

Targeting Cariogenic Enzymes Through Food Design: A Combined Docking and pH Kinetics Study of Sweeteners and Polyphenols

Saisha Sikka
saishasikka9@gmail.com

ABSTRACT

Biofilm formation is a biological process driven by complex interactions between acid-producing microorganisms and fermentable carbohydrates, influencing oral microbial balance and overall oral health. The acids produced by bacteria such as *Streptococcus mutans* lower the pH, which demineralises and erodes the enamel subsurface. This study addresses the gap in the integration of enzyme-level computational screening with practical food testing and dietary reformulation strategies. This is accomplished by utilising a computational approach that combines molecular docking on AutoDock Vina (Tamarind Bio) and literature mapping to identify specific sweeteners that exhibit favourable interactions with key carbohydrate-processing enzymes. Molecular docking was done with enzymes commonly produced by oral, acidogenic bacteria such as *S. mutans* (Glucosyltransferase B - GtfB, Glucosyltransferase C - GtfC, Lactate Dehydrogenase- LDH and Human α -Amylase) against ligands from sweeteners (Xylitol and Erythritol) and ligands from polyphenols (Catechin, Epicatechin, Quercetin, Ferulic acid and Chlorogenic acid). Sucrose was tested as the control. Finally, the study also reformulates the best candidates into 2–3 tooth-friendly baked prototypes in an effort to mitigate the effects of acidogenic bacteria and carbohydrate metabolism in commonly consumed foods. It was concluded that Epicatechin was the best-performing polyphenol ligand due to its high binding affinity for GtfB, GtfC, and human α -amylase. For sweeteners, Erythritol was the best result due to the lowest binding affinities when tested against GtfB, GtfC, and LDH. Human α -Amylase was the only exception, with Xylitol as the best result, presenting the lowest binding affinity of -4.94 kcal/mol. LDH was the only exception, with Quercetin as the best result, with the highest binding affinity of -8.666 kcal/mol. Lastly, the study lists common food sources which contain beneficial ligands so they can be compiled to create functional recipes which do not promote biofilm or caries formation. Moreover, the outcomes of this study can be utilised to create oral care products and preventative strategies aimed at decreasing the occurrence of dental caries.

Keywords: Molecular docking, Dental caries, Polyphenols, Sugar substitutes and Acidification kinetics

INTRODUCTION

Dental caries, or tooth decay, is a disease prevalent across all age groups. It is one of the oldest diseases humans have faced. ¹ It develops in tooth crowns and roots and can even affect the primary teeth of infants and toddlers. It results from the complex interaction between acid-producing bacteria and fermentable carbohydrates from dietary molecules. ² The end products of bacterial metabolism accumulate on the tooth surface, resulting in a drop in pH. Acids can cause further damage by eroding the pores of the enamel and demineralising the subsurface. ³ Fluoride from products can help protect from surface demineralisation as well as the reaction products of demineralisation, such as calcium and phosphate, which also accumulate in the enamel surface and prevent further mineral loss. ⁴ Sugar plays a vital role in the formation of dental caries because it is readily metabolised by many bacteria involved in dental biofilm formation. Different sugars have different acidogenic properties. For example, lactose (milk sugar) is considered to be less acidogenic and cariogenic. This suggests that diet is a modifiable risk factor. ⁵

Research has increasingly focused on preventive strategies to reduce acid production rather than on treating it after the cavity or damage has occurred. ⁶ Since caries are driven by microbial acid production, targeting dietary acidogenicity and sugar intake enables the development of non-invasive preventive strategies that reduce enamel demineralisation without invasive intervention. ⁷ Several studies have examined microbial ecology as the root cause of caries and have focused on diet as a determinant of caries risk. ⁸ Moreover, carbohydrate metabolic pathways are unregulated in caries, which justifies targeting metabolism in Caries prevention. ⁹ Furthermore, sugar type influences cariogenicity, and several studies have shown that sugar substitutes reduce acid production. ¹⁰ Lastly, research on plant polyphenols and their anti-cariogenic properties has shown that they inhibit microbial enzymes by limiting biofilm formation and acid production. ¹¹ Studies suggest enamel repair as a non-invasive biomimetic strategy of amelogenesis. ¹²

Studies utilise molecular docking as a screening tool to predict enzyme-ligand interactions and find the advantages of in silico approaches. Metabolic pathways have been modelled to link carbohydrate metabolism and acid production through the use of bioinformatics. ⁹ Studies have also provided evidence of enzyme inhibition. Lastly, transcriptomics has been used to elucidate the importance of gene expression analysis. ¹³ In this paper, we will also use bioinformatics to analyse and screen for molecular interactions relevant to carbohydrate metabolism and acid production.

Collectively, existing studies focus on biological mechanisms/repair but not food reformulation. While many have examined the effects of acid on caries and its prevention, no ingredient-level screening has been conducted. Despite extensive research, there's limited integration of enzyme-level computational screening with practical food testing and dietary reformulation strategies to mitigate the effects of acidogenic bacteria and carbohydrate metabolism in commonly consumed foods. Thus, in this study, it was hypothesised that a computational approach combining docking and literature mapping can be used

to identify specific sweeteners that exhibit favourable interactions with key carbohydrate-processing enzymes. Finally, the best candidates can be reformulated into 2–3 tooth-friendly baked prototypes.

METHODOLOGY

Data collection

Large amounts of information relevant to the topic of this research paper were gathered. Databases such as FooDB, Phenol-Explorer, and HOMD (Human Oral Microbiome Database) were explored using the same relevant keywords and combinations used for the literature review. These include ‘cavity’, ‘enzymes’, ‘bioinformatics’, ‘sweeteners’, ‘plant polyphenols’, and ‘microbes’. In addition, research papers that contain data sets were also analysed. Information on the numerous polyphenols present in common baking ingredients such as whole-grain flour, common sugar, and cacao powder was also noted. From all the collected data, information about which kind of glucotransferase enzyme encode all known species of *Staphylococcus mutans* bacteria make were identified and the type of glucan (α -1,3 or α -1,6) they produce for dental plaque formation, the effect of consuming the same weight of foods of various glycemic indexes were tested with dental plaque pH, the impact of *S.mutans* in different baby formulas were tested to see biofilm formation, pH and lactic acid production, the implication of bacteria which participate in anaerobic nitrate-nitrate metabolism in the mouth to influence the oral microbiome balance, plaque pH and dental health and molecular docking data about the compounds from sugarcane extract and their binding affinity with α -glucosidase. All this data was compiled to make a final dataset, which was used to create a Final Enzyme Docking sheet.

Data processing

After collecting data, the obtained information was filtered and ordered systematically to create a dataset applicable to molecular docking analysis. First, all redundant, irrelevant, or non-food-grade compounds were eliminated, and all polyphenols and sweeteners that were common and safe to consume and had a known structure were retained. Equally, out of the number of enzymes of interest that have been identified in the oral cavity, four functionally competent and structurally solved enzymes, including glucosyltransferase B (GtfB), glucosyltransferase C (GtfC), lactate dehydrogenase (LDH), and human salivary amylase, were picked due to their known functions in biofilm formation, carbohydrate metabolism, and acid production. PubChem Compound IDs (CIDs) and 3D structural files of each chosen ligand were checked. In contrast, experimentally determined PDB structures of every enzyme were authenticated to maintain the uniformity and dependability of docking simulations. Each curated ligand and enzyme was then randomly matched to form all the possible combinations of receptor-ligand complexes, which were then tabulated into a standardised Final Enzyme Docking Sheet containing enzyme name, PDB ID, ligand name and PubChem CID. This organised sheet served as structured input for uniform and reproducible docking experiments across all chosen combinations.

Molecular docking

The process of molecular docking began by downloading the Legacy PDB format of enzyme structures from the RCSB Protein Data Bank (PDB IDs 3AIB, 3AIC, 4LMR and 1HNY) and the 3D SDF structures of the ligands from PubChem. Docking was performed using AutoDock Vina on Tamarind Bio, a managed cloud platform which converts the uploaded structures to PDBQT format using its default settings. Each docking job was named according to the combination being tested, for example Amylase_Catechin (Enzyme_Ligand). The active site was not known and neither were any known ligands used. Hence, a blind docking approach was adopted: For each docking run, the grid box was adjusted manually to enclose the full receptor, ensuring no part of the enzyme protruded beyond the box walls while permitting free ligand rotation. A grid spacing of 0.375 Å was applied throughout. No explicit pH-based protonation prediction was performed; ionisable residues and ligand hydroxyl groups were therefore modelled in their default states rather than being adjusted to a specified pH. Grid parameters for all four receptors are given in Table 1.

Receptor	PDB ID	Centre x, y, z / Å	Box size x, y, z / Å	Spacing of Grid Map Calculation / Å
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	206 , 50 , 201	143 x 115 x 133	0.375
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	215, 54, 191	117 x 94 x 116	0.375
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	10, 128, 25	120 x 90 x 83	0.375
Human α -Amylase	1HNY	9, 61, 16	84 x 83 x 73	0.375

Table 1. The grid box parameters used for the blind docking of each of the four enzymes.

Next, the sdf ligand file was uploaded from the folder, and the job was submitted. Docking was performed using AutoDock Vina v1.2.5 with the default vina scoring function, and binding affinities were reported in kcal/mol. An exhaustiveness value of 8 was used, with the receptor treated as rigid and ligand torsions free to rotate. For each docking run, nine binding modes were generated within a default energy range of 3 kcal/mol, and the highest-scoring pose was taken as the predicted binding affinity. Identical scoring and search settings were applied.

Once the job had finished running, the result was downloaded and converted from a ZIP file to a regular desktop file. Within the file, the 'log.txt' was accessed to view the binding affinity of the ligand to the enzyme. The binding affinities were noted in the Enzyme Docking Sheet. The same process was repeated for all the combinations.

RESULTS

Molecular docking analysis

Molecular docking was conducted by testing enzymes from *S. mutans* such as Glucosyltransferase B (GtfB), Glucosyltransferase C (GtfC), Lactate Dehydrogenase (LDH), and Human α -Amylase with 2 categories of ligands. This included polyphenols such as Catechin, Epicatechin, Quercetin, Ferulic acid, and Chlorogenic acid, and sweeteners such as Xylitol, Erythritol, and, finally, Sucrose as the control (Figure 1). Sucrose was used as a control as it is a commonly consumed cariogenic sugar and provides a baseline for comparison with other sweeteners and polyphenols. The binding affinities for all the combinations of enzymes and ligands are listed in Table 2. Every possible combination was tested. For GtfB, GtfC, and LDH, Erythritol was the best result for the sweeteners, with the lowest binding affinity, as shown in Figure 2. Human α -Amylase was the only exception, with Xylitol as the best result, presenting the lowest binding affinity of -4.94 kcal/mol. In terms of polyphenols, Epicatechin shows the best result due to its high binding affinity for GtfB, GtfC, and Human α -Amylase, as shown in Figure 3. LDH was the only exception, with Quercetin as the best result, with the highest binding affinity of -8.666 kcal/mol. In table 2, The best-performing polyphenol and best-performing sweetener for each enzyme have been emboldened.

Enzyme (Receptor)	PDB ID	Ligand Name	PubChem CID	Known Inhibitors	Binding Affinity (kcal/mol)
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Catechin	9064	Acarbose; compound G43	-7.912
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Epicatechin	72276	Acarbose; compound G43	-8.373
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Quercetin	5280343	Acarbose; compound G43	-7.749

Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Ferulic acid	445858	Acarbose; compound G43	-6.034
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Chlorogenic acid	1794427	Acarbose; compound G43	-7.305
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Xylitol	6912	Acarbose; compound G43	-4.915
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Erythritol	222285	Acarbose; compound G43	-4.718
Glucosyltransferase B (GtfB) – <i>S. mutans</i>	3AIB	Sucrose (reference substrate/control)	5988	Acarbose; compound G43	-6.65
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Catechin	9064	Acarbose; compound G43	-7.594
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Epicatechin	72276	Acarbose; compound G43	-8.39
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Quercetin	5280343	Acarbose; compound G43	-8.238
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Ferulic acid	445858	Acarbose; compound G43	-6.55
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Chlorogenic acid	1794427	Acarbose; compound G43	-7.91
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Xylitol	6912	Acarbose; compound G43	-5.414

Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Erythritol	222285	Acarbose; compound G43	-4.547
Glucosyltransferase C (GtfC) – <i>S. mutans</i>	3AIC	Sucrose (reference substrate/control)	5988	Acarbose; compound G43	-6.676
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Catechin	9064	Oxamate (oxamic acid); oxalate	-8.145
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Epicatechin	72276	Oxamate (oxamic acid); oxalate	-8.362
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Quercetin	5280343	Oxamate (oxamic acid); oxalate	-8.666
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Ferulic acid	445858	Oxamate (oxamic acid); oxalate	-6.009
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Chlorogenic acid	1794427	Oxamate (oxamic acid); oxalate	-7.786
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Xylitol	6912	Oxamate (oxamic acid); oxalate	-4.623
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Erythritol	222285	Oxamate (oxamic acid); oxalate	-4.362
Lactate Dehydrogenase (LDH) – <i>S. mutans</i>	4LMR	Sucrose (reference substrate/control)	5988	Oxamate (oxamic acid); oxalate	-6.544
Human α -Amylase	1HNY	Catechin	9064	Acarbose; quercetin	-8.589

Human α -Amylase	1HNY	Epicatechin	72276	Acarbose; quercetin	-8.611
Human α -Amylase	1HNY	Quercetin	5280343	Acarbose; quercetin	-8.605
Human α -Amylase	1HNY	Ferulic acid	445858	Acarbose; quercetin	-6.223
Human α -Amylase	1HNY	Chlorogenic acid	1794427	Acarbose; quercetin	-7.909
Human α -Amylase	1HNY	Xylitol	6912	Acarbose; quercetin	-4.94
Human α -Amylase	1HNY	Erythritol	222285	Acarbose; quercetin	-6.271
Human α -Amylase	1HNY	Sucrose (reference substrate/ control)	5988	Acarbose; quercetin	-6.095

Table 2. The binding affinity values for each combination of the enzymes and the ligands. The best-performing polyphenol and best-performing sweetener for each enzyme are shown in bold.

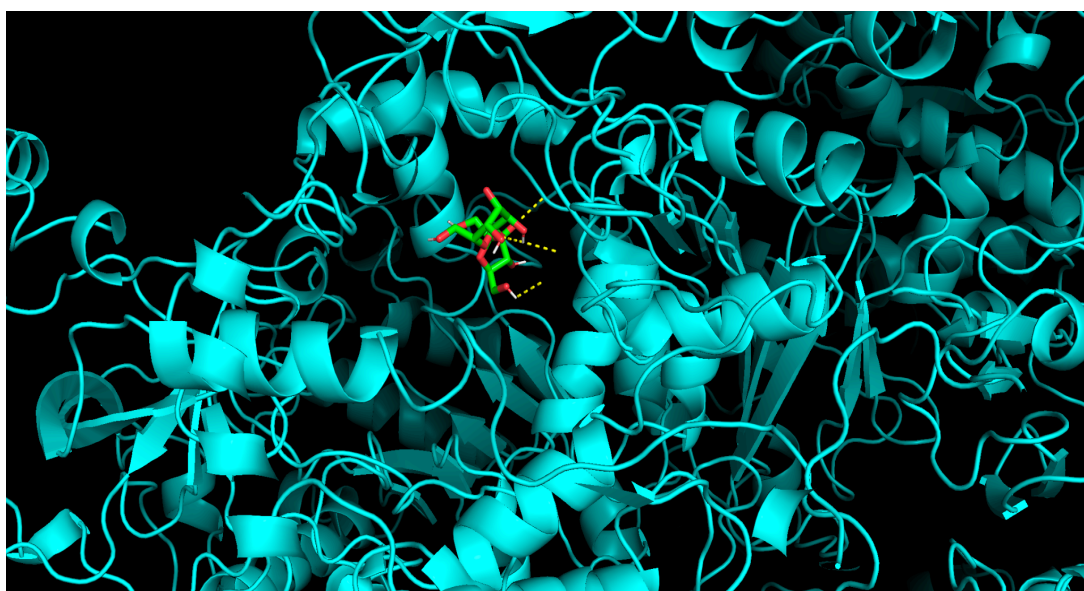


Figure 1. Molecular docking of sucrose (control) with glucosyltransferase B (GtfB). This figure shows the result of docking sucrose (ligand) with the enzyme GtfB from *Streptococcus mutans* (receptor) on Autodock Vina (Tamarind Bio). Yellow dashed lines represent the polar contacts between the ligand

and the receptor. Sucrose is the control substrate, and the model presents its binding within the active site of GtfB. This presents a reference for comparing the binding affinities and interactions of other tested compounds.

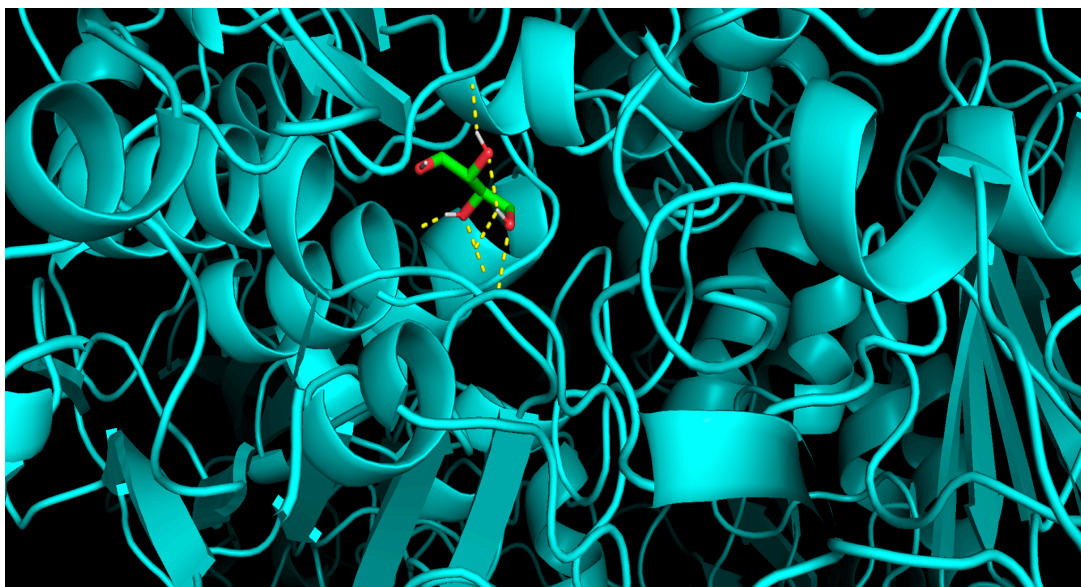


Figure 2. Three-dimensional structure of erythritol, the best-performing sweetener ligand against glucosyltransferase B (GtfB). GtfB, derived from *Streptococcus mutans* (receptor), was docked with erythritol (ligand) (PubChem CID 222285) using AutoDock Vina on the Tamarind Bio platform. Erythritol gave a predicted binding affinity of $-4.718 \text{ kcal mol}^{-1}$, the least negative value among the sweetener ligands tested, indicating the weakest predicted binding to the enzyme of all sweeteners in this study. Yellow dashed lines represent the polar contacts between the ligand and the receptor.

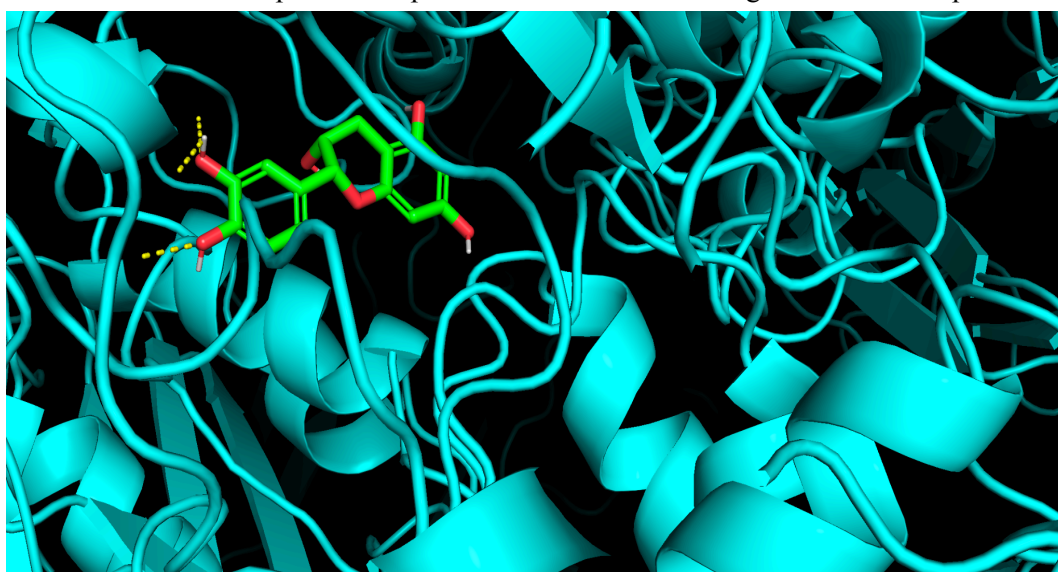


Figure 3. Three-dimensional structure of epicatechin, the best-performing polyphenol ligand against glucosyltransferase B (GtfB). GtfB, derived from *Streptococcus mutans*(receptor), was docked with epicatechin (ligand) (PubChem CID 72276) using AutoDock Vina on the Tamarind Bio platform. Epicatechin gave a predicted binding affinity of $-8.373 \text{ kcal mol}^{-1}$, the most negative value among the polyphenol ligands tested, indicating the strongest predicted binding to the enzyme of all polyphenols in this study. Yellow dashed lines represent the polar contacts between the ligand and the receptor.

Binding Analysis

The binding analysis helped in interpreting the best polyphenol and sweetener. The interpretations of binding affinity differed between polyphenols and sweeteners. Polyphenols are large, structurally complex, bioactive compounds that can modulate the oral microbiota and reduce the risk of dental caries.¹ This is enabled by their chemical structure², which includes multiple hydroxyl (-OH) groups and aromatic rings, allowing diverse intermolecular interactions with biological targets such as enzymes involved in biofilm formation. For polyphenols, a higher binding affinity is desirable because stronger interactions with enzyme active sites lead to more effective inhibition of enzymatic activities involved in biofilm formation. In contrast, sweeteners are not primary enzyme inhibitors; rather, they serve as noncariogenic sugar substrates that do not promote bacterial metabolism. Therefore, lower-binding-affinity sweeteners were considered more desirable, such as erythritol and xylitol. Among all polyphenol ligands tested, Epicatechin showed the best results for GtfB (-8.373 kcal/mol), GtfC (-7.749 kcal/mol), and Human α -Amylase (-8.611 kcal/mol), as its binding affinities for these enzymes were more negative than those of all other polyphenols. A more negative value indicates a stronger binding to the enzyme's active site. This suggests a greater potential for enzyme inhibition. In contrast, Erythritol was established as the best result for ligands derived from sweeteners due to its low binding affinity for 3 out of 4 enzymes, which include GtfB (4.718 kcal/mol), GtfC (-4.547 kcal/mol), and LDH (-4.362 kcal/mol).

Identification of food sources for polyphenols and sweeteners

Plant-based food sources for each ligand were investigated. Unique sources and sources which were common to more polyphenols were identified. For unique sources, it was found that Catechin is present in black tea, Epicatechin in cocoa, Quercetin in onions, apples, and berries, Ferulic acid in whole grains like rice bran, wheat, and oats, and chlorogenic acid in potatoes. Common food sources which contained more than one type of polyphenol ligand were green tea and dark chocolate, which contain catechin and Epicatechin; berries and apples, which contain quercetin and flavonoids like Catechin and Epicatechin. Finally, coffee, which contains chlorogenic acid and ferulic acid. The same was done for sweeteners. Xylitol is present in birch bark, corn cobs, strawberries, raspberries, plums, cauliflower, pumpkin, spinach, and lettuce, while Erythritol is found in grapes, pears, melons and watermelons. Mushrooms are the only food which contains a small amount of Erythritol and Xylitol.

Since polyphenols are widely present in plant sweeteners like Xylitol and Erythritol occur only in trace amounts naturally, there is a very small overlap in the foods which contain both polyphenols and

sweeteners. This overlap includes fruits like grapes, pears and watermelon, which contain a significant amount of polyphenols but only traces of erythritol, which is possible via fermentation or natural metabolism of the sugars in the fruit.

However, the possibility of the presence of the polyphenols and sweeteners identified in this study is much higher in processed foods. Based on these results, all these sources can be combined to develop functional recipes that may help prevent biofilm formation and acid production. To elaborate, non-cariogenic sweeteners, like Xylitol and Erythritol, can be used as sugar substitutes when integrated with enzyme-inhibiting, polyphenol-rich foods.

DISCUSSION

To predict binding affinity values as an indicator of potential enzyme inhibition, molecular docking was performed. The process started by selecting enzymes and downloading their respective downloads from the PDB database. These were enzymes produced by *S.mutans*, which are commonly found in the oral microbiome. These included Glucosyltransferase C, Glucosyltransferase B, Lactate Dehydrogenase, and Human α -Amylase. The ligand structures were downloaded from the PubChem database and comprised 2 categories: polyphenol ligands (Catechin, Epicatechin, Quercetin, Ferulic acid, Chlorogenic acid) and sweetener ligands (Xylitol, Erythritol, and Sucrose [control]). Docking was performed using AutoDock Vina, with Tamarind Bio used to determine binding affinity. The binding affinity of each ligand was analysed to determine the best result. It was predicted that Epicatechin was the best-performing polyphenol due to the most negative binding affinity (for all enzymes except Lactate Dehydrogenase). In contrast, Erythritol was predicted as the best-performing sweetener due to the least negative binding affinity (for all enzymes except Human α -Amylase). The binding affinity of sucrose stayed relatively consistent across GtfB, GtfC, LDH, and Human α -Amylase: -6.65 kcal/mol, -6.676 kcal/mol, -6.544 kcal/mol, -6.095 kcal/mol, respectively. The high binding affinity of epicatechin and, conversely, the relatively low binding affinity of erythritol suggest a greater likelihood of interaction with enzymes involved in biofilm formation, as they may be able to interact with enzymes involved in biofilm formation effectively.

After reviewing the literature, it was understood that catechin derivatives are considered strong inhibitors of enzyme activity due to their ability to form stable enzyme-ligand complexes. This is possible through hydrogen bonding and van der Waals interactions, which often lead to conformational changes and thus inhibit enzyme activity.² Therefore, the known structural and biochemical properties of polyphenols are consistent with the strong binding affinity of epicatechin observed in this study. On the other hand, sugar alcohols like erythritol and xylitol are structurally simpler molecules and highly polar. They are used more commonly as non-nutritive sweeteners. These compounds can influence microbial biofilm formation while reducing bacterial adhesion and growth compared to sucrose.³ In this study, erythritol showed a more favourable predicted binding affinity than xylitol, due to lower binding affinity values for GtfB, GtfC, and LDH. However, previous studies have reported that xylitol may be a stronger inhibitor than erythritol due to its lower binding affinity for certain carbohydrate-digesting enzymes.⁴ This

contrasts with the results of this study. This discrepancy may be because other studies employed some in vitro experiments too, whereas this study relies solely on in silico experiments using AutoDock Vina on Tamarin Bio. This helps make a theoretical prediction about the binding of the enzyme with the ligands. As a result, variations in binding affinities may arise due to differences in predicted interactions and actual biological interactions.

FUTURE WORK AND LIMITATIONS

Several limitations can arise from the docking method itself. Firstly, the receptors were treated like rigid, hence induced-fit effects and side-chain rearrangement on ligand bonding could not be modelled. Also, the docking was performed without any explicit solvent, suggesting that water-mediated hydrogen bonds and solvation effects were not accounted for. No molecular dynamics simulations were performed so the stability of the predicted poses over time could not be assessed.

Structure preparation was performed automatically by the Tamarind Bio platform, and the exact handling of hydrogens, partial charges and non-protein heteroatoms during PDBQT conversion is not publicly documented. Protonation states were therefore not assigned at a defined pH, and the acidic conditions characteristic of cariogenic plaque were not represented in the model.

The findings displayed in this study are solely computational predictions : they have not been experimentally validated. Molecular docking simulations can estimate the affinity with which a ligand binds a target, but does not account for or measure multiple the dynamic nature of biological interactions like enzyme activity, bacterial growth, or any physiological outcome. The absence of experimental validations through in vivo studies or in-vitro enzyme inhibition assays makes it hard to determine the actual effectiveness of the results of this study.

Therefore, the food reformulation strategies proposed in this study should therefore be regarded as hypotheses generated through computational screening as compared to evidence-based dietary recommendations. A staged programme of experimental work would be essential before their production could support such recommendations.

First, in-vitro enzyme inhibition assays need to be done to confirm whether the predicted binding translated into functional inhibition, with IC₅₀ values found for each compound against purified glucosyltransferase, lactate dehydrogenase and α -amylase. Further, comparing the resulting potency ranking against the predicted affinity will also indicate how well the docking model in this study performed.

Next, microbial assays would test whether compounds affect *S. mutans* in culture. This can be done by measuring growth inhibition and , separately, biofilm formation. Measuring both separately prevents any confusion since a compound may reduce one biofilm formation without affecting growth inhibition (or the other way around).

Third, pH kinetics studied could measure the acid produced in *S. mutans* cultures or biofilm models following sucrose challenge. This could help establish whether the compounds reduce the pH drop associated with enamel demineralisation.

Only once these stages are complete, the results can be translated into justified dietary recommendation or oral health. Further work on bioavailability, stability during food processing and effective concentration in the oral cavity would also be needed then. Expanding the range of enzymes and ligands analysed could help create a more comprehensive understanding of the anti-cariogenic agents.

CONCLUSION

Conclusively, the results of this study provide predictions about the interactions between enzymes and ligands to indicate which compounds may have potential anti-cariogenic properties, based on predicted binding to cariogenic enzymes. The results are consistent with epicatechin and erythritol as candidate compounds for further investigation into biofilm reduction. Together, these findings suggest that dietary habits can influence the activity of the oral microbiome at the molecular level. Experimental validation, for example through enzyme inhibition assays measuring IC₅₀ values, would be required to confirm whether the predicted binding affinities translate into functional inhibition. Moreover, the study investigates common food sources that contain beneficial ligands that can be used as ingredients to create functional recipes/ingredient lists that do not promote biofilm or caries formation. Finally, the outcomes of this study may inform future development of oral care products, subject to experimental validation. Overall, the research underscores the importance of combining biochemical analysis with nutritional choices to support improved oral health outcomes.

References

1. [Dental Caries \(Tooth Decay\) in Adults \(Ages 20 to 64 Years\).](https://www.nidcr.nih.gov/research/data-statistics/dental-caries/adults)
<https://www.nidcr.nih.gov/research/data-statistics/dental-caries/adults>.
2. [Chen, X. *et al.* Microbial Etiology and Prevention of Dental Caries: Exploiting Natural Products to Inhibit Cariogenic Biofilms. *Pathogens* **9**, 569 \(2020\).](#)
3. [Dental caries. *The Lancet* **369**, 51–59 \(2007\).](#)
4. [Tuloglu, N., Bayrak, S., Tunc, E. S. & Ozer, F. Effect of fluoride varnish with added casein phosphopeptide-amorphous calcium phosphate on the acid resistance of the primary enamel. *BMC Oral Health* **16**, 103 \(2016\).](#)
5. [Lactose triggers biofilm formation by *Streptococcus mutans*. *International Dairy Journal* **42**, 51–57 \(2015\).](#)
6. [Horst, J. A., Tanzer, J. M. & Milgrom, P. M. Fluorides and Other Preventive Strategies for Tooth Decay. *Dental clinics of North America* **62**, \(2018\).](#)

7. [Spatafora, G., Li, Y., He, X., Cowan, A. & Tanner, A. C. R. The Evolving Microbiome of Dental Caries. *Microorganisms* **12**, 121 \(2024\).](#)
8. [Bradshaw, D. J. & Lynch, R. J. M. Diet and the microbial aetiology of dental caries: new paradigms. *International Dental Journal* **63**, 64 \(2020\).](#)
9. [Yu, S. *et al.* Pyruvate metabolism is involved in adaptability and cariogenicity of *Streptococcus mutans*. *Journal of oral microbiology* **18**, \(2026\).](#)
10. [Durso, S. C. *et al.* Sucrose substitutes affect the cariogenic potential of *Streptococcus mutans* biofilms. *Caries research* **48**, \(2014\).](#)
11. [Najarzadeh, Z. *et al.* Plant Polyphenols Inhibit Functional Amyloid and Biofilm Formation in *Pseudomonas* Strains by Directing Monomers to Off-Pathway Oligomers. *Biomolecules* **9**, \(2019\).](#)
12. [A novel biomimetic strategy for mimicking amelogenesis to repair enamel. *Dental Materials* **41**, 513–522 \(2025\).](#)
13. [Guanidination carbon dots combat *Streptococcus mutans* biofilm through downregulating carbohydrate metabolism to prevent dental caries. *Carbon* **247**, 120980 \(2026\).](#)
14. [Guo, Y., Li, Z., Chen, F. & Chai, Y. Polyphenols in Oral Health: Homeostasis Maintenance, Disease Prevention, and Therapeutic Applications. *Nutrients* **15**, 4384 \(2023\).](#)
15. [Zhu, M., Pan, J., Hu, X. & Zhang, G. Epicatechin Gallate as Xanthine Oxidase Inhibitor: Inhibitory Kinetics, Binding Characteristics, Synergistic Inhibition, and Action Mechanism. *Foods* **10**, 2191 \(2021\).](#)
16. [Jeong, G.-J., Khan, F., Tabassum, N. & Kim, Y.-M. Alteration of oral microbial biofilms by sweeteners. *Biofilm* **7**, 100171 \(2023\).](#)
17. [Comparative effects of xylitol and erythritol on inhibition of key digestive enzymes and modulating dysfunctional enzyme activities in oxidative hepatic injury. *Scientific African*, **29**, e02911 \(2025\).](#)

ABOUT THE AUTHOR

Saisha Sikka is a high school senior at Bombay International School in Mumbai, India. She enjoys learning biology and medical biosciences, hoping to explore these subjects in a STEM-related degree in college too.