Quantum-chemical and docking analysis on the binding potential of hydroxybenzoic acids from *Graptopetalum paraguayense* E. Walther to HSV thymidine kinase active site

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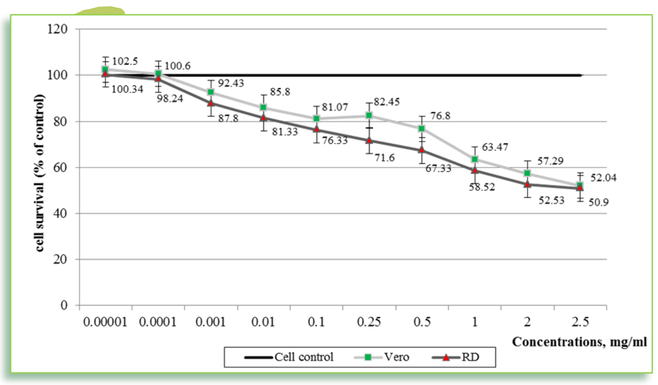
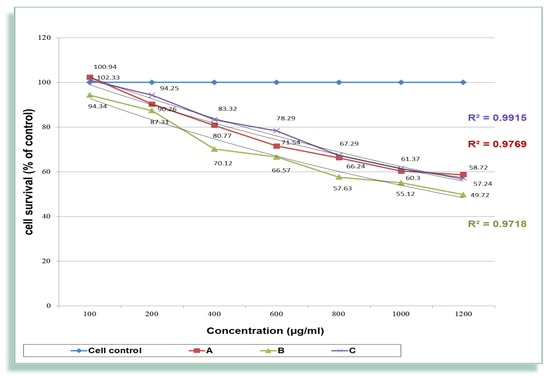
The total methanol extract and phenolic fraction from the plant *Graptopetalum paraguayense* demonstrates a significant inhibitory effect on HSV-1. Since thymidine kinase appears to be a key feature in the replication of HSV DNA virus, we present a theoretical study on the binding expedient of phenols from this fraction to viral DNA polymerase amino acids. Twelve different hydroxybenzoic acids were found by GC/MS analyses. MOE 2016 software package was used to dock selected structures in the active site defined in published X-ray diffraction structures of the HSV 1 DNApol. The structure was protonated and scored by implemented GBVI/WSA dG scoring function. According to this function, trans-ferulic and gentisic acids have optimal interactions with the receptor. Some hydrogen-bonded complexes between phenolic and amino acids at B3LYP/6-31+G(d,p) level were modeled. The received data suggest that all phenolic acids could form stable complexes with amino acids from the DNA polymerase active site.

In vitro anti-herpetic and cytotoxic activity of the GP extracts



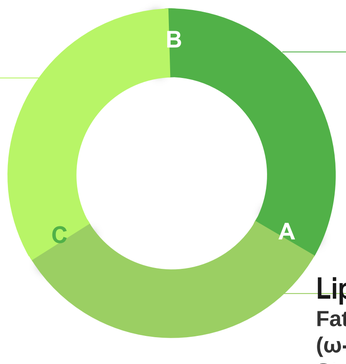
Cytotoxic activity of GP total extract on the viability of Vero and RD cell lines at 72 h treatment.



Cytotoxicity of fractions A, B and C of G. paraguayense E. Walther on green monkey kidney cell line (Vero).

Fractions C and A have no CPE on green monkey kidney cell line (Vero) and inhibited HSV replication in dose-dependent manner more efficiently against HSV-1, whereas their effect on HSV-2 was significantly lower. B fraction showed no antiviral effect.

Main metabolites in *G. paraguayense*



Phenolic Acids
Syringic acid
Gallic acid
trans-Ferulic acid
Salicylic acid
Vanillic acid
Gentisic acid
Protocatechuic acid
p-Coumaric acid
and others

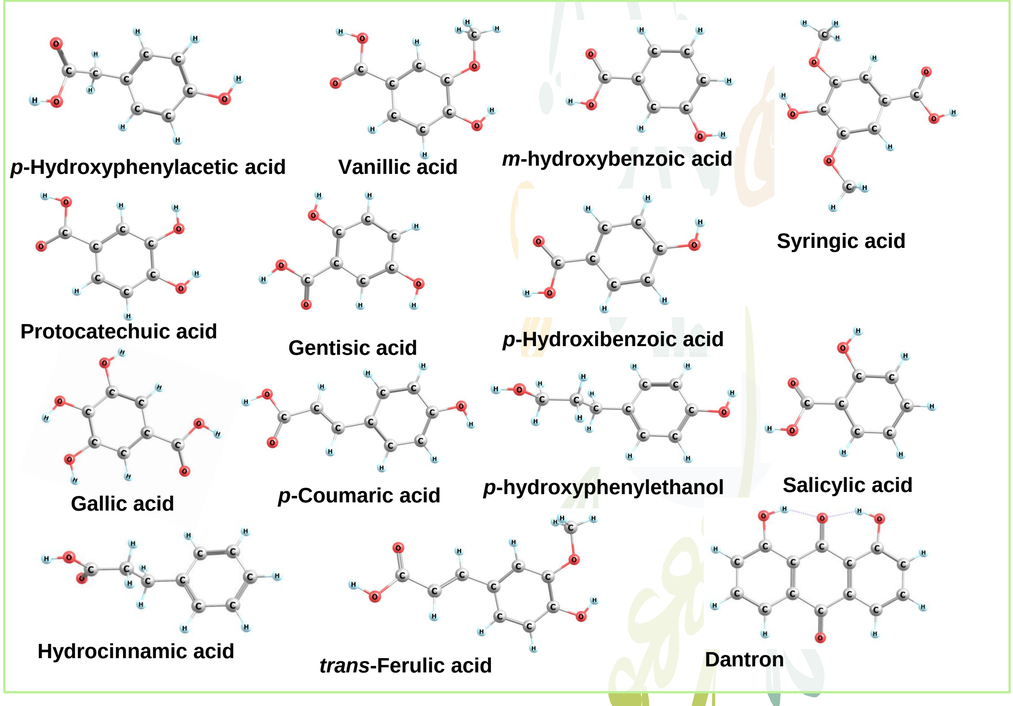
Polar Metabolites
Amino acids
Other organic acids
Sugars

Lipids
Fatty acids
(ω-3, ω-6, ω-7, ω-9)
Sterols and terpenoids
α-Tocopherol, β-Sitosterol,
β-Amyrin, Lanosterol

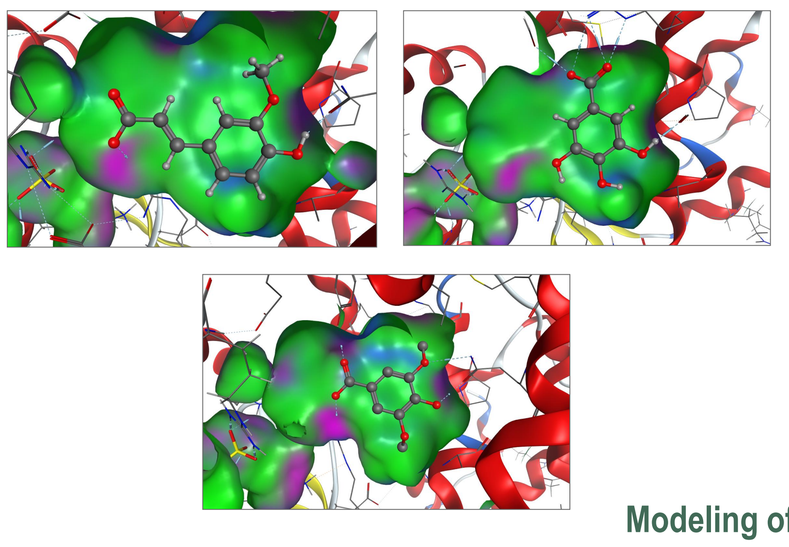
Inhibition of the replication:

ACV sensitive strains	ACV resistant strains
Victoria (HSV-1)	***DD (HSV-1)***
GP extract – **97.5%**	GP extract - **65.5%**
ACV – 100%	ACV – 10.8%
Bja (HSV-2)	***PU (HSV-2)***
GP extract – 25.5%	GP extract – **13%**,
ACV – 95.7%	ACV – 0%

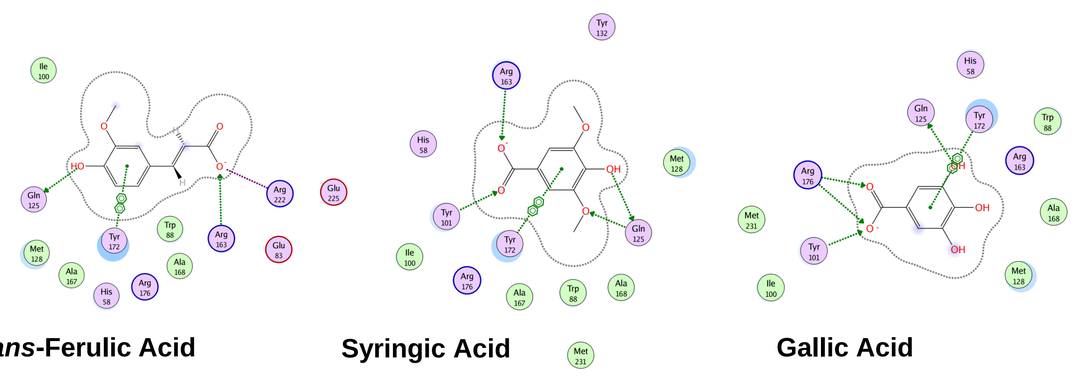
Hydroxybenzoic acids structures from GP extract optimized at B3LYP level



Hydroxybenzoic acid interacting with HSV-1 TK active site



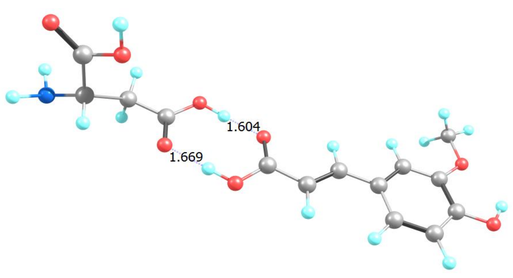
Fitting of phenolic acids in the HSV-1 TK pocket



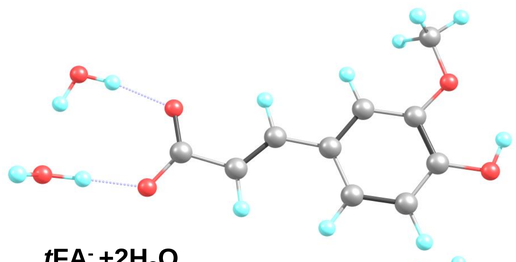
trans-Ferulic Acid | Syringic Acid | Gallic Acid

HSV thymidine kinase (TK) catalyses the transfer of the gamma-phosphate group of ATP to thymidine to generate dTMP in the salvage pathway of pyrimidine synthesis. The dTMP serves as a substrate for DNA polymerase during viral DNA replication. TK allows the virus to be reactivated and to grow in non-proliferative cells lacking a high concentration of phosphorylated nucleic acid precursors. Thus, TKs are the main targets in anti-herpes virus treatments and potential therapeutic targets in antitumor gene therapy strategies.

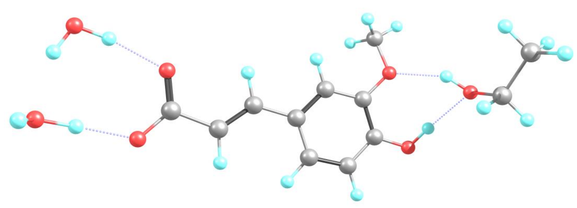
Modeling of phenolic acids-HSV-1 TK amino acids complexes



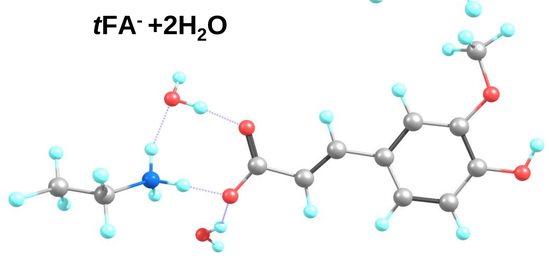
*t*FA+ Asp The length of the intermolecular hydrogen bonds is in Å. E_{int}= 16.93 kcal.mol^{-1}



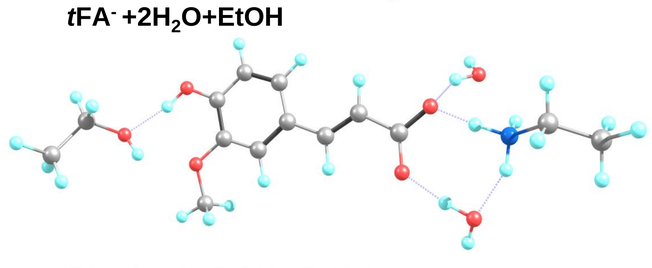
*t*FA$^-$ +2H_2O



*t*FA$^-$ +2H_2O+EtOH



tFA$^-$ +2H_2O+ EtAH$^+$



*t*FA$^-$ +2H_2O+EtOH+ EtAH$^+$

trans-Ferulic acid complexes, optimized at B3LYP/6-31+G(d,p) level.

Structures of the *trans*-Ferulic acid complexes calculated at B3LYP/6-31+G(d,p) level

$$E_{int} = (E_{water} + E_{GA^-} + E_{EtOH} + E_{EtAH^+}) - E_{complex}$$

$$E_{int} = 34.27 \text{ kcal.mol}^{-1}$$

*t*FA forms stable hydrogen-bonded complex with HSV-1 TK active site residues. Therefore, the complex formed is stable and *trans*-Ferulic acid demonstrates great binding affinity to the active site of NSV-1 TK where it can exhibit its inhibitory properties.

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