Tropical Journal of Drug Research June 2025; 2 (6): 180 - 185
ISSN: 2476-8065 (Electronic)
Available online at http://www.tjdr.org
https://doi.org/10.26538/tjdr/v2i6.1

Original Research Article

Effects of Quercetin on Brain Architecture in Lipopolysaccharide-Induced Neuroinflammation in Mice

Owemidu O Idowu[1], Ekpe A Christian[1], Atidoga Philip[1], Oladejo D Mariam[1], Saliu I Abdulrahman[1], Okewusi O Esther[1], Odufuwa F Eniola[1], Akhigbe F Omoighele[2]

[1] *Department of Physiology, Faculty of Basic Medical Sciences, Kogi State University, Anyigba, Nigeria.*
[2] *Department of Pharmacology, Faculty of Basic Medical Sciences, Kogi State University, Anyigba, Nigeria*.
*For correspondence: Email: owemidu.io@ksu.edu.ng; +234 7032603785

Sent for review: 02 June 2025 Revised accepted: 28 June 2025

Abstract

Purpose: Neuroinflammation is associated with oxidative stress, thereby leading to neuronal dysfunctions in the central nervous system. Quercetin is a well-known potent anti-oxidant capable of reducing the generation of free radicals. Preventing the generation of free radical may be a valuable therapeutic approach in the management of disorders such as Parkinson's disease, multiples sclerosis and Alzheimer's disease associated with neuroinflammation. This work was therefore designed to investigate the effect of quercetin on brain architecture in lipopolysaccharide (LPS) induced neuroinflammation in mice.
Method: Fifteen (15) animals (mice) were randomly divided into a group of three consisting of five animals in each group. Neuroinflammation was induced by a single injection of 2 mg/kg Escherichia *coli* bacterial lipopolysaccharide (serotype 055:B5, Sigma, St Louis, MO, USA) dissolved in 0.9% sterile saline intra-peritonially (IP).
Group A served as the control group and was administered distilled water for a period of three days while group B received only LPS meanwhile group C was induced and treated with quercetin (40 mg/kg p.o) for three days. After three days, all animals were sacrificed, and their brain tissues were carefully collected for analysis. Dendritic arborization and deoxyribonucleic acid (DNA) were quantified using silver stain and feulgen stain respectively
Results: Silver stain revealed that quercetin in group C prevented axonal loss as compared to untreated group B. Meanwhile in the feulgen stain, the DNA was preserved by quercetin as compared to untreated group B.
Conclusion: This study has clearly revealed that quercetin prevented neuronal dysfunction during neuroinflammation through mechanisms that may involve preventing axonal and DNA loss.

Keywords: Brain, Quercetin, Feulgen stain, Silver stain, neuroinflammation

This is an Open Access article that uses a funding model which does not charge readers or their institutions for access and distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0) and the Budapest Open Access Initiative (http://www.budapestopenaccessinitiative.org/read), which permit unrestricted use, distribution, and reproduction in any medium, provided the original work is properly credited.

Tropical Journal of Drug Research is indexed by Chemical Abstracts, Embase, Index Copernicus, EBSCO, African Index Medicus, JournalSeek, Directory of Open Access Journals
(DOAJ), African Journal Online, Bioline International, Open-J-Gate and Pharmacy Abstracts

© 2025 The authors. This work is licensed under the Creative Commons Attribution 4.0 International License

INTRODUCTION

Quercetin is one of the most widely distributed flavonoids characterized by a crystalline, yellow material which is completely insoluble in cold water, soluble in alcohol and lipids, and marginally soluble in hot water and is distinguished by its bitter taste. Structurally, it is an aglucone or aglycon that does not contain any carbohydrate molecules, and it gives a range of flowers great colors [1]. Quercetin is distributed in nature and commonly present in floral components, bark, stems, roots of various species. Additionally, it is found in numerous dietary sources such as wine, vegetables, tea, and fruits such as apples, capers, berries, cilantro, lovage, onions, and dill. Chemically, Quercetin is composed of benzene rings and hydroxyl groups. Quercetin has been extensively studied for its wide spectrum of pharmacological properties including its intriguing treatments for different health issues such as anticancer activity[2], antiallergic activity [3], anti-diabetes activity [4] anti-obesity activity[5], and its efficacy in managing hyperuricemia and gouty arthritis[6] etc. The most important impact of quercetin is its ability to inhibit the spread of certain cancers including those of the breast, cervix, lung, colon, prostate, and liver [7].

The therapeutic potential of quercetin is largely attributed to its antioxidant capacity. It directly scavenges free radicals, chelate metals ions perhaps inhibiting the formation of free radicals by the ions, and its ability to modulate signaling molecules [8] and also interacts with glutathione (GSH) [9]. It has been found that quercetin can modulate metallothionein which is an important metal detoxification protein [10]. The metabolites of quercetin has been shown to promote growth of beneficial gut microbiota, indicating its role in maintaining intestinal health[11].

Neuroinflammation refers to the inflammatory response within the brain or spinal cord. This process involves the release of signaling molecules like cytokines, chemokines and reactive oxygen species. These mediators are produced by resident CNS glia (microglia and astrocytes), endothelial cells, and peripherally derived immune cells. When tissue in the brain (or spinal cord) becomes inflamed, they can lead to inflammation in neural tissues, potentially contributing to various neurological conditions. Although inflammation on the skin is often visible and painful, inflammation in the brain cannot be seen or felt in the same way. However, during neuroinflammation, brain cells are directly affected, and individuals may experience temporary changes in mood, behavior, concentration, memory, fatigue and motivation. Neuroinflammatory responses are mediated by several key pro-inflammatory cytokines (IL-1β, IL-6, and TNFα), chemokines (CCL2, CCL5, CXCL1), secondary messengers (NO and prostaglandins) and reactive oxygen species (ROS). Many of these mediators are produced by activated resident CNS cells including microglia and astrocytes[12] Lipopolysaccharide (LPS), a toxic compound found in the outer membrane of Gram-negative bacteria, is commonly used to model neuroinflammation in animal studies because of its strong ability to activate microgalia and triggers the release of pro-inflammatory cytokines such as (IL-1β, IL-6, and TNFα)[13].

Quercetin has been widely studied for its antioxidant effects [14], as well as its roles in anti-aging[15], anti cancer[16], treatment in kidney and tumor tissues[17], it also helps reduce oxidative stress and blood pressure related markers in both urine and blood[18], Antidepressant activity[19]. Despite its potentials, research specifically investigating the effect of quercetin on neuronal architecture in the context of LPS induced neuroinflammation remains limited. This study aims to investigate the effect of quercetin on brain architecture in LPS induced neuroinflammation in mice

MATERIALS AND METHODS

Animals

Mice weighing between 20-30g were used for this study. They were acclimatized for 2 weeks at the Central Animal House, College of Health Sciences, Prince Abubakar Audu University, Anyigba. They were kept under standard laboratory conditions, and fed on rodent cubes. All experimental procedures on rodents were conducted in accordance with established protocols under the guidelines of the Principle of Laboratory Animal Care (National Institute of Health publication No. 85–23). Ethical approval number (CREC-CHS/PAAU/2025/0011) was obtained from the College of Health Sciences, Prince Abubakar Audu University ethical committee.

Induction of Neuroinflammation

Following the method of (Yun et al., 2019) the overnight fasted rats were injected intra-peritoneal (i.p.) with 2 mg/kg *Escherichia coli*

bacterial lipopolysaccharide (serotype 055:B5, Sigma, St Louis, MO, USA) dissolved in 0.9% sterile saline. Treatment was done on the day of induction and continuously on a daily basis up to the third day. Mice were sacrificed three days after Lipopolysaccharide injection. After sacrifice, brains were removed. Silver and Feulgen stain were used to quantify the dendritic arborization and DNA intensity respectively.

Animal grouping

Group A served as the control (not induced) received distilled water for three days while group B (induced) and received distilled water for three days after administration of LPS meanwhile group C was induced and treated with Quercetin (40 mg/kg p.o) for three days.

Statistical Analysis

Data were presented as mean ± Standard Error of Mean (SEM) using GraphPad Prism version 10. Comparisons between groups were made using the one-way analysis of variance (ANOVA) followed by Duncan post-hoc test. 95% confidence level, and at $p < 0.05$ was considered statistically significant.

RESULT AND DISCUSSION

This present study has investigated the role of quercetin in preserving the cytoarchitecture of the brain using the LPS induced model of neuroinflammation, meanwhile quantifying cytoarchitecture by silver stain and feulgen stain. Feulgen stain has earlier been used various purposes, including the precise estimation of chromatin condensation and cellular rearrangements of the cerebellar layers in X-ray exposed mice[20], and the quantitation of nuclear DNA content in lung carcincoma[21]. From the results as shown in Figure 1, Group A had intensity of +++ (indicating the measure of nuclear stain concentration) in the cell and a cell volume of ++ while Group B shows lesser intensity of stain (+), although the same volume of stain in group C, meanwhile Group A and C are comparable. This result indicates that quercetin given at 40 mg/kg preserved the brain cytoarhitecture specifically preserving the DNA in the brain cells as compared to untreated group B, our findings suggest that quercetin maybe preserving the cell via mechanism that are associated with preventing free radicals in the cell. Quercetin has earlier been reported for its anti-oxidant capacity[22]–its ability to prevent DNA loss may be attributed to this anti-oxidant properties.

Both animal and plant cell research has relied on the Feulgen reaction to perform specific DNA analysis. In 2020, the application of Feulgen staining was described for imaging of ovaries and developing embryo sacs in maize[23].

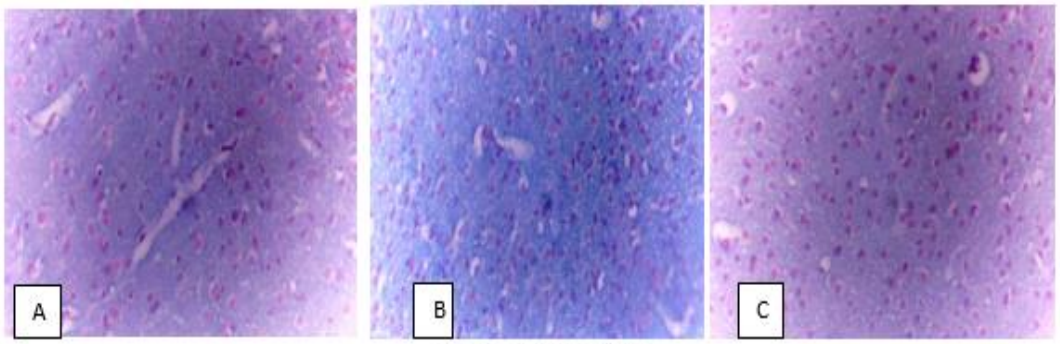

Figure 1: Effects of Quercetin on Feulgen stain

A- The photomicrograph above in feulgen stain show intensity(+++),vol(_++)
B- The photomicrograph above in feulgen stain shows intensity(+), volume(+++)
C- The photomicrograph above in feulgen stain show intensity(+++), vol(+++)

Silver stain is a highly sensitive colorimetric technique used to detect proteins and low molecular-weight nucleic acids in polyacrylamide gels, enabling visualization of even trace amount of the biomolecules. Silver staining techniques have been widely utilized to visualize diverse physical and biological

structures, for instance, Van Bregt et al[24] observed that silver staining detects progressive neuronal damage in the substantia nigra after moderate diffuse brain injury. Burk et al[25] also

demonstrated the use of silver staining in identifying axonal degeneration resulting from combined deficiencies in Vitamin E and Vitamin C in guinea pigs. Their findings revealed fragmented axons and degenerating fibers, particularly in the spinal cord and brainstem regions.

From the results as shown in Figure 2, Group A and B shows slight loss of axons while Group C shows high volume of normal axons. From these results, quercetin given at 40 mg/kg prevented axonal loss when compared with untreated group B. This observation can be attributed to the antioxidant activity of quercetin[14]. Since free radicals are generated during neuroinflammation. Mitigating this would be a valuable therapeutic approach in the management of neurodegenerative disorder associated with free radicals linked neuroinflammation.

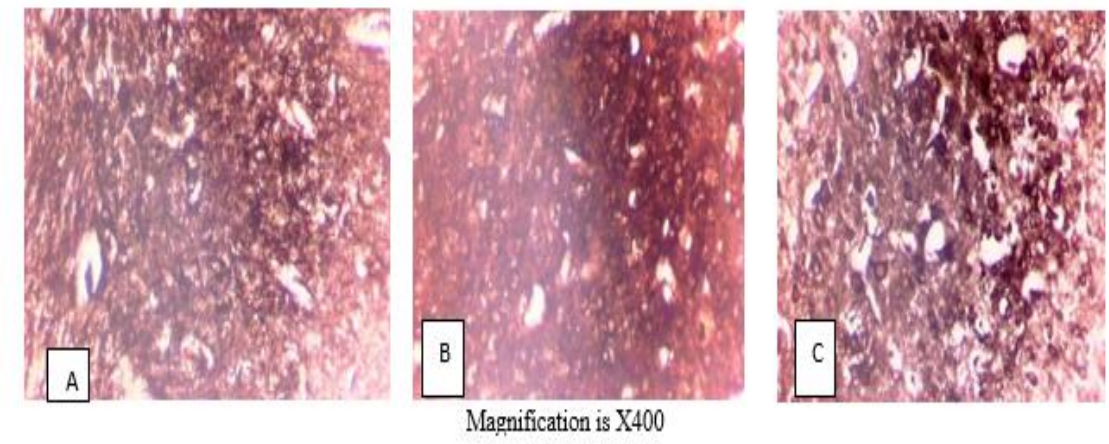

Figure 2: Effects of Quercetin on Silver stain
A- The photomicrograph above in silver stain shows slight loss of axons
B- The photomicrograph above in silver stain shows slight loss of axons
C- The photomicrograph above in silver stain shows high volume normal axons

CONCLUSION

This study has clearly indicated that quercetin protected the brain cytoarchitecture through mechanisms that are likely to be associated with preventing DNA and axonal loss.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHORS DECLARATION

The authors hereby declare that the works presented in this article are original and that any liability for claims relating to the content of this article will be borne by them.

Open Access

This is an Open Access article that uses a funding model which does not charge readers or their institutions for access and distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/ 4.0) and the Budapest Open Access Initiative (http://www.budapestopenaccessinitiative.org/read), which permit unrestricted use, distribution, and reproduction in any medium, provided the original work is properly credited

REFERENCES

1. Gaber EB, Amany MB, Muhammad I, Zohair SM, Mohamed EA, Ayman ET, Abdelazeem MA, Yaser HA. The Pharmacological Activity, Biochemical Properties, and Pharmacokinetics of the Major Natural Polyphenolic Flavonoid: Quercetin. Foods. 2020; 9: 374.
2. Rauf A, Imran M, Khan IA, Ur-Rehman M, Gilani SA, Mehmood Z, Mubarak MS. Anticancer Potential of Quercetin: A Comprehensive Review. Phytother Res. 2018; 32: 2109–2130.
3. Yuanyuan D, Chaomei L, Yongjing Z, Pengyu M, Tingting Z, Delu C, Jiao C, Jue W, Rui L, Tao Z, Langchong H. Quercetin as a Lyn Kinase Inhibitor Inhibits IgE-Mediated Allergic Conjunctivitis. Food Chem Toxicol. 2020; 135: 110924.

4. Zhanxin Y, Yeqing G, Qing Z, Li L, Ge M, Hongmei W, Yang X, Xue B, Hongbin S, Shaomei S, Xing W, Ming Z, Qiyu J, Yuntang W, Kun S, Weina G, Changjiang G, Kaijun N. Estimated Daily Quercetin Intake and Association with the Prevalence of Type 2 Diabetes Mellitus in Chinese Adults. Eur J Nutr. 2019; 58: 819–830.
5. Perdicaro DJ, Rodriguez Lanzi C, Gambarte Tudela J, Miatello RM, Oteiza PI, Vazquez Prieto MA. Quercetin Attenuates Adipose Hypertrophy, in Part through Activation of Adipogenesis in Rats Fed a High-Fat Diet. J Nutr Biochem. 2020; 79: 108352.
6. Nutmakul T. A Review on Benefits of Quercetin in Hyperuricemia and Gouty Arthritis. Saudi Pharm J. 2022; 30: 918–926.
7. Yue L, Zhen-Gang T, Yi L , Xin-Guo Q, Wei L, Guo-Bin W, Cai-Li L. Effects of Quercetin on Proliferation and Migration of Human Glioblastoma U251 Cells. Biomed Pharmacother. 2017; 92: 33–38.
8. Klara Z, Vedrana R , Josipa V, Patrick R, Nada O, Goran Š, Maja J. PI3K/Akt and ERK1/2 signaling are involved in quercetin-mediated neuroprotection against copper-induced injury Oxid Med Cell Longev. 2020; Article ID 9834742.
9. Xu. D, Hu. M.J, Wang. Y.Q, Cui. Y.L. Antioxidant activities of quercetin and its complexes for medicinal application Molecules. 2019; 24: 1123
10. Kuo SM, Huang CT, Blum P, Chang C. Quercetin cumulatively enhances copper induction of metallothionein in intestinal cells. Biol Trace Elem Res. 2001; 84: 1-10.
11. Hai Y, Zhang Y, Liang Y, Ma X, Qi X , Xiao J, Xue W, Luo Y, Yue T. Advance on the absorption, metabolism and efficacy exertion of quercetin and its important derivatives. Food Front. 2020; 1: 420-434.
12. Norden DM, Trojanowski PJ, Villanueva E, Navarro E, Godbout JP. Sequential activation of microglia and astrocyte cytokine expression precedes increased Iba-1 or GFAP immunoreactivity following systemic immune challenge. Glia. 2016; 64: 300–316.
13. Liya Q, Xuefei W, Michelle LB, Yuxin L, George RB, Jau-Shyong H, Darin JK, Fulton TC. Systemic LPS causes of chronic neuroinflammation and progressive neurodegeneration. Gila. 2007; 55(5): 453-462.
14. Boots AW, Haenen GR, Bast A. Health Effects of Quercetin: From Antioxidant to Nutraceutical. Eur J Pharmacol. 2008; 585: 325–337.
15. Cao Y, Zhao H, Wang Z, Zhang C, Bian Y, Liu X, Zhang C, Zhang X, Zhao Y. Quercetin Promotes in Vitro Maturation of Oocytes from Humans and Aged Mice. Cell Death Dis. 2020; 11: 965.
16. Ezzati M, Yousefi B, Velaei K, Safa A. A Review on Anti-Cancer Properties of Quercetin in Breast Cancer. Life Sci. 2020; 248: 117463.
17. Sánchez-González PD, López-Hernández FJ, Dueñas M, Prieto M, Sánchez-López E, Thomale J, Ruiz-Ortega M, López-Novoa JM, Morales AI. Differential Effect of Quercetin on Cisplatin-Induced Toxicity in Kidney and Tumor Tissues. Food Chem Toxicol. 2017; 107: 226–236.
18. Edwards RL, Lyon T, Litwin SE, Rabovsky A, Symons JD, Jalili T. Quercetin Reduces Blood Pressure in Hypertensive Subjects. J Nutr. 2007; 137: 2405–2411.
19. Singh V, Chauhan G, Shri R. Anti-Depressant like Effects of Quercetin 4'-O-Glucoside from Allium Cepa via Regulation of Brain Oxidative Stress and Monoamine Levels in Mice Subjected to Unpredictable Chronic Mild Stress. Nutr Neurosci. 2019; 24: 35–44.
20. Fidler Y, Gomes J. Effects of a single dose of X-ray irradiation on MMP-9 expression and morphology of the cerebellum cortex of adult rats. Cerebellum 2023; 22(2): 240–248.
21. Kobayashi S, Saio M, Fukuda T, Kimura K, Hirato J, Oyama T. Image analysis of the nuclear characteristics of emerin protein and the correlation with nuclear grooves and intranuclear cytoplasmic inclusions in lung adenocarcinoma. Oncol Rep. 2019; 41:133–142.
22. Fatemeh A, Milad H. Recent Advances in Potential Health Benefits of Quercetin. Pharmaceuticals. 2023; 16(7): 1020.
23. Kalinowska K, Chen J, Dresselhaus T. Imaging of embryo sac and early seed development in maize after Feulgen staining. Methods Mol Biol. 2020; 2122: 191–203.
24. Van Bregt DR, Thomas TC, Hinzman JM, Cao T, Liu M, Bing G, Gerhardt GA, Pauly JR, LIfshitz J. *Substantia nigra* vulnerability after a single moderate

diffuse brain injury in the rat. Exp Neurol. 2012; 233(1): 282-292

25. Burk RF, Hill KE, Motley AK, Winfrey VP, Kurokawa S, Mitchell SL, Zhang W, Torano JS, Birringer M, Saito Y, Takahashi K. Combined deficiency of vitamins E and C causes severe central nervous system damage in guinea pigs. J Nutr. 2006; 136(10): 2540-2545.