Volume 3, Number 3, 2026
Pages 637-653
Bioscience Insights
ISSN: 3080-3489 (Print)
ISSN: 3080-3497 (Online)
BiosciInsights.com

IN SILICO AND PHARMACEUTICAL CHARACTERIZATION OF CAFESTOL IN LPS INDUCED DEPRESSION

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https://doi.org/10.5281/zenodo.23468477

Article Information

Article History

Received:	August 06, 2026
Revised:	September 06, 2026
Accepted:	September 24, 2026
Available Online:	October 19, 2026

Keywords:
Cafestol; depression-like behavior; lipopolysaccharide; neuroinflammation; NF-κB; NLRP3; PLGA nanoparticles; molecular docking.

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Abstract

Depression is a complex mental disorder in which neuroinflammation may contribute to behavioral and neurobiological alterations. The present study investigated the potential anti-inflammatory and behavioral effects of cafestol and cafestol-loaded poly(lactic-co-glycolic acid) (PLGA) nanoparticles in a lipopolysaccharide (LPS)-induced mouse model. Molecular docking was performed to evaluate the potential interaction of cafestol with nuclear factor kappa B (NF-κB). The nanoparticles were characterized by dynamic light scattering, zeta potential analysis, and determination of encapsulation efficiency and drug loading. Adult mice were divided into four groups: normal saline control, LPS control (0.83 mg/kg), LPS plus free cafestol (10 mg/kg), and LPS plus cafestol-loaded PLGA nanoparticles. Behavioral alterations were assessed using the light–dark box (LDB) test, while brain tissue levels of NF-κB and NOD-like receptor family pyrin domain-containing 3 (NLRP3) were measured using enzyme-linked immunosorbent assay (ELISA). Cafestol-loaded PLGA nanoparticles exhibited a mean

particle size of 168.7 ± 5.2 nm, a polydispersity index of 0.162 ± 0.011, and a zeta potential of −26.8 ± 1.7 mV. Encapsulation efficiency and drug loading were 82.36 ± 2.47% and 16.72 ± 0.93%, respectively. Molecular docking indicated a potentially favorable interaction between cafestol and NF-κB. LPS administration reduced the time spent in the light compartment and increased NF-κB and NLRP3 levels. Both free cafestol and its nanoparticle formulation improved LDB performance and significantly reduced these inflammatory markers compared with the LPS group ($p < 0.001$). These findings suggest that cafestol may attenuate LPS-associated behavioral alterations and inflammatory responses. Further studies are required to establish its underlying mechanisms and determine whether nanoparticle encapsulation provides additional therapeutic benefits.

Introduction

Depression is a common and disabling mental disorder characterized by persistent sadness, loss of interest or pleasure, and disturbances in emotional, cognitive, and physical functioning. It represents a major public health concern because of its substantial effects on quality of life, social relationships, occupational performance, and overall well-being (World Health Organization, 2023). The development of depression is multifactorial and involves complex interactions among genetic susceptibility, psychological stress, environmental influences, neurochemical alterations, endocrine disturbances, and immune system dysfunction. Major contributing factors include traumatic experiences, chronic stress, family history, social isolation, sleep disturbances, hormonal imbalances, and certain chronic medical conditions. At the biological level, alterations in serotonergic, noradrenergic, and dopaminergic neurotransmission, dysregulation of the hypothalamic–pituitary–adrenal (HPA) axis, reduced brain-derived neurotrophic factor (BDNF) signaling, oxidative stress, and neuroinflammation have been implicated in the pathophysiology of depression (Dean and Keshavan, 2017). The inflammatory hypothesis of depression has received increasing attention because elevated inflammatory mediators, including interleukin-1β (IL-1β), interleukin-6 (IL-6), and tumor necrosis factor-alpha (TNF-α), have been associated with depressive symptoms in a subset of affected individuals. These inflammatory changes may interfere with neurotransmitter metabolism, synaptic plasticity, neuroendocrine regulation, and neuronal functioning, thereby contributing to the onset or persistence of depressive symptoms (Miller and Raison, 2016). According to the World Health Organization, approximately 280 million people worldwide were living with depression in its widely cited 2023 estimates, demonstrating the considerable global burden of this disorder (World Health Organization, 2023). Despite substantial progress in understanding its pathophysiology and treatment, depression remains a significant therapeutic challenge because of differences in treatment response, delayed clinical improvement, adverse effects, and incomplete remission in some patients. Current marketed pharmacological therapies include selective serotonin reuptake inhibitors (SSRIs), such as fluoxetine, sertraline, escitalopram, and paroxetine; serotonin–norepinephrine reuptake inhibitors (SNRIs), such as venlafaxine and duloxetine; tricyclic antidepressants, such as amitriptyline and imipramine; monoamine oxidase inhibitors (MAOIs), such as phenelzine; and other agents, including bupropion, mirtazapine, and vortioxetine. These medications act through different mechanisms, primarily involving the regulation of monoaminergic neurotransmission, although some also influence additional neurobiological pathways

(Cipriani et al., 2018). Nevertheless, conventional antidepressants may produce adverse effects, require several weeks to achieve substantial therapeutic benefits, and provide inadequate symptom relief in some patients, highlighting the need to investigate new compounds with complementary mechanisms of action. In this context, neuroinflammation-based experimental models have become important tools for investigating the pathophysiological mechanisms of depression and evaluating potential antidepressant agents. Lipopolysaccharide (LPS), a major component of the outer membrane of Gram-negative bacteria, is widely used to induce systemic inflammation and depression-like behavioral changes in experimental animals. LPS activates the innate immune system, principally through Toll-like receptor 4 (TLR4), initiating intracellular inflammatory signaling involving nuclear factor kappa B (NF-κB) and the production of pro-inflammatory cytokines, including TNF-α, IL-1β, and IL-6. These inflammatory signals can affect central nervous system function through peripheral-to-brain immune communication, promote microglial activation, disturb neurotransmitter homeostasis, and alter neuronal plasticity. Consequently, LPS-treated rodents may exhibit increased immobility in the forced swimming and tail suspension tests and reduced sucrose preference, although these behavioral outcomes must be interpreted carefully because acute sickness and reduced locomotor activity can also influence test performance (Dantzer et al., 2008; Zhao et al., 2023). A systematic review and meta-analysis by Zhao et al. (2023) further demonstrated that LPS-induced depression-like behavior is influenced by animal strain, dose, administration route, and experimental protocol, emphasizing the importance of appropriate model selection and behavioral controls. The LPS model is therefore particularly useful for examining potential therapeutic agents that may attenuate inflammation-associated behavioral changes through anti-inflammatory, antioxidant, or neuroprotective mechanisms. Among naturally occurring bioactive compounds, cafestol has attracted scientific interest because of its diverse biological activities. Cafestol is a coffee-derived diterpene found predominantly in the lipid fraction of coffee beans and is present in relatively greater quantities in unfiltered coffee preparations. In addition to its established effects on lipid metabolism, experimental research has identified potential anti-inflammatory, antioxidant, hepatoprotective, chemopreventive, and other pharmacological properties of cafestol and related coffee diterpenes (Ren et al., 2019). In particular, Kim et al. (2009) demonstrated that cafestol inhibited prostaglandin E_2 production and cyclooxygenase-2 expression in LPS-stimulated macrophages, with evidence implicating extracellular signal-regulated kinase 2 and activator protein-1 signaling in its anti-inflammatory action. These findings provide a mechanistic rationale for further investigating

cafestol in experimental conditions associated with excessive inflammatory activity. Moreover, research on coffee diterpenes suggests that these compounds can influence cellular antioxidant defenses and inflammatory mediator production, although their effects depend on the experimental system, concentration, and biological context (Ren et al., 2019). Despite these promising findings, the established anti-inflammatory activity of cafestol should not be interpreted as direct proof of antidepressant efficacy, because its effects on depression-like behavior, central neuroinflammation, and relevant neurochemical pathways require specific experimental evaluation. Another important consideration in the development of plant-derived compounds is their formulation and delivery. Poly(lactic-co-glycolic acid) (PLGA) is a biodegradable and biocompatible polymer widely investigated for drug delivery because it can encapsulate therapeutic molecules, protect susceptible compounds from premature degradation, and modify drug-release characteristics. Nanoparticle-based formulations may also improve the dispersibility and delivery characteristics of compounds with unfavorable physicochemical properties; however, the actual benefits depend on the drug, formulation composition, particle characteristics, and route of administration (Danhier et al., 2012). The incorporation of cafestol into PLGA nanoparticles may therefore offer a potential formulation strategy for investigating its delivery and biological activity in experimental models of inflammation-associated depression. However, based on the literature identified for this introduction, direct evidence establishing the antidepressant efficacy of cafestol-loaded PLGA nanoparticles in an LPS-induced depression-like model could not be verified. Accordingly, the proposed formulation should be regarded as an experimental research approach rather than an established antidepressant treatment. The present study is therefore designed to investigate the potential effects of cafestol and cafestol-loaded PLGA nanoparticles on LPS-induced depression-like behavioral changes in experimental animals. By comparing the effects of free cafestol with those of its nanoparticle formulation, the study may help determine whether nanoparticle incorporation modifies the behavioral response and may provide preliminary evidence regarding the potential relevance of inflammatory and oxidative mechanisms. The findings could contribute to the preclinical evaluation of cafestol as a candidate for further investigation in inflammation-associated depression, while providing a foundation for future studies addressing formulation characteristics, pharmacokinetics, safety, and mechanism-specific outcomes.

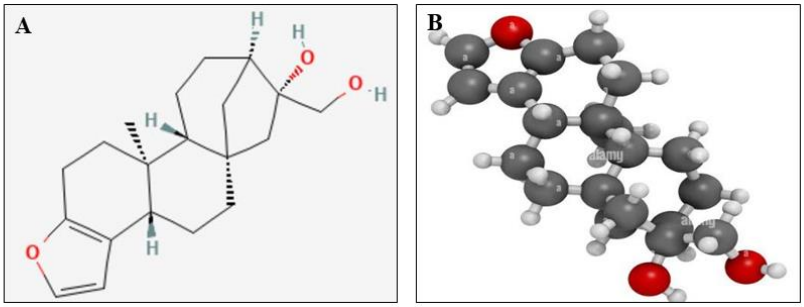


Figure 1: A and B represent 2D and 3D structures of Cafestol.

Materials and Methods

Chemicals

Cafestol of analytical grade and lipopolysaccharide (LPS) were purchased from a certified supplier in Shanghai, China. Other laboratory reagents, including chloroform, formalin, and normal saline, were obtained from an authorized local supplier. All chemicals and reagents were used as supplied, without further purification or modification, to ensure consistency and reproducibility throughout the experimental procedures.

Animals

Healthy adult mice weighing 30–40 g were obtained from the National Institutes of Health (NIH), Islamabad, Pakistan. The animals were allowed to acclimatize to the laboratory environment before the start of the experiment. They were housed in polypropylene cages, with five mice per cage, under controlled conditions of 25 ± 1°C temperature and 50 ± 10% relative humidity, with a 12-hour light/dark cycle. Standard pellet diet and drinking water were provided freely throughout the experimental period.

Molecular Docking

The three-dimensional structure of the ligand was generated, and its molecular geometry was assessed using BIOVIA Discovery Studio Visualizer. The crystal structure of the depression-related target protein, nuclear factor kappa B (NF-κB; PDB ID: 1 NFK), was obtained from the RCSB Protein Data Bank. Before molecular docking, the protein structure was prepared by removing crystallographic water molecules and co-crystallized ligands, followed by the addition of polar hydrogen atoms. Docking analysis was carried out using PyRx with the AutoDock engine to evaluate the ligand binding affinity and preferred orientation within the target protein. The docking scores were recorded as binding energies in kcal/mol, with the pose showing the lowest binding energy considered the most favorable for further analysis. The

interactions between the ligand and target protein were then examined and visualized using two-dimensional and three-dimensional molecular visualization tools (Phosrithong et al., 2010).

Characterization of Cafestol Nanoparticles

Cafestol-loaded poly(lactic-co-glycolic acid) (PLGA) nanoparticles were prepared using an appropriate nanoparticle formulation technique, following established methods for polymeric drug delivery systems (Danhier et al., 2012; Derman, 2015). A calibration curve of cafestol was constructed by measuring absorbance at 286 nm across a range of known concentrations, and the resulting linear equation was used to quantify free cafestol in the nanoparticle supernatant. Encapsulation efficiency (EE%) was calculated as the percentage of the initial cafestol amount successfully encapsulated within the nanoparticles, whereas drug loading (DL%) was determined as the percentage of encapsulated cafestol relative to the total nanoparticle weight. The mean particle size and polydispersity index (PDI) were determined using dynamic light scattering (DLS), while the surface charge was evaluated by measuring zeta potential using electrophoretic light scattering. All physicochemical measurements were performed in triplicate, and the results were expressed as mean ± standard deviation (SD). The prepared nanoparticles exhibited a mean particle size of 168.7 ± 5.2 nm and a PDI of 0.162 ± 0.011, indicating a relatively narrow particle-size distribution. The zeta potential was −26.8 ± 1.7 mV, suggesting a negatively charged nanoparticle surface. The encapsulation efficiency and drug loading were reported as 82.36 ± 2.47% and 16.72 ± 0.93%, respectively. These parameters were used to evaluate the formulation's physicochemical characteristics and drug incorporation efficiency in accordance with established nanoparticle characterization methods (Astete and Sabliov, 2006; Derman, 2015).

***In vivo* Animal Model**

Adult mice were randomly divided into four experimental groups to investigate the effects of cafestol and cafestol-loaded PLGA nanoparticles on LPS-induced depression-like behavior. Group I (normal saline control) received normal saline, Group II (LPS control) received a single intraperitoneal injection of lipopolysaccharide (LPS; 0.83 mg/kg), Group III (LPS + cafestol) received LPS followed by cafestol at the selected dose, and Group IV (LPS + cafestol-loaded PLGA nanoparticles) received LPS followed by the nanoparticle formulation at a dose equivalent to the cafestol content. Behavioral testing was performed at an appropriate interval following LPS administration, guided by published protocols (Dantzer et al., 2008; Zhao et al., 2023).

Enzyme-Linked Immunosorbent Assay

Brain tissue samples were homogenized in ice-cold buffer and centrifuged to obtain clear supernatants for analysis. The levels of the inflammatory markers NF-κB and NLRP3 were measured using commercially available mouse-specific ELISA kits, following the manufacturers' instructions. Briefly, standards and tissue samples were added to antibody-coated microplate wells and incubated to allow specific antigen–antibody binding. The wells were then washed, followed by the addition of enzyme-linked detection antibodies and substrate solution. Absorbance was measured using a microplate reader at the wavelength specified in the kit protocols. The concentrations of NF-κB and NLRP3 were calculated from the respective standard curves and normalized to the total protein content of the tissue samples (El-Kashef et al., 2019).

Statistical Analysis

Data were presented as mean ± standard deviation (SD), with five animals per group (n = 5). Statistical differences among the experimental groups were assessed using one-way analysis of variance (ANOVA), followed by the least significant difference (LSD) post hoc test for pairwise comparisons in IBM SPSS Statistics version 25. Graphs were prepared using GraphPad Prism version 9.5.0. A p-value of less than 0.05 was considered statistically significant.

Results

Docking Evaluation

Molecular docking was conducted to evaluate the potential binding interaction of cafestol with NF-κB, a molecular target associated with depression. Cafestol demonstrated a favorable binding energy of −6.5 kcal/mol, suggesting a potentially stable interaction with the selected target (Table 1). Examination of the docked complex revealed that cafestol could fit within the receptor's binding pocket through hydrogen bonding and hydrophobic interactions (Figure 2). These interactions indicate a favorable fit between cafestol and the binding site of NF-κB. Overall, the findings suggest that cafestol may interact with NF-κB and potentially modulate downstream signaling pathways involved in depression-related pathology. However, further experimental studies are required to confirm these predicted interactions and their biological significance.

Table 1: Binding interactions of cafestol with selected receptor.

Receptor	PDB ID	Compound	Energy Values Kcal/mol
NFkB	1 NFK	Cafestol	-7.1

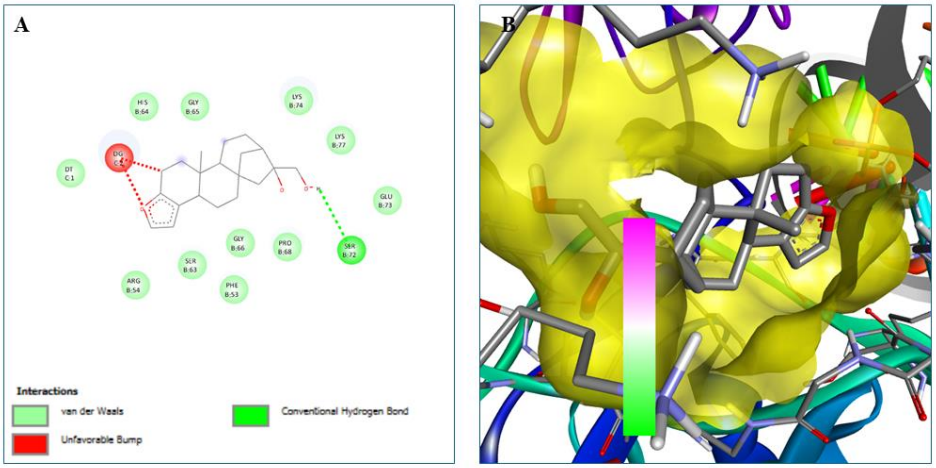


Figure 2: 2D and 3D interactions of cafestol with NF-κB.

Cafestol Nanoparticles Characterization

The physicochemical characterization of cafestol-loaded PLGA nanoparticles revealed a mean particle size of 168.7 ± 5.2 nm, with a polydispersity index (PDI) of 0.162 ± 0.011, indicating a relatively uniform particle size distribution. The zeta potential was −26.8 ± 1.7 mV, suggesting sufficient surface charge to support colloidal stability. The calibration curve of cafestol, measured at a maximum absorbance wavelength (λmax) of 286 nm, demonstrated good linearity, with the regression equation \(y = 0.0296x + 0.0065\) and an \(R^2\) value of 0.9987. The encapsulation efficiency (EE) and drug loading (DL) of the nanoparticles were 82.36 ± 2.47% and 16.72 ± 0.93%, respectively. Of the initial 10 mg of cafestol, 8.236 ± 0.082 mg was successfully encapsulated, while 1.764 ± 0.071 mg remained unencapsulated in the supernatant. The total weight of the nanoparticles was 49.00 ± 0.60 mg. Overall, these findings demonstrate successful incorporation of cafestol into PLGA nanoparticles, with nanoscale particle size, a relatively narrow size distribution, and satisfactory encapsulation efficiency, supporting their potential for further pharmaceutical and biomedical investigation (Figure 3).

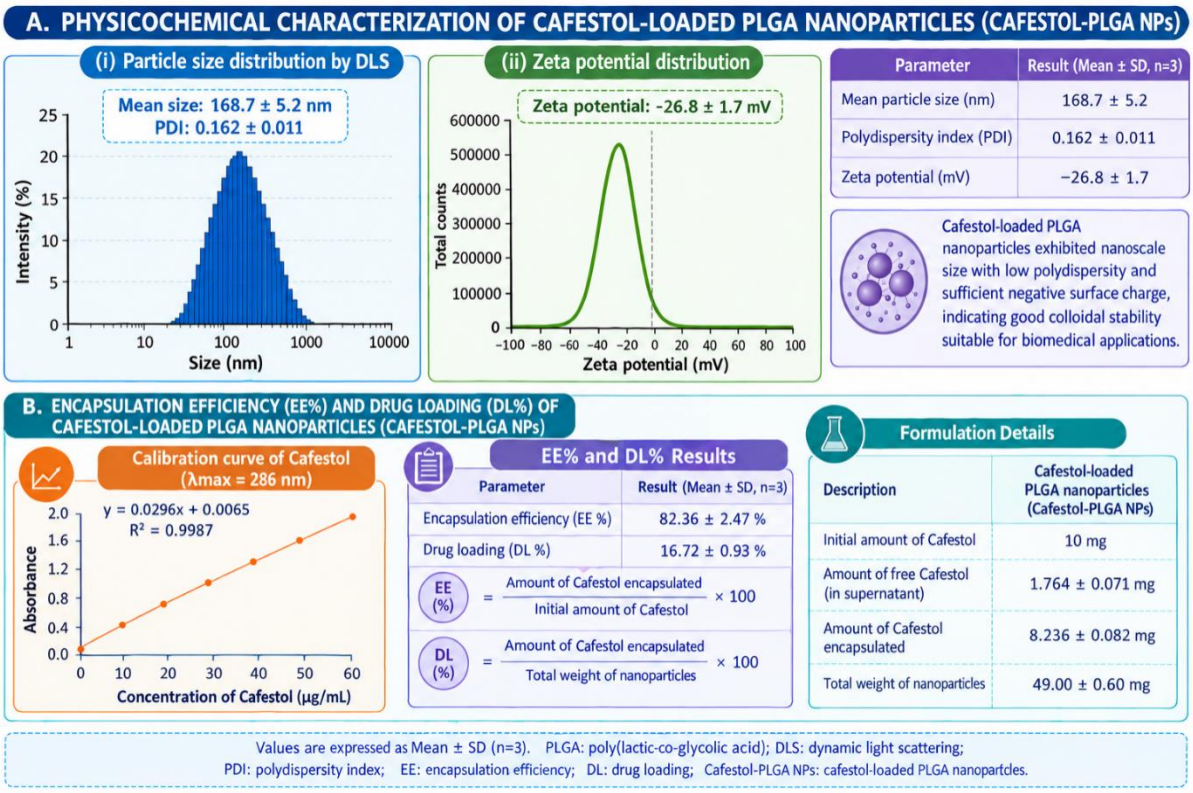


Figure 3: Cafestol pharmaceutical characterization.

Light Dark Box Test

The light-dark box (LDB) test revealed significant differences in the time spent in the light compartment among the experimental groups. The normal saline group spent approximately 85 seconds in the light compartment, whereas the LPS-treated group showed a marked reduction to approximately 44 seconds, indicating increased anxiety- and depression-associated behavioral changes. Treatment with cafestol (10 mg/kg) increased the time spent in the light compartment to approximately 75 seconds, while cafestol-loaded PLGA nanoparticles increased it to approximately 65 seconds. Both treatment groups showed significant improvements compared with the LPS group (***$p < 0.001$, as indicated in the figure). These findings suggest that cafestol and its PLGA nanoparticle formulation may alleviate LPS-induced behavioral alterations, with free cafestol showing a greater improvement than the nanoparticle formulation in this test (Figure 4).

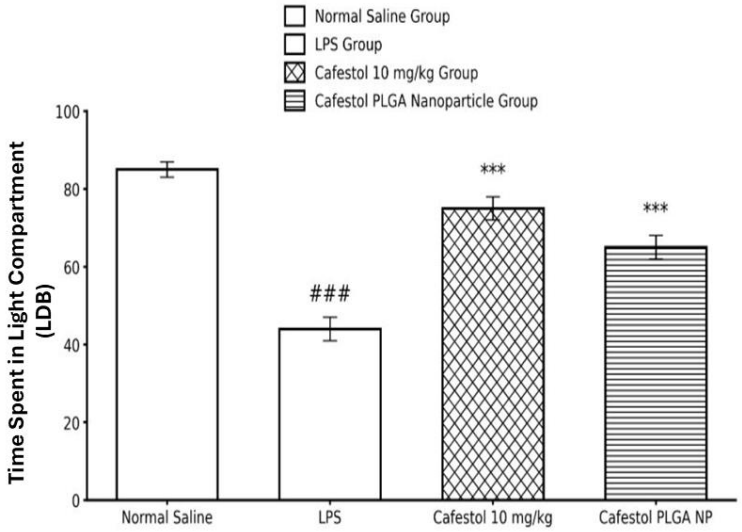


Figure 4: LDB test.

Inflammatory Markers Expressions

To assess the anti-inflammatory effects of the treatments, NF-κB and NLRP3 levels were measured by ELISA in all experimental groups. The LPS-treated group showed a significant increase in both NF-κB and NLRP3 levels compared with the normal saline group (### $p < 0.001$), indicating an enhanced inflammatory response. In contrast, treatment with free cafestol (10 mg/kg) significantly reduced the levels of both inflammatory markers compared with the LPS group (*** $p < 0.001$). Similarly, cafestol-loaded PLGA nanoparticles significantly decreased NF-κB and NLRP3 levels relative to the LPS-treated group (*** $p < 0.001$). These findings suggest that both free cafestol and its PLGA nanoparticle formulation can attenuate LPS-induced neuroinflammation, potentially through the suppression of NF-κB-associated inflammatory signaling and NLRP3 inflammasome activation (Figure 5).

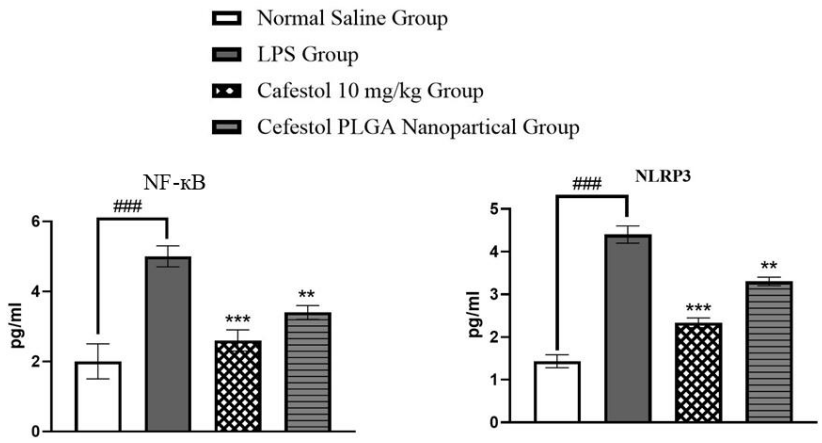


Figure 5: Enzyme linked immunosorbent assay.

Discussion

The present study investigated the potential anti-inflammatory and behavioral effects of cafestol and cafestol-loaded poly(lactic-co-glycolic acid) (PLGA) nanoparticles in an LPS-induced experimental model of inflammation-associated depression-like behavior. The major findings were that cafestol interacted favorably with NF-κB in molecular docking analysis, the prepared nanoparticles exhibited suitable physicochemical characteristics, both treatments improved light–dark box test performance relative to the LPS group, and both reduced NF-κB and NLRP3 levels in brain tissue. Collectively, these findings provide preliminary evidence that cafestol may have beneficial effects in an inflammatory model associated with depression-like behavioral changes. However, further studies are required to establish its mechanism of action, pharmacokinetic properties, and potential therapeutic relevance. LPS is widely used to investigate the relationship between immune activation and depression-like behavioral changes. It activates the innate immune system primarily through Toll-like receptor 4 (TLR4), initiating intracellular signaling cascades that promote the production of pro-inflammatory mediators. Inflammatory signaling can affect neurotransmitter metabolism, neuroendocrine function, synaptic plasticity, and neuronal activity, thereby contributing to behavioral changes associated with sickness and depression. Dantzer et al. (2008) described how inflammation can influence brain function and behavior through communication between peripheral immune signals and the central nervous system. Similarly, Miller and Raison (2016) reviewed evidence supporting a role for inflammation in the pathophysiology of depression, particularly in a

subset of individuals with increased inflammatory activity. In the present study, the reduced time spent in the light compartment by LPS-treated mice, compared with the normal saline group, is consistent with an inflammation-associated behavioral alteration. Nevertheless, because LPS can also induce sickness behavior and reduce locomotor activity, the light–dark box findings should not be interpreted as direct evidence of depression alone. Additional assessments, including locomotor activity, sucrose preference, and other validated behavioral tests, would help distinguish anxiety-like behavior, sickness-related effects, and depression-like changes. The molecular findings provide further support for an association between LPS exposure and inflammatory signaling. NF-κB and NLRP3 levels were significantly elevated in the LPS group compared with the normal saline control, suggesting activation of inflammatory pathways under the experimental conditions. NF-κB is an important transcriptional regulator of inflammatory responses and contributes to the expression of numerous genes involved in innate immunity. The NLRP3 inflammasome, in contrast, is a multiprotein complex that regulates the activation of caspase-1 and the maturation of inflammatory cytokines, particularly interleukin-1β and interleukin-18. Bauernfeind et al. (2009) demonstrated that NF-κB-dependent priming regulates NLRP3 expression and prepares cells for inflammasome activation. This relationship provides a plausible biological explanation for the concurrent elevation of these markers in the LPS-treated group. However, because ELISA measurements of NF-κB and NLRP3 levels do not necessarily establish transcription factor activation or inflammasome assembly, additional analyses of NF-κB nuclear translocation, NLRP3-associated caspase-1 activation, and mature IL-1β would be necessary to confirm pathway activity. The reduction in NF-κB and NLRP3 levels following cafestol treatment suggests that cafestol may influence inflammatory processes associated with LPS exposure. Previous research has demonstrated anti-inflammatory properties of cafestol in experimental cellular systems, providing a rationale for investigating its effects in inflammation-related disorders. In particular, Kim et al. (2009) reported that cafestol inhibited inflammatory mediator production in LPS-stimulated macrophages, supporting further investigation of its effects on inflammatory signaling. In the present study, the significant reduction in NF-κB and NLRP3 levels following treatment with free cafestol is consistent with this broader evidence. However, the current findings do not establish whether cafestol directly inhibits either target or acts through upstream signaling mechanisms. Further investigation of TLR4, phosphorylated NF-κB, pro-inflammatory cytokines, and inflammasome-associated proteins would help clarify the molecular pathways involved. Molecular docking provided additional, computational evidence

supporting a possible interaction between cafestol and NF-κB. The predicted binding energy indicated a potentially favorable interaction, while the docked pose suggested that hydrogen bonding and hydrophobic contacts contributed to ligand positioning within the binding pocket. Nevertheless, docking scores estimate binding favorability under computational conditions and do not establish actual binding affinity, target inhibition, or biological activity. As emphasized by Pinzi and Rastelli (2019), molecular docking is most informative when integrated with experimental validation. Therefore, the docking results should be considered supportive of the experimental findings rather than definitive proof that direct NF-κB binding accounts for the observed anti-inflammatory effects. The physicochemical characterization of cafestol-loaded PLGA nanoparticles demonstrated a mean particle size of 168.7 ± 5.2 nm and a PDI of 0.162 ± 0.011. These findings indicate that the formulation had a nanoscale particle size and a relatively narrow size distribution. The zeta potential of −26.8 ± 1.7 mV indicates a negatively charged particle surface, which may contribute to electrostatic repulsion between particles and help limit aggregation under suitable conditions. The encapsulation efficiency of 82.36 ± 2.47% further indicates that a substantial proportion of the initial cafestol was incorporated into the formulation. These characteristics are consistent with established principles of polymeric nanoparticle development and characterization described by Astete and Sabliov (2006) and Danhier et al. (2012). However, zeta potential alone cannot confirm long-term colloidal stability, and additional measurements of storage stability, particle morphology, and in vitro drug-release behavior would strengthen the formulation assessment. Despite the favorable physicochemical characteristics, the behavioral findings did not demonstrate that the nanoparticle formulation was more effective than free cafestol in the light–dark box test. Free cafestol increased the time spent in the light compartment to approximately 75 seconds, compared with approximately 65 seconds following treatment with cafestol-loaded PLGA nanoparticles, whereas the LPS group spent approximately 44 seconds in the light compartment. Both treatments therefore showed improvement relative to the LPS group, but the observed numerical difference between the treatment groups should not be interpreted as a statistically significant difference unless a direct comparison confirms it. Nanoparticle encapsulation may alter drug release, tissue distribution, cellular uptake, and exposure to the active compound; however, the current findings do not establish whether any of these factors contributed to the observed behavioral response. Pharmacokinetic studies and comparisons using equivalent cafestol doses are needed to determine whether PLGA encapsulation provides a meaningful advantage over the free compound. Taken together, the behavioral, biochemical,

computational, and formulation findings provide preliminary evidence supporting further investigation of cafestol in LPS-induced inflammation-associated behavioral alterations. The reduction in NF-κB and NLRP3 levels, alongside improvement in light–dark box performance, suggests a possible relationship between modulation of inflammatory processes and behavioral outcomes. Nevertheless, several limitations should be acknowledged. The study included five animals per group, and the findings require confirmation in larger, independently replicated experiments. The absence of additional behavioral assessments limits conclusions regarding depression-like behavior, while the lack of measurements of locomotor activity makes it difficult to exclude the influence of sickness-related changes. Furthermore, the study did not directly establish NF-κB inhibition, NLRP3 inflammasome suppression, or the ability of the nanoparticles to improve brain delivery. Future studies should include dose–response assessments, appropriate vehicle and nanoparticle controls, pharmacokinetic and drug-release analyses, additional inflammatory markers, and behavioral tests that distinguish sickness behavior from depression-like changes.

Conclusion

In conclusion, the present study suggests that free cafestol and cafestol-loaded PLGA nanoparticles may attenuate selected inflammatory and behavioral changes associated with LPS administration. The nanoparticle formulation demonstrated suitable preliminary physicochemical properties and satisfactory encapsulation efficiency, while molecular docking identified a potentially favorable interaction between cafestol and NF-κB. These findings support further preclinical investigation but do not yet establish the clinical efficacy of cafestol as an antidepressant or demonstrate the superiority of its nanoparticle formulation.

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