Vanillin attenuates AGEs, Glycation-Induced Oxidation & Aggregation of Serum Transferrin: Insights from biophysical and *in silico* studies

Bhuvana Kumar, Saraswathi N.T.

Molecular Biophysics lab, School of Chemical and Biotechnology,

SASTRA Deemed to be University, Thanjavur-613401.

(Presenting author Email: bhuvana@scbt.sastra.ac.in)

Abstract

Hyperglycemic conditions observed during diabetes results in the formation of advanced glycation endproducts (AGEs) by the non enzymatic modification termed glycation. AGEs are the covalent adducts formed between carbonyl groups of sugars and free amino groups of proteins. AGEs formed play multiple roles in progression and severity of diabetic complications. They also lead to increased oxidative stress and activation of inflammatory pathways. Transferrin, the major serum glycoprotein involved in transport of iron is reported to undergo variable glycation in diabetic patients resulting in impaired iron binding capacity. Reported AGE inhibitors such as aminoguanidine, are limited in their use owing to toxicity in clinical trials. Natural compounds showing reduced toxicity, are highly suitable to explore their role as anti-glycation agents. Here, we report the glycation inhibition observed in glycated transferrin by vanillin (used as flavoring agent) which is a phenolic aldehyde present at 1-2% (w/w) in cured vanilla beans. Quantification of AGEs fluorescence showed that vanillin hindered AGE formation with increased reduction in vesperlysine and total AGEs. The presence of vanillin curtailed oxidative damage addressed by reduced fluorescent oxidative markers such as dityrosine, kynurenine and N'-formyl-l-kynurenine. It also reduced glycation induced fibrillation further confirmed by reduced ThT bound fibrils in glycated transferrin with vanillin. The extent of arginine modification was also reduced in the presence of vanillin. Vanillin binds transferrin exhibiting a binding affinity of 0.512623x 104 M-1 with one binding site as assessed by fluorescence quenching studies. *In silico* docking revealed binding of vanillin in the C-lobe of transferrin. Molecular dynamics simulation supported the stability of the complex and continuous interaction between vanillin and transferrin. MM PBSA binding energy calculations revealed the electrostatic energy as the highest contributor. Thereby, vanillin exerts antiglycative potential against glycated transferrin and prevents subsequent oxidative damage and aggregates formation.

Keywords: Advanced Glycation Endproducts; Vanillin; Fibrillation