# Using AutoDock Vina to Create a Quercetin Derivative for Binding Xanthine Oxidase

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## Abstract

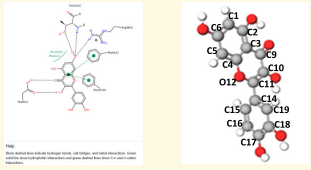
Quercetin is an anti-hyperuricemia agent that competitively inhibits the natural ligands for binding at the active site of Xanthine Oxidase (XOD), which catalyzes the oxidation of hypoxanthine to xanthine and then to urate. It is an old treatment for gout, a disease resulting from the buildup of uric acid crystals in the joints of limbs. The long term goal of the project is to find a water soluble quercetin derivative with higher affinity for the XOD active site. The quercetin molecule contains three six-membered rings with peripheral hydroxyl groups that have specific interactions with the side chains of XOD. In the project, computer programs are used to design quercetin derivative compounds by replacing the peripheral hydroxyl groups with aldehyde and carboxylic acid groups. These quercetin derivatives were tested for binding XOD by the AutoDock Vina software. Of the several aldehyde and carboxylic acid derivatives tested for docking, their results showed less affinity for the active site. Once a promising derivative is found, it will be evaluated for suitability and possibility of synthesis, then the binding studied by NMR spectroscopy.

## Introduction

Hyperuricemia is a condition where the level of uric acid in the blood is much higher than normal. The uric acid builds up in body fluids which eventually crystallizes in the joints, resulting in the disease gout. Gout severely reduces mobility and causes extreme pain and chronic inflammation. The majority of cases of high serum uric acid level is due to under excretion; sometimes it is a result of overproduction and often is a combination of the two (Choi, Mount, & Reginato, 2005). Xanthine oxidase (XOD) is the final enzyme in the purine degradation pathway, which converts it to the final waste product, uric acid. It catalyzes the oxidation of hypoxanthine to xanthine and then to urate (Harrison, 2002). Some drugs for treating gout exploit the XOD activity to reduce the amount of uric acid production. One of these drugs is the competitive inhibitor quercetin.

## Objectives

Quercetin is a relatively old treatment for gout, and it is no longer the most effective. In changing specific functional groups on the quercetin molecule, we can more fully understand the XOD active site and utilize the bonding capability of the active site. Our objective is to find a quercetin derivative with an equal or higher affinity for binding XOD and one that would be more suitable for wet laboratory analysis later.




Picture: http://www.rcsb.org/pdb/explore/explore.do?structureId=3NVY

## Methods

We used Protein Data Bank files and Autodock tools to convert files of both the derivatives and XOD into .pdbqt files for Autodock Vina. After finding the appropriate box dimensions, AutoDock Vina was used to dock the different modified ligands into the XOD binding site (Trott et. al, 2010). New derivatives were made using Spartan10, and the Vina results were inspected in PyMOL to visualize the interactions. We analyzed the residues and atoms of XOD interacting with quercetin, as well as atoms we could exploit with the derivatives in order to increase affinity for the active site.

## Results

### Test box dimensions (nxnxn)

| Dimension | RMSD |
|---|---|
| 11 | 1.071077 |
| 12 | 1.026314 |
| 16 | 1.039792 |
| 20 | 1.04297 |

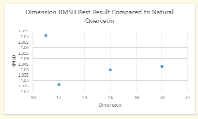



Figure 1

| Dimension | Number of results |
|---|---|
| 11 | 6 |
| 12 | 7 |
| 14 | 14 |
| 16 | 11 |
| 20 | 9 |

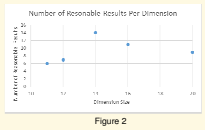



Figure 2

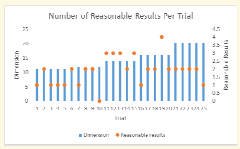



Figure 3

Final box dimensions and placement
center_x = 87.5
center_y = 8
center_z = 16

size_x = 16
size_y = 16
size_z = 16

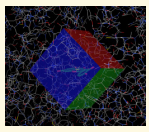
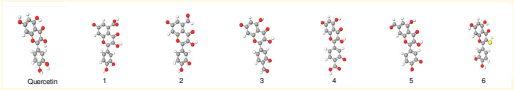


Figure 4

Figure 1: RMSD values between natural quercetin and the best result of docked quercetin into the active site. Lower RMSD values are better results.
Figure 2: Total number of reasonable results for all five trials at each dimension size. A reasonable result is a docked quercetin result that appears to have the same mode of binding as seen in the original PDB file.
Figure 3: Each dimension size was run five times, resulting in a certain number of reasonable results per trial. Blue bars represent dimension sizes and refer to the left y-axis scale. The number of reasonable results per trial is seen as orange dots and refer to the right y-axis scale. A box size of 16x16x16 was chosen to be used in the rest of the docking experiments for its second highest total number of reasonable results from all five trials.
Figure 4: Example of test box dimension and center point.

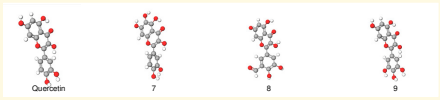
### Tested derivatives

| Atom name | C1 | C2 | C5 | C6 | C9 | C10 | C15 | C16 | C17 | C18 | C19 | O12 | Affinity | RMSD |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Quercetin | H | OH | H | OH | O | OH | H | H | OH | OH | H | O12 | -9.1 | 1.04 |



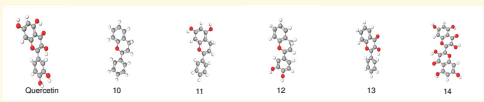

### Replaced functional groups

| Atom name | C1 | C2 | C5 | C6 | C9 | C10 | C15 | C16 | C17 | C18 | C19 | O12 | Affinity |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | | COO- | | | | | | | | | | | -7.2 |
| 2 | | CHO | | | | | | | | | | | -8.2 |
| 3 | | | | | | | | | CHO | | | | -8.6 |
| 4 | | | | | | | | | COO | | | | -8.4 |
| 5 | | | | CHO | | | | | | | | | -7.2 |
| 6 | | | | | | SH | | | | | | | -7.5 |



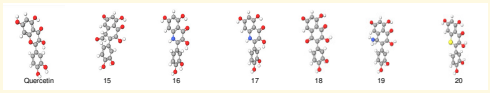

### Added functional groups

| Atom name | C1 | C2 | C5 | C6 | C9 | C10 | C15 | C16 | C17 | C18 | C19 | O12 | Affinity |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 7 | OH | | | | | | | | | | | | -8.2 |
| 8 | | | | | | | | CHO | | | | | -8.3 |
| 9 | | | | | | | | OH | | | | | -7.9 |




### Deleted functional groups

| Atom name | C1 | C2 | C5 | C6 | C9 | C10 | C15 | C16 | C17 | C18 | C19 | O12 | Affinity |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 10 | | H | | H | H2 | H | | | H | H | | | -8.7 |
| 11 | | | | | H2 | H | | | H | H | | | -9.5 |
| 12 | | H | | H | H2 | H | | | | | | | -8.3 |
| 13 | | H | | H | | | | | H | H | | | -8.2 |
| 14 | mirror at C11-C14, C11 side | | | | | | | | | | | | -9.2 |

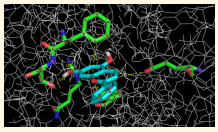



### Changes within a ring

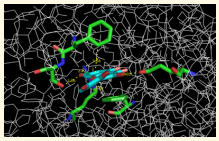
| Atom name | C1 | C2 | C5 | C6 | C9 | C10 | C15 | C16 | C17 | C18 | C19 | O12 | Affinity |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 15 | | | | | | | | | | | | CH-CHO | NA |
| 16 | | | | | | | | | | | | NH(up) | -8.2 |
| 17 | | | | | | | | | | | | NH(down) | -8.1 |
| 18 | | | | | | | | | | | | CO | -8.2 |
| 19 | | | | | | | | | | | | CHN | -6.9 |
| 20 | | | | | | | | | | | | S | -7.8 |

Distance of interactions (Å) between key XOD amino acid side chain atoms to certain atoms on natural quercetin compared to derivative 11 (derivative with highest affinity).

| | Q | D11 |
|---|---|---|
| Arg880 to O29 | 2.77 | 2.77 |
| Thr1010 to O29 | 2.66 | 2.66 |
| Phe914 to C4 | 3.57 | 3.57 |
| Phe1009 to C4 | 3.68 | 3.68 |
| Glu802 to C10 | 3.02 | 3.17 |



D11



Q

## Conclusions

1. All of the derivatives tested, except for D15, have the correct mode of bonding similar to natural quercetin.
2. D11 had the highest affinity, followed by D14, D10, and D3. The interactions seen with D11 are very close to being the same as natural quercetin, but with slightly more distance from Glu802.
3. D15 did not have the correct mode of binding or interactions that were reasonable.
4. Derivatives with fewer oxygen atoms seem to have higher affinity.

## Future Outlook

Finding a derivative with the same or greater affinity for the active site is possible. Once a promising derivative is found, we will see if it is naturally occurring or possible to synthesize. The binding interactions will then be analyzed using NMR spectroscopy.

## References

Choi, H. K., Mount, D. B., & Reginato, A. M. (4 October 2005). Pathogenesis of gout. *Annals of Internal Medicine, vol. 143(7),* p499-516. Retrieved from http://www.the-aps.org/mm/publications/journals/pim/choi-pdf.pdf.

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O. Trott, A. J. Olson, AutoDock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization and multithreading, *Journal of Computational Chemistry 31 (2010) 455-461.*

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