docking.

### **Selection and Preparation of Acarbose, Isorhamnetin, Caffeic acid and Chlorogenic acid**

The three-dimensional (3D) structures of Acarbose, Isorhamnetin, Caffeic acid and Chlorogenic acid were obtained from the pubchem database [10]. The optimized ligand molecules (Acarbose, Isorhamnetin, Caffeic acid, and Chlorogenic acid) were docked into the refined human pancreatic alpha-amylase model using “LigandFit” in the AutoDock 4.2.

### **Molecular Docking**

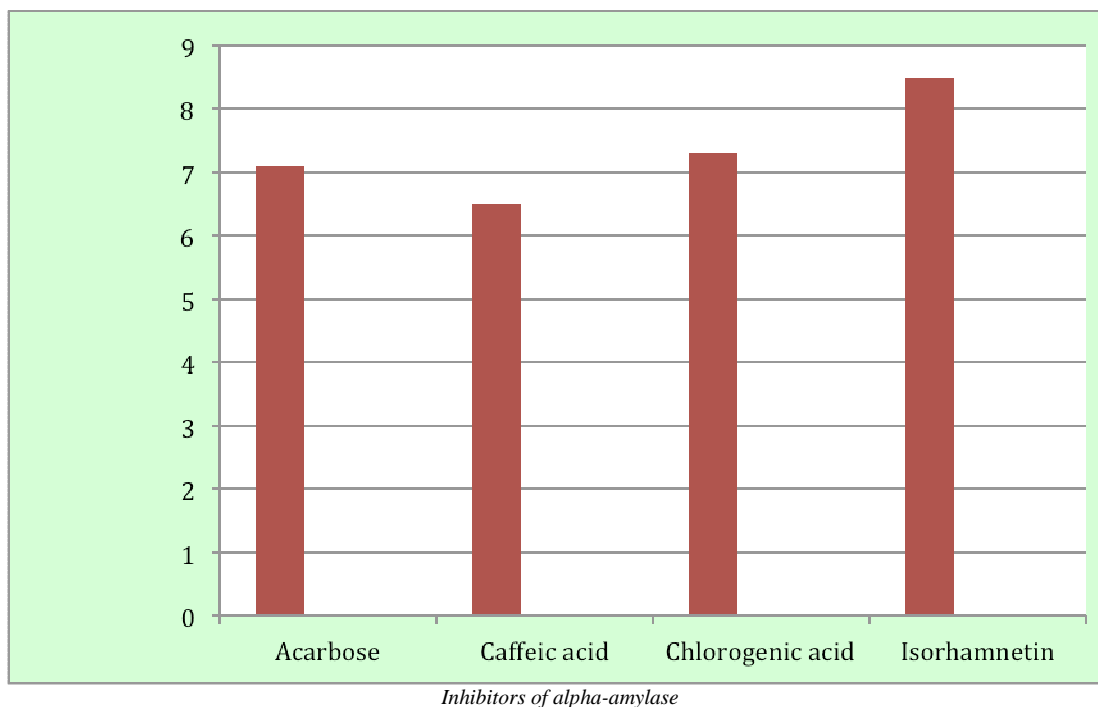
The docking of the of Acarbose, Isorhamnetin, Caffeic acid and Chlorogenic acid to the binding site of human alpha-amylase was done using the autodock vina 4.2 [9]. Water molecules and the ligand (myricetin) complex with alpha-amylase were removed for better docking scores. The protein was treated as a rigid body [3], while the

rotatable bonds of the ligands were set to be free. The grid box size was set at 13.44, 15.14 and 39.44 Å<sup>o</sup> (x, y, and z) to include all the amino acid residues. The spacing between grid points was 0.375 angstroms.

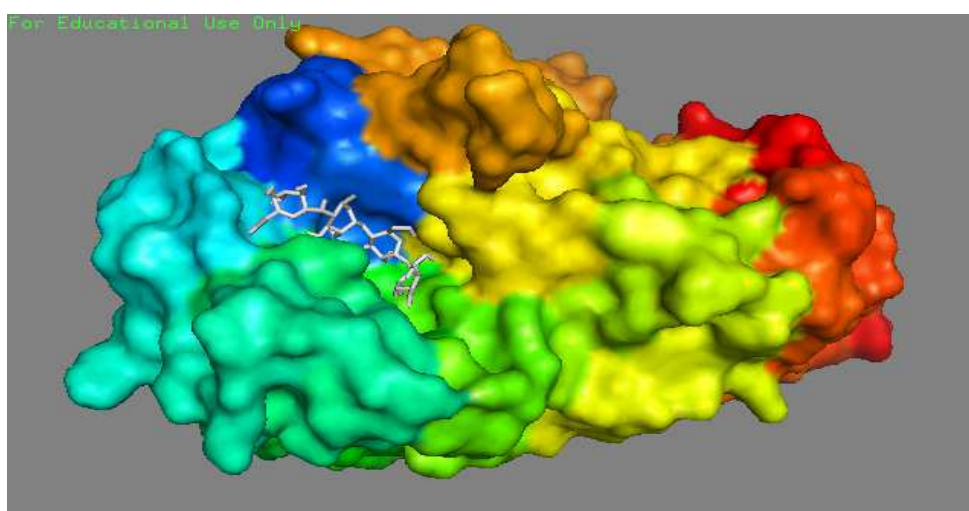
## RESULTS AND DISCUSSION

**Table 1: Showing Acarbose, Isorhamnetin, Caffeic acid and Chlorogenic acid with corresponding binding energies obtained from docking with human alpha-amylase using AutoDock program**

Compound Name	Binding Energy (Kcal/mol)
Acarbose	-7.1
Caffeic acid	-6.5
Chlorogenic acid	-7.3
Isorhamnetin	-8.5



**Figure 3: A bar chart showing the binding energy/ affinity of Acarbose, Caffeic acid, Chlorogenic acid, Isorhamnetin**



**Figure 4: alpha-amylase enzyme with acarbose at its active site**

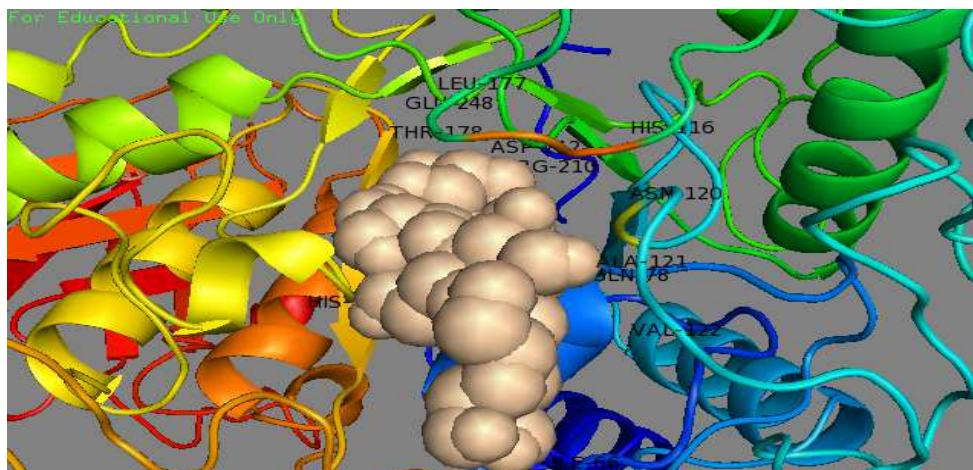


Figure 5: Molecular interaction of acarbose with alpha-amylase

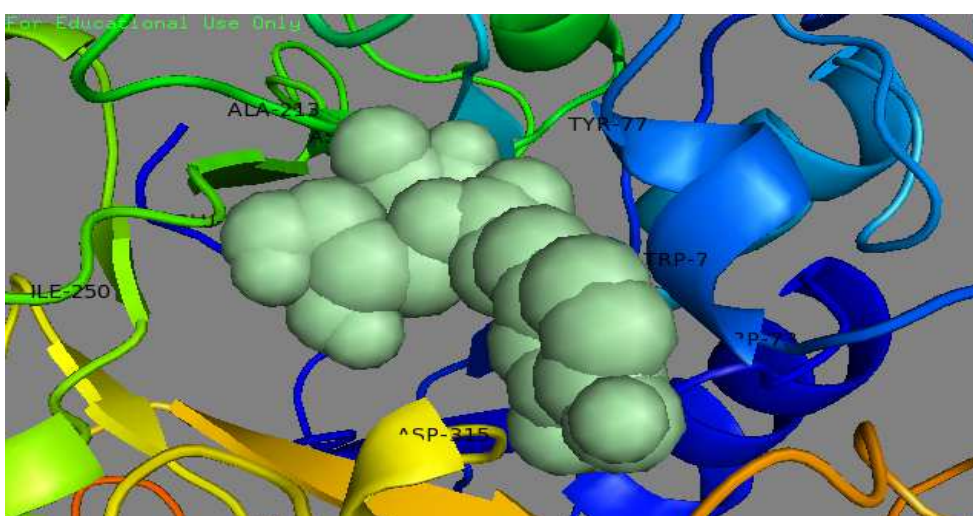


Figure 6: A molecular interaction of Chlorogenic acid with alpha-amylase

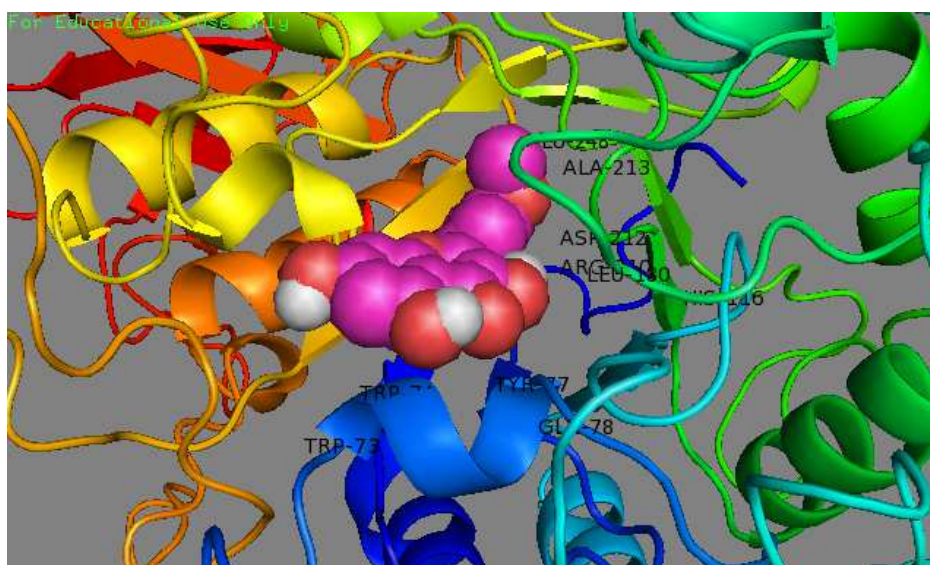


Figure 7: A molecular interaction of Isorhmnetin with alpha-amylase

Acarbose is a drug that have been used to decrease postprandial hyperglycemia by inhibition of the  $\alpha$ -amylase enzyme, however with its own side effects. In the present study through the aid of molecular docking studies, we

have identified isorhamnetin (a natural compound) from *Corchorus olerius* to be a better inhibitor of alpha-amylase.

Acarbose, an established anti-diabetic drug was docked with the aid of autodock/vina to our modeled alpha-amylase enzyme (receptor) (figure 5) and showed a binding energy of -7.1Kcal/mol. Caffeic acid, chlorogenic acid and isorhamnetin characterized from *Corchorus olerius* plant were also docked with our modeled alpha-amylase and with binding energies of -6.5 Kcal/mol, -7.3 Kcal/mol and -8.5 Kcal/mol respectively (table 1). From the result of our molecular docking scores it is evident that isorhamnetin has a better inhibitory effect than acarbose, also chlorogenic acid is a slight better than acarbose, however acarbose has a better inhibitory effect than caffeic acid, though caffeic acid with a binding energy of -65Kcal/mol is still a good inhibitor of alpha-amylase( figure 3). The alpha-amylase-isorhamnetin better interactions when compared with the rest (acarbose, caffeic acid and chlorogenic acid) are a function of the amino acid residues in the active site of the alpha amylase. The amino acid residues within the 4A that stabilized the alpha-amylase –isorhamnetin interaction are GLN 78, TRY 77, HIS 116, ALA 213, HIS 314 LEU 180, TRP 73, ASP 212 TRP 74, ARG 210, GLU 248, ALA 213. The extensive hydrogen bonds interactions with GLN 78, TRY 77, GLU 248, ASP 212 residues, pi-sigma stacking interactions between TRP 73 and TRP 74 of alpha-amylase and coupled with the electrostatic attractions between the amino acid residues of alpha-amylase enzyme and isorhamnetin , may account for its lowest binding energy (8.5Kcal/mol), better interaction and better inhibitory potential.

### CONCLUSION

Based on the present *in silico* study, Isorhamnetin is the best inhibitor of alpha amylase, when compared with acarbose, caffeic acid and chlorogenic acid. This result confirmed the earlier wet experiment, that isorhamnetin has inhibitory potential on alpha-amylase and as such isorhamnetin should be consider for clinical trials as an anti-diabetic drug.

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