Damanhour Journal of Veterinary Sciences 12 (2), (2024) 55-63

Damanhour Journal of Veterinary Sciences

Journal homepage: https://djvs.journals.ekb.eg/

E-ISSN 2636-3003 | ISSN 2636-3011

Anti-aging Effects of Natural Bioactive Compounds by Target mTOR: Molecular Docking Approach

Ali H. El-Far[1, *], Abdelwahab A. Alsenosy[1], Shymaa M. Mokadem[1], Manar A. Elshrief[1], Shaimaa M. Saafan[1, 2], Doaa A. Elsabagh[1], Hadeer H. Mohamed[1], Mona M. Elghaity[1], Shymaa A. Mohamed[3]

[1]Department of Biochemistry, Faculty of Veterinary Medicine, Damanhour University، Damanhour 22511, Egypt
[2]Department of Biochemistry, Animal Health Research Institute (AHRI), Damanhour Branch, Agriculture Research Centre (ARC) Damanhour 22511, Egypt
[3]Molecular Biology, Molecular Biology Unit, Medical Technology Centre, Medical Research Institute, Alexandria University, Egypt

Abstract:

mTOR pathway is involved in the aging process but the understanding of all mechanisms remains needs more extensive research. In the current study, we investigated the molecular docking interactions of catechin, caffeic acid, carvacrol, diosgenin, eugenol, and rutin against mTOR. Results indicated the ability of investigated natural bioactive compounds to interact with mTOR with binding energy of -5.68, -4.87, -4.57, -7.34, -7.34, -9.72 kcal/mol. More in vitro and in vivo experiments are extremely needed to understand the mechanistic role of these natural bioactive compounds to hinder mTOR's aging process participation.

Keywords: Aging; mTOR; Natural bioactive compounds; Molecular docking

*Correspondence: **Ali H. Elfar**
Department of Biochemistry, Faculty of Veterinary Medicine, Damanhour University، Damanhour 22511, Egypt
; Email: ali.elfar@damanhour.edu.eg
P ISSN: 2636-3003
EISSN: 2636-2996
DOI: 10.21608/djvs.2024.302055.1133
Received: July 6, 2024; Received in revised form: July 16, 2024; accepted: July 18, 2024
Editor-in-Chief: Ass. Prof .Dr/Abdelwahab A. Alsenosy (editor@vetmed.dmu.edu.eg)

1. Introduction

The aging physiologic and pathologic process is marked by a gradual deterioration of cellular functioning, facilitated by modifications in several molecular pathways that heighten the vulnerability of the cell to harm. This decrease is a major risk factor for several major human disorders (Tenchov et al. 2024). Numerous cellular processes are common to the aging process in many different organisms, including mitochondrial dysfunction, exhaustion of stem cells, telomere erosion, epigenetic modifications, deregulated nutrition-sensing, and altered intercellular signal transduction (Kennedy et al. 2014).

The nutrient-sensing protein kinase called mammalian target of rapamycin (mTOR) controls the growth and metabolism of all eukaryotic cells (Yang et al. 2008). The notion that the mTOR signaling network is crucial for regulating aging is supported by research done on mice, worms, yeast, and flies. There is evidence that the preventive benefits of various dietary restriction strategies, which have been shown to extend animal lifespans and postpone the onset of age-related disorders, are mostly mediated by mTOR (Stallone et al. 2019).

Cellular senescence, an irreversible cell cycle termination, is considered a major tumor suppression mechanism in conventional wisdom (López-Otín et al. 2013). This implies that treating cancer and delaying aging may be accomplished by focusing on mTOR. Although the mechanism of mTOR inhibition for anti-aging or cancer therapy is complicated and sometimes confusing, it is believed that mTOR inhibition generally has favorable effects for anti-aging and anti-cancer (Kim et al. 2017). Because of its remarkable ability to control a wide range of important cellular activities, the mTORC1 signaling pathway is likely important in cellular senescence. The mTOR signaling pathway can be made to act differently by a polyphenol. Furthermore, a key component of aging is cellular senescence, the underlying cause of age-related disorders. Polyphenols have anti-aging characteristics because they can slow down important signaling pathways, such the mTOR signaling pathway, that affect an organism's longevity (Weichhart 2018).

Caffeine is a member of the phenolics class and has methoxy and hydroxyl groups on its structure (Khan et al. 2016). Rutin is a member of the flavonoid glycoside group, which is also known as vitamin P. Its ability to scavenge free radicals suggests that it might have antiviral and antihypertensive qualities (Khomsi et al. 2022). Rhodanase, tyrosinase, elastase, and collagenase were all suppressed by rutin and caffeic acid, indicating their anti-aging characteristics. The capacity of rutin and caffeic acid, which are found in snake fruit peels, to bind to the proteins that promote aging was validated by a prior study (Saafan et al. 2023; Widowati et al. 2023).

Phosphorylation of Ser2448 and auto-phosphorylation of Ser2481 in the mTOR protein occur via the PI3 kinase/Akt signaling pathway (Huang and Houghton 2003). While phospho-mTOR Ser2481 and phospho-p70S6kinase, a downstream signaling protein of the mTOR pathway, were significantly decreased by catechin treatment, mTOR and phosphor-mTOR Ser2448 were only slightly reduced (Chung et al. 1992; Kuo et al. 1992). As mTOR and p70 S6 kinase control the production of new proteins, maintain the equilibrium of food absorption, control the levels of ATP and amino acids in the body, and control the growth of cells (Gingras et al. 2001; Lee et al. 2009).

Natural bioactive compounds of antioxidant potential are extremely beneficial as anti-aging nutraceuticals. Therefore, the current study has been assigned to determine the probability of molecular interactions of catechin, caffeic acid, carvacrol, diosgenin, eugenol, and rutin against mTOR.

2. Materials and Methods

2.1. Ligands preparation

The three-dimensional structures of catechin, caffeic acid, carvacrol, diosgenin, eugenol, and rutin were retrieved from PubChem (https://pubchem.ncbi.nlm.nih.gov/) database in SDF format. Ligands energy minimization and docking with target proteins were done using The MOE 2015.10 (Salim and Noureddine 2020) software.

2.2. Protein preparation

The three-dimensional structures of rats' mTOR was retrieved from UniProt database (https://www.uniprot.org/). Target proteins were prepared for docking using MOE software along with target protein energy minimization.

2.3. Molecular docking analysis and visualization

mTOR was docked with ligands using MOE software through identification of binding site and molecular docking.

Finally, the protein-ligand interactions were visualized by MOE.

3. Results and Discussion

As we thoroughly investigated the experimentally induced ageing by subcutaneous or intraperitoneal injections that generated the identical aspects of natural ageing, the current investigation was conducted on the mTOR of rats (El-Far et al. 2020, 2021, 2022, 2024; Saafan et al. 2023).

Results represented in **Figure 1** revealed the molecular interaction of catechin against mTOR with binding free energy of -5.68 kcal/mol whereas catechin bound with ASP911 (H-donor) and GLN1937 (pi-H) residues.

Found in a wide range of fruits, vegetables, and plant-based beverages, catechin belongs to the flavonoid family of polyphenolic chemicals, or flavonols (Braicu et al. 2013).Catechin was shown to be able to bind mTOR and reduce ageing simultaneously by using molecular docking models. Although catechin is a metal ion chelator and a scavenger of reactive oxygen species (ROS), its indirect antioxidant actions include inducing antioxidant enzymes, which may account for some of its anti-aging properties (Bernatoniene and Kopustinskiene 2018).

Caffeic acid is an organic antioxidant compound that prevents oxidative stress alterations in the body. It is found naturally in a wide range of plants (especially in coffee) (Gülçin 2006; Khoshdel et al. 2022). As represented in **Figure 2**, caffeic acid interacted with mTOR by binding free energy of -4.87 kcal/mol indication its role as anti-aging compound. Also, *in vivo* studies evidenced the anti-aging effect of caffeic acid against testicular damages in mice (Khoshdel et al. 2022) and memory and hippocampal neurogenesis deficits in aging rats induced by D-galactose (Saenno et al. 2022).

Carvacrol is a phenolic monoterpenoid found in essential oils of oregano (*Origanum vulgare*), thyme (*Thymus vulgaris*), pepperwort (*Lepidium flavum*), wild bergamot (*Citrus aurantium bergamia*), *Nigella sativa*, and other plants with a wide range of bioactivities (Sharifi-Rad et al. 2018). In the current study, carvacrol interacted with mTOR by binding free energy of -4.57 kcal/mol as represented in **Figure 3**. The ability of carvacrol to interact with mTOR could alleviate the aging features. The *in vivo* study of El-Far et al (2022) revealed that carvacrol attenuate brain D-galactose-induced aging-related oxidative alterations in rats.

Many such bioactive compounds, like as diosgenin, are found naturally in *Trigonella foenum graecum*, *Dioscorea alata*, *Smilax china*, and other plants (Arya et al. 2023). Diosgenin is a phytochemical that is used to cure fatal conditions such diabetes, cancer, arthritis, asthma, cardiovascular disease, and problems of the nervous system (Parama et al. 2020). Molecular docking revealed that diosgenin interacted with mTOR by binding free energy of -7.34 kcal/mol as represented in **Figure 4**. Previous studies have indicated that diosgenin is a bioactive chemical with a range of biological effects, including the amelioration of cognitive deficits related to aging (Chiu et al. 2011; El-Far et al. 2024).

The naturally occurring substance eugenol can be found in a variety of foods and plants, including peppers, *Zingiber officinale*, *tulsi* or holy basil leaves, *Eugenia caryophyllata*, bark from *Cinnamomum verum*, and *Curcuma longa* (Nisar et al. 2021). In the present study, eugenol interacted with mTOR by binding free energy of -7.34 kcal/mol as represented in **Figure 5**. Also, eugenol thought to be potential naturally occurring protective agents that could delay the aging process and maintain health (El-Far et al. 2022).

Rutin (3,3′,4′,5,7-pentahydroxyflavone-3-rhamnoglucoside) is a flavonol abundant in plants such as passionflower، buckwheat, tea, and apple (Ganeshpurkar and Saluja 2017). Data in **Figure 6** explored that rutin bound with ASN2147 (H-acceptor) and GLY2203 (pi-H) residues in mTOR by energy of -9.72 kcal/mol. Saafan et al (2023) stated that rutin attenuates the D-galactose-induced oxidative stress in rats' brain and liver.

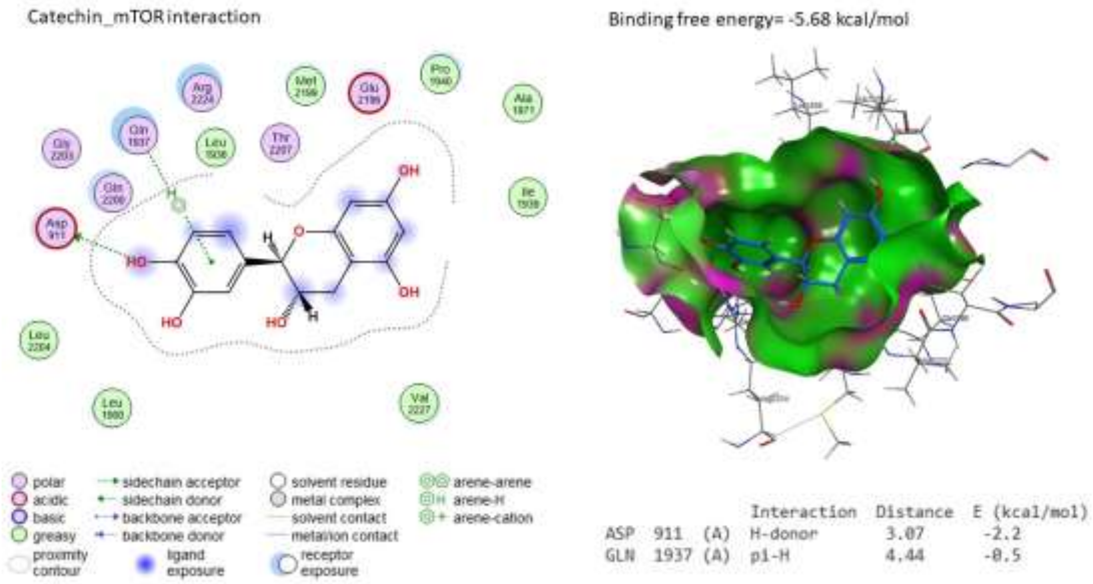

Figure 1. Molecular interaction of catechin against mammalian target of rapamycin (mTOR)

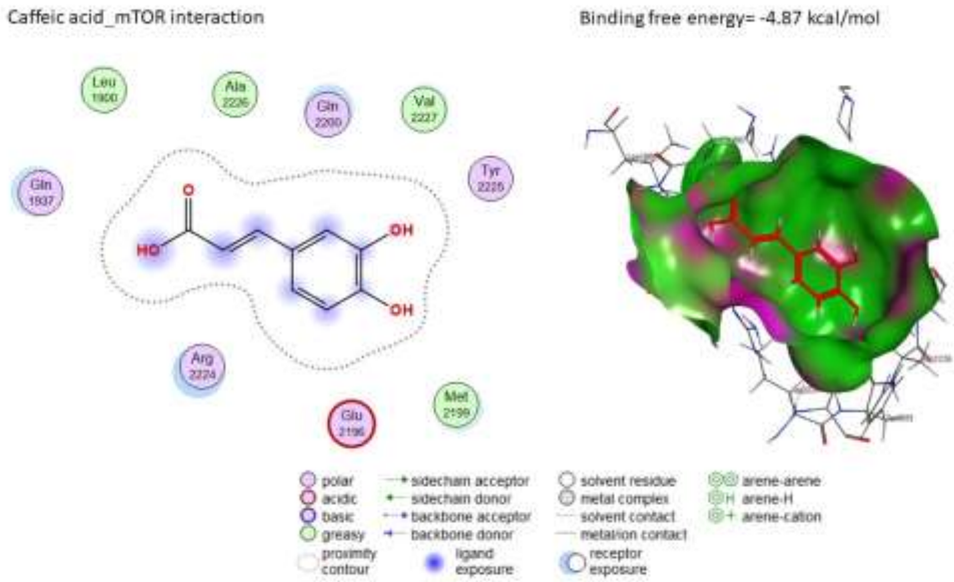

Figure 2. Molecular interaction of caffeic acid against mammalian target of rapamycin (mTOR)

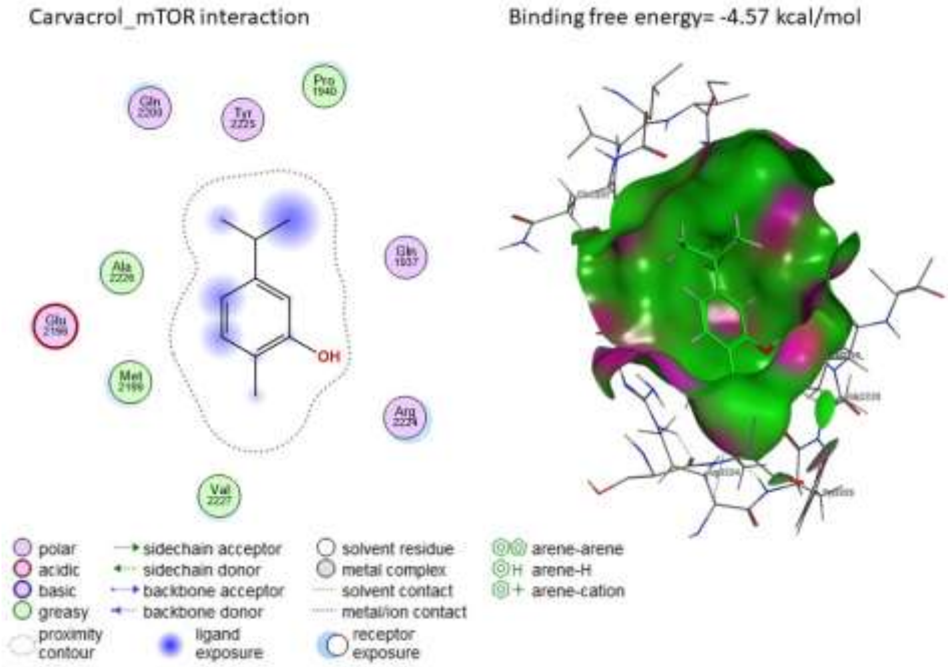

Figure 3. Molecular interaction of carvacrol against mammalian target of rapamycin (mTOR)

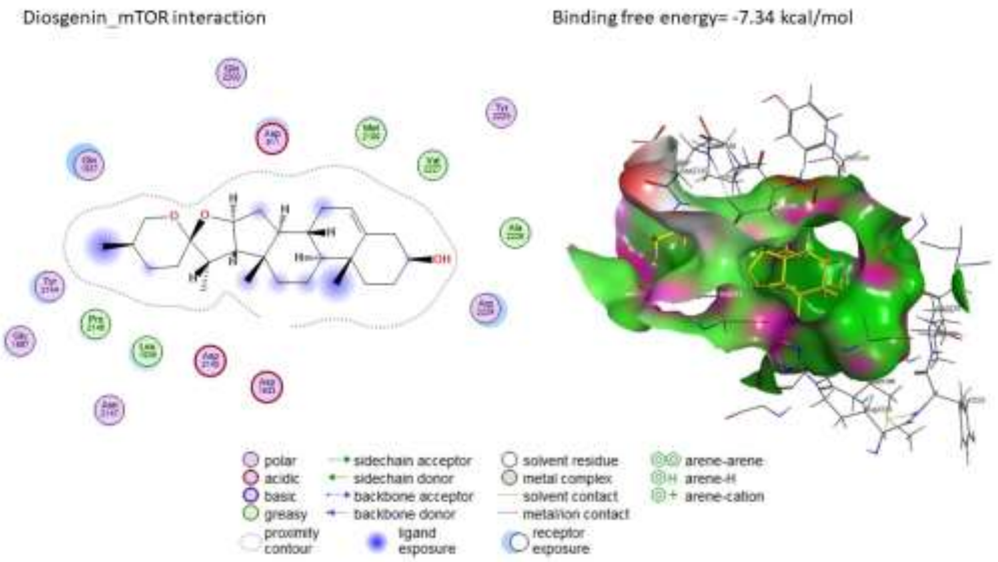

Figure 4. Molecular interaction of diosgenin against mammalian target of rapamycin (mTOR)

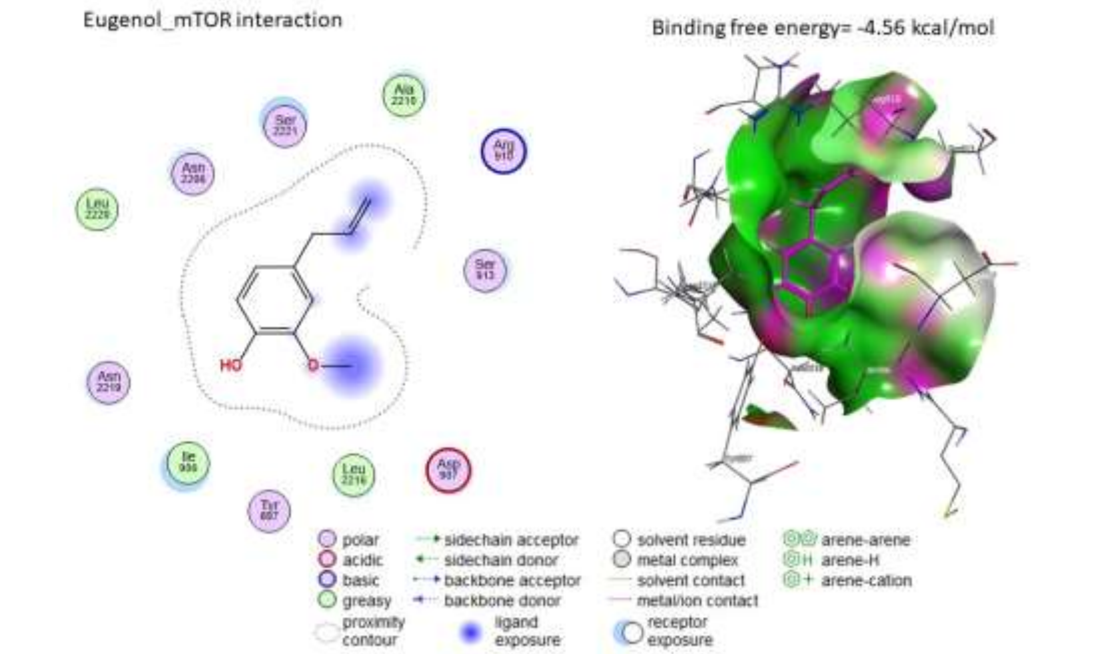

Figure 5. Molecular interaction of eugenol against mammalian target of rapamycin (mTOR)

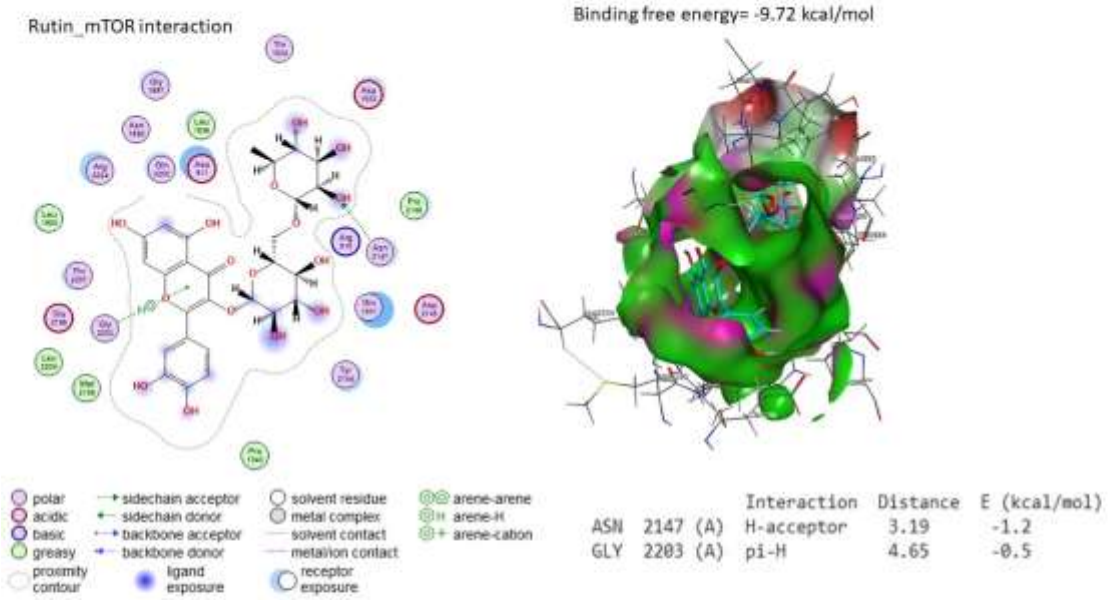

Figure 6. Molecular interaction of rutin against mammalian target of rapamycin (mTOR)

4. Conclusion

As natural anti-aging bioactive substances, catechin, caffeic acid, carvacrol, diosgenin, eugenol, and rutin have been shown to interact with mTOR by the use of the molecular docking technique in this work. Previous research has validated the in-silico model used in this work, but a deeper comprehension of the mechanisms behind their action is nevertheless required.

5. References

Arya P, Munshi M, Kumar P (2023) Diosgenin: Chemistry, extraction, quantification and health benefits. Food Chem Adv 2:100170. https://doi.org/10.1016/j.focha.2022.100170

Bernatoniene J, Kopustinskiene DM (2018) The Role of Catechins in Cellular Responses to Oxidative Stress. Molecules 23:965. https://doi.org/10.3390/molecules23040965

Braicu C, Ladomery MR, Chedea VS, et al (2013) The relationship between the structure and biological actions of green tea catechins. Food Chem 141:3282–9. https://doi.org/10.1016/j.foodchem.2013.05.122

Chiu C-SS, Chiu Y-JJ, Wu L-YY, et al (2011) Diosgenin ameliorates cognition deficit and attenuates oxidative damage in senescent mice induced by D-galactose. Am J Chin Med 39:551–63. https://doi.org/10.1142/S0192415X11009020

Chung J, Kuo CJ, Crabtree GR, Blenis J (1992) Rapamycin-FKBP specifically blocks growth-dependent activation of and signaling by the 70 kd S6 protein kinases. Cell 69:1227–36. https://doi.org/10.1016/0092-8674(92)90643-q

El-Far AH, Elewa YHA, Abdelfattah E-ZA, et al (2021) Thymoquinone and curcumin defeat aging-associated oxidative alterations induced by d-galactose in rats' brain and heart. Int J Mol Sci 22:6839. https://doi.org/10.3390/ijms22136839

El-Far AH, Elghaity MM, Mohamed SA, et al (2024) Diosgenin alleviates D-galactose-induced oxidative stress in rats' brain and liver targeting aging and apoptotic marker genes. Front Mol Biosci 11:1303379. https://doi.org/10.3389/fmolb.2024.1303379

El-Far AH, Lebda MA, Noreldin AE, et al (2020) Quercetin Attenuates Pancreatic and Renal D-Galactose-Induced Aging-Related Oxidative Alterations in Rats. Int J Mol Sci 21:4348. https://doi.org/10.3390/ijms21124348

El-Far AH, Mohamed HH, Elsabagh DA, et al (2022) Eugenol and carvacrol attenuate brain D-galactose-induced aging-related oxidative alterations in rats. Environ Sci Pollut Res Int 29:47436–47447. https://doi.org/10.1007/s11356-022-18984-8

Ganeshpurkar A, Saluja AK (2017) The Pharmacological Potential of Rutin. Saudi Pharm J SPJ Off Publ Saudi Pharm Soc 25:149–164. https://doi.org/10.1016/j.jsps.2016.04.025

Gingras AC, Raught B, Sonenberg N (2001) Regulation of translation initiation by FRAP/mTOR. Genes Dev 15:807–26. https://doi.org/10.1101/gad.887201

Gülçin I (2006) Antioxidant activity of caffeic acid (3,4-dihydroxycinnamic acid). Toxicology 217:213–20. https://doi.org/10.1016/j.tox.2005.09.011

Huang S, Houghton PJ (2003) Targeting mTOR signaling for cancer therapy. Curr Opin Pharmacol 3:371–7. https://doi.org/10.1016/s1471-4892(03)00071-7

Kennedy BK, Berger SL, Brunet A, et al (2014) Geroscience: linking aging to chronic disease. Cell 159:709–13. https://doi.org/10.1016/j.cell.2014.10.039

Khan FA, Maalik A, Murtaza G (2016) Inhibitory mechanism against oxidative stress of caffeic acid. J Food Drug Anal 24:695–702. https://doi.org/10.1016/j.jfda.2016.05.003

Khomsi M El, Imtara H, Kara M, et al (2022) Antimicrobial and Antioxidant Properties of Total Polyphenols of Anchusa italica Retz. Molecules 27:416. https://doi.org/10.3390/molecules27020416

Khoshdel F, Golmohammadi MG, Jannat Dost M, et al (2022) Impact of caffeic acid on the testicular damages in D-galactose-induced aging model in mice. Iran J Basic Med Sci 25:1190–1195. https://doi.org/10.22038/IJBMS.2022.63977.14092

Kim LC, Cook RS, Chen J (2017) mTORC1 and mTORC2 in cancer and the tumor microenvironment. Oncogene 36:2191–2201. https://doi.org/10.1038/onc.2016.363

Kuo CJ, Chung J, Fiorentino DF, et al (1992) Rapamycin selectively inhibits interleukin-2 activation of p70 S6 kinase. Nature 358:70–3. https://doi.org/10.1038/358070a0

Lee ES, Lee J-O, Lee SK, et al (2009) Caffeic acid phenethyl ester accumulates beta-catenin through GSK-3beta and participates in proliferation through mTOR in C2C12 cells. Life Sci 84:755–9. https://doi.org/10.1016/j.lfs.2009.03.004

López-Otín C, Blasco MA, Partridge L, et al (2013) The Hallmarks of Aging. Cell 153:1194–1217. https://doi.org/10.1016/j.cell.2013.05.039

Nisar MF, Khadim M, Rafiq M, et al (2021) Pharmacological Properties and Health Benefits of Eugenol: A Comprehensive Review. Oxid Med Cell Longev 2021:2497354. https://doi.org/10.1155/2021/2497354

Parama D, Boruah M, Yachna K, et al (2020) Diosgenin, a steroidal saponin, and its analogs: Effective therapies against different chronic diseases. Life Sci 260:118182. https://doi.org/10.1016/j.lfs.2020.118182

Saafan SM, Mohamed SA, Noreldin AE, et al (2023) Rutin attenuates D-galactose-induced oxidative stress in rats' brain and liver: molecular docking and experimental approaches. Food Funct 14:5728–5751. https://doi.org/10.1039/d2fo03301a

Saenno R, Dornlakorn O, Anosri T, et al (2022) Caffeic Acid Alleviates Memory and Hippocampal Neurogenesis Deficits in Aging Rats Induced by D-Galactose. Nutrients 14:2169. https://doi.org/10.3390/nu14102169

Salim B, Noureddine M (2020) Identification of Compounds from Nigella Sativa as New Potential Inhibitors of 2019 Novel Coronasvirus (Covid-19): Molecular Docking Study. chemRxiv 19:1–12. https://doi.org/10.26434/chemrxiv.12055716.v1

Sharifi-Rad M, Varoni EM, Iriti M, et al (2018) Carvacrol and human health: A comprehensive review. Phytother Res 32:1675–1687. https://doi.org/10.1002/ptr.6103

Stallone G, Infante B, Prisciandaro C, Grandaliano G (2019) mTOR and Aging: An Old Fashioned Dress. Int J Mol Sci 20:2774. https://doi.org/10.3390/ijms20112774

Tenchov R, Sasso JM, Wang X, Zhou QA (2024) Aging Hallmarks and Progression and Age-Related Diseases: A Landscape View of Research Advancement. ACS Chem Neurosci 15:1–30. https://doi.org/10.1021/acschemneuro.3c00531

Weichhart T (2018) mTOR as Regulator of Lifespan, Aging, and Cellular Senescence: A Mini-Review. Gerontology 64:127–134. https://doi.org/10.1159/000484629

Widowati W, Dani D, Vera V, Yuninda VA (2023) Antioxidant and Antiaging Potential of Salak Fruit Extract (Salacca zalacca (Gaert.)Voss). Maj Obat Tradis 28:230–236. https://doi.org/10.22146/mot.83995

Yang X, Yang C, Farberman A, et al (2008) The mammalian target of rapamycin-signaling pathway in regulating metabolism and growth. J Anim Sci 86:E36-50. https://doi.org/10.2527/jas.2007-0567