

Supplementary Data

Title: Cytoprotective effect exerted by geraniin in HepG2 cells is through microRNA mediated regulation of BACH-1 and HO-1

Author's name: Hoda Aayadi^a, Smriti P.K. Mittal^{a1}, Anjali Deshpande^b, Makarand Gore^b, Saroj S. Ghaskadbi^{a*}

Affiliation: ^aDepartment of Zoology, Savitribai Phule Pune University, Pune – 411007, India ^bAJ Organica Pvt. Ltd., Pune – 411007, India

¹Present address: Department of Biotechnology, Savitribai Phule Pune University, Pune – 411007, India

Running Title: Regulation of HO-1 induction by geraniin

Keywords: Geraniin; Antioxidants; HO-1; Nrf-2; miRNAs

Corresponding Author's Information: Tel:+91-9850993508; Fax:+91-20-25690617; E-mail: ssg@unipune.ac.in

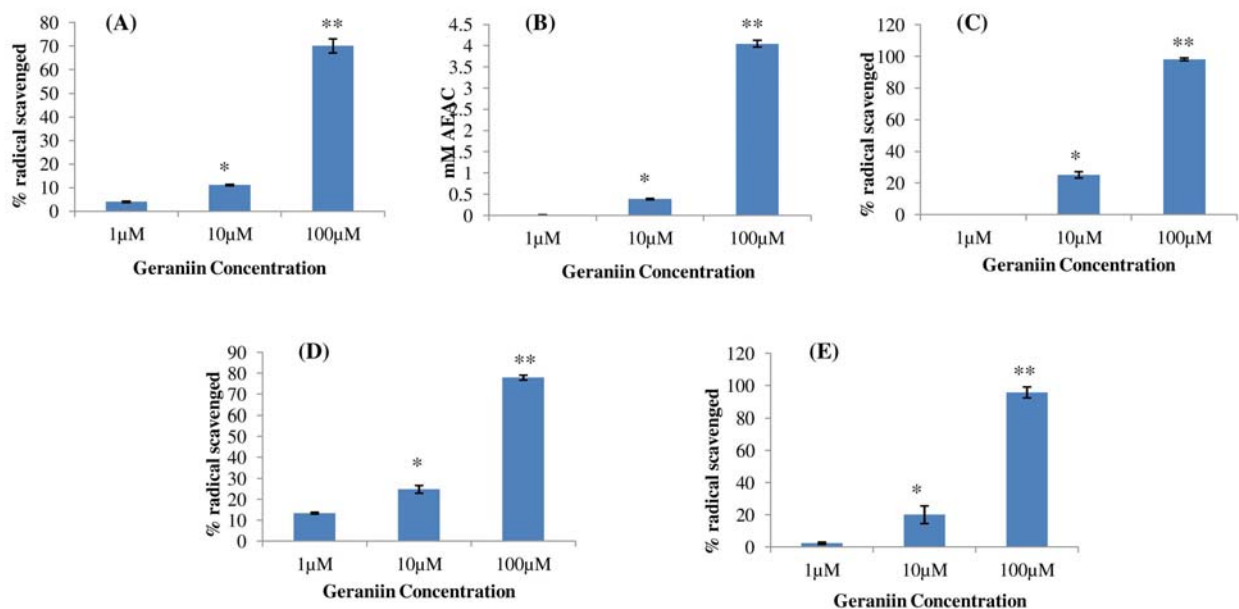


Figure 1: Radical scavenging by Geraniin. (A) DPPH, (B) FRAP Assay, (C) Superoxide radical, (D) Hydroxyl radical and (E) H₂O₂ assay

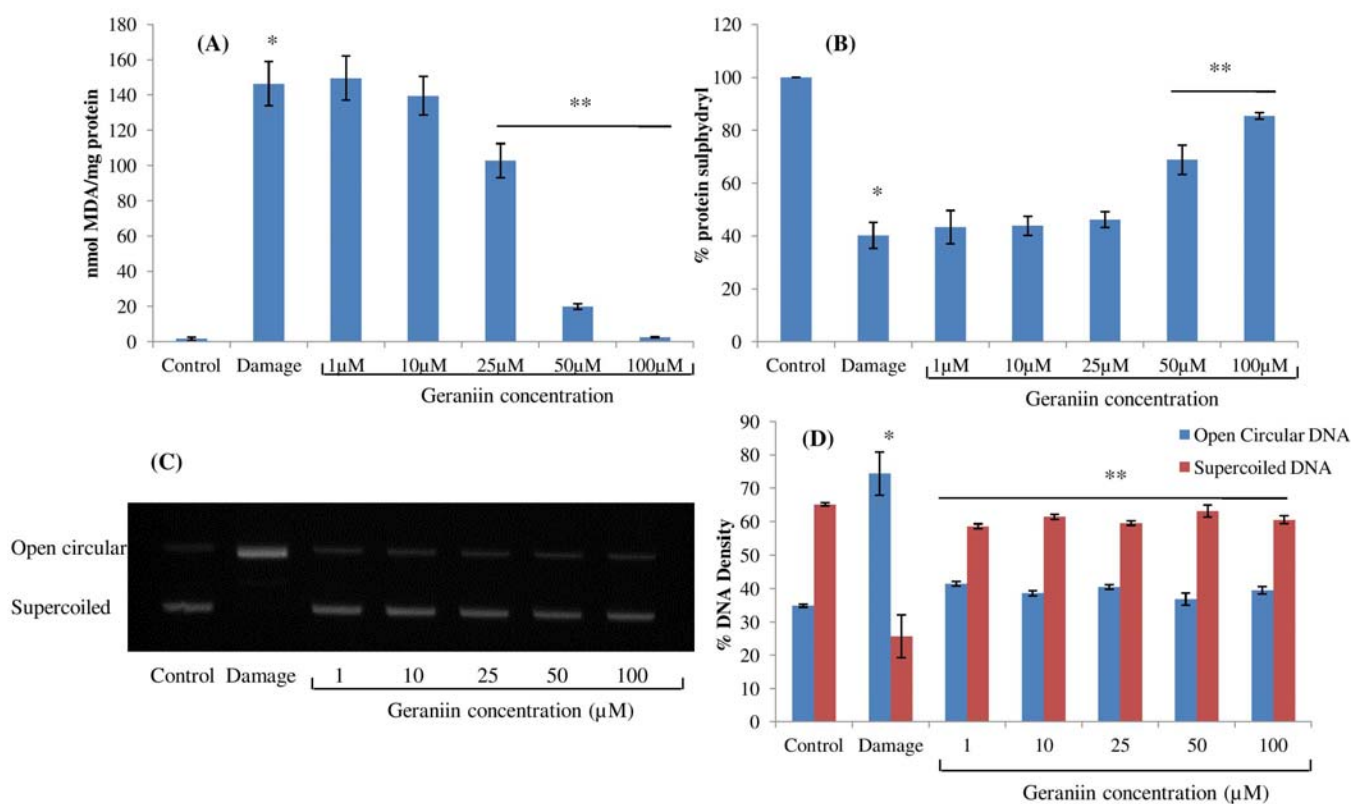


Figure 2: Protective effect of Geraniin against oxidatively damaged biomolecules. (A) Inhibition of lipid peroxide formation and (B) Restoration of protein sulphhydryl by Geraniin in oxidatively damaged rat liver mitochondria. (C) and (D) Gel electrophoresis pattern of pBR322 plasmid DNA and its densitogram analysis after induction of oxidative damage in the presence of different concentrations of Geraniin. Data shown represent the mean values of three experiments \pm SE. * p < 0.05 vs. control. ** p < 0.05 vs. damage.

Table 1: Primers used for the amplification of gene specific qRT-PCR products.

Gene	Primer Sequence
CAT	Forward:5'-TGCTGAATGAGGAACAGAGG-3' Reverse:5'-GTGTGAATCGCATTCTTAGG-3'
GPx	Forward:5'- GCGGCGGCCAGTCGGTGTA -3' Reverse: 5'- GAGCTTGGGGTCGGTCATAA -3'
SOD	Forward:5'- TGTGGCCGATGTGTCTATTG -3' Reverse: 5'- GCGTTTCCTGTCTTTC -3'
HO-1	Forward:5'- ACTGCGTTCCTGCTCAACAT -3' Reverse: 5'- GGGCAGAATCTTGCACTTT -3'
BACH-1	Forward:5'- TGCGATGTCACCATCTTTGT -3' Reverse: 5'- CCTGGCCTACGATTCTTGAG -3'
GAPDH	Forward: 5'- TCGACCACCGACTGCTTAGC -3' Reverse: 5'- GGGATGGACTGTGGTTATG -3'
Let7b	Stem-loop: 5'- GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACAACCAT -3' Forward:5'- GCGCATTGAGGTAGTAGGTTGTGT -3'
Let7c	Stem-loop: 5'- GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACAACCAT -3' Forward:5'- GCGCATTGAGGTAGTAGGTTGTAT -3'
miR-98	Stem-loop: 5'- GTCGATTCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACAACAAT -3' Forward:5'- GCGCCATTGAGGTAGTAAGTTGTA -3'
miR-196	Stem-loop: 5'- GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACCCCAAC -3' Forward:5'- GCGCATTAGGTAGTTTCCTGTTGT -3'
miR-217	Stem-loop: 5'- GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACTCCAAT -3' Forward:5'- GCCATTACTGCATCAGGAACTGA -3'
miR-377	Stem-loop: 5'- GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACACAAAA -3' Forward:5'- GCGAATATCACACAAAGGCAACT -3'