Abstract

The CoronaVirus Disease 2019 (COVID-19) caused by SARS-CoV-2 continues to be a global pandemic with substantial socioeconomic damages. Exploration of bioactive compounds from herbal resources with potential anti SARS-CoV-2 effects are currently on the rise. In particular, naturally-derived flavonoids are reported for its capability to inhibit 3-chymotrypsin like-protease ($3CL^{pro}$), a promising drug target that is essential for SARS-CoV-2 replication and formation. In this paper, the flavonoid compounds in *Foeniculum vulgare* semen (Fennel seed) ethanolic crude extract are identified using UPLC-ESI-MS/MS, followed by binding affinity prediction of the identified flavonoid compounds toward SARS-CoV-2 $3CL^{pro}$ crystal structure through *in silico* approaches, before ultimately assessing the inhibition level of the extract towards recombinant SARS-CoV-2 $3CL^{pro}$ *in vitro*. The results indicated that *F. vulgare* seed extract contains the flavonol class of flavonoids, specifically quercetin, kaempferol, and its glycosylated moieties which are well documented by previous studies to exert SARS-CoV-2 $3CL^{pro}$ inhibitory effects. Molecular docking analysis revealed strong binding affinity values of <-7 kcal/mol for all identified flavonols, with glycosylated moieties displaying a more negative value than aglycone flavonols. Quercetin-3-O-rutinoside exhibited the highest binding affinity of -8.8 kcal/mol, and most flavonols have interactions with the Cys145 and Glu166 active site residues of SARS-CoV-2 $3CL^{pro}$ crystal structure (6LU7). *F. vulgare* seed extract is also revealed to successfully inhibit SARS-CoV-2 $3CL^{pro}$ with a half-maximum inhibitory concentration (IC_{50}) of 58.55 ± 7.29 ppm. Overall, the findings are able to provide important insights for further research and innovation of novel anti SARS-CoV-2 drugs.

Keywords: $3CL^{pro}$, Anti SARS-CoV-2, COVID-19, Fennel Seeds, Flavonols