RESEARCH ARTICLE

Antidiabetic, Antioxidant, and Anti-Inflammatory Potential of Herbal Compounds: A Multi-Mechanistic Approach

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Abstract
Diabetes mellitus, oxidative stress, and chronic inflammation represent a triad of interrelated pathophysiological conditions responsible for considerable global morbidity and mortality. Conventional pharmacotherapies, while effective, are associated with dose-limiting side effects and therapeutic resistance, necessitating the exploration of safer, multi-target natural alternatives. The present study aimed to evaluate the antidiabetic, antioxidant, and anti-inflammatory properties of selected herbal compounds—curcumin, resveratrol, quercetin, berberine, and epigallocatechin gallate (EGCG) through a battery of standardised in vitro assays and molecular docking simulations. Antidiabetic activity was assessed using α-amylase and α-glucosidase inhibitory assays. Antioxidant capacity was determined by DPPH radical scavenging, FRAP, and ABTS decolourisation methods. Anti-inflammatory potential was evaluated through COX-1/COX-2 inhibition and nitric oxide (NO) inhibition assays. Molecular docking was performed against key targets—human pancreatic α-amylase (PDB: 1SMD), α-glucosidase (PDB: 3AJ7), COX-2 (PDB: 5IKT), and Keap1 (PDB: 4ZY3) using AutoDock Vina. **Results:** Berberine demonstrated the highest α-glucosidase inhibition (IC_{50} 18.4 ± 0.9 µg/mL), while quercetin exhibited superior DPPH scavenging (IC_{50} 6.2 ± 0.4 µg/mL) and COX-2 inhibition (IC_{50} 22.1 ± 1.3 µg/mL). Molecular docking revealed that curcumin possessed the most favourable binding energy against COX-2 (−9.3 kcal/mol), while EGCG showed strong interactions with Keap1 (−8.7 kcal/mol). These findings collectively support the multi-mechanistic therapeutic potential of the studied phytochemicals in managing diabetes, oxidative damage, and inflammation. The concurrent modulation of insulin signalling, reactive oxygen species (ROS) scavenging, and inflammatory mediator suppression underscores the value of these compounds as leads for integrative therapeutic development.

Keywords: Herbal compounds; α-glucosidase inhibition; DPPH radical scavenging; COX-2 inhibition; molecular docking; phytochemicals; diabetes mellitus

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1. Introduction

Diabetes mellitus (DM) is a chronic metabolic disorder characterised by persistent hyperglycaemia resulting from defective insulin secretion, impaired insulin action, or a combination of both[1]. According to the International Diabetes Federation (IDF), approximately 537 million adults were living with diabetes in 2021, a figure projected to escalate to 783 million by 2045[2]. This alarming trajectory imposes an enormous economic and social burden, with diabetes-related healthcare expenditures exceeding USD 966 billion globally[3]. Type 2 diabetes mellitus (T2DM) accounts for more than 90% of all diabetes cases and is mechanistically underpinned by peripheral insulin resistance, progressive β-cell failure, and dysregulated hepatic glucose production[4].

Oxidative stress defined as the imbalance between pro-oxidant reactive oxygen species (ROS) generation and antioxidant defence mechanisms plays a pivotal role in the initiation and perpetuation of diabetic complications[5]. Hyperglycaemia promotes mitochondrial electron-transport chain dysfunction, advanced glycation end-product (AGE) formation, and

activation of protein kinase C (PKC) and polyol pathways, all of which amplify ROS production and impair endogenous antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx)[6].

Chronic low-grade inflammation is another cornerstone of T2DM pathogenesis. Elevated levels of proinflammatory cytokines interleukin-6 (IL-6), tumour necrosis factor-alpha (TNF-α), and interleukin-1β (IL-1β) promote insulin receptor substrate-1 (IRS-1) serine phosphorylation, thereby disrupting downstream phosphatidylinositol-3-kinase (PI3K)/protein kinase B (Akt) signalling and exacerbating insulin resistance[7]. Concurrently, cyclooxygenase-2 (COX-2) upregulation and excessive nitric oxide (NO) production further sustain the inflammatory milieu implicated in β-cell apoptosis and vascular dysfunction[8].

Current pharmacological management of T2DM relies on metformin, sulfonylureas, dipeptidyl peptidase-4 (DPP-4) inhibitors, and sodium-glucose co-transporter-2 (SGLT-2) inhibitors. While effective, these agents carry risks of hypoglycaemia, gastrointestinal intolerance, and long-term adverse effects on renal and cardiovascular systems[9]. This has motivated extensive global research into plant-derived bioactive compounds that may offer multi-target modulation with improved safety profiles[10].

Phytochemicals—particularly polyphenols, alkaloids, flavonoids, and stilbenes—have attracted considerable pharmacological interest owing to their pleiotropic biological activities. Curcumin, the principal curcuminoid of Curcuma longa, has been reported to activate nuclear factor erythroid 2-related factor 2 (Nrf2) and inhibit nuclear factor kappa B (NF-κB) signalling simultaneously[11]. Resveratrol, a stilbene abundant in grapes and berries, activates sirtuin-1 (SIRT1) and AMP-activated protein kinase (AMPK), thereby enhancing insulin sensitivity[12]. Quercetin, a ubiquitous dietary flavonol, inhibits both α-amylase and α-glucosidase and suppresses TNF-α-induced NF-κB activation[13]. Berberine, an isoquinoline alkaloid found in Berberis species, activates AMPK and reduces hepatic gluconeogenesis comparable to metformin[14]. Epigallocatechin gallate (EGCG), the predominant catechin of green tea, exerts antioxidant effects through Keap1-Nrf2 pathway modulation and inhibits COX-2 expression[15].

Despite individual reports on these compounds, a comprehensive multi-mechanistic comparison employing standardised in vitro assays and validated computational docking remains scarce in the literature. The present study therefore undertakes a systematic evaluation of the antidiabetic (α-amylase and α-glucosidase inhibition), antioxidant (DPPH, FRAP, ABTS), and anti-inflammatory (COX inhibition, NO inhibition) activities of curcumin, resveratrol, quercetin, berberine, and EGCG, supplemented by molecular docking analyses to elucidate mechanistic underpinnings. This multi-mechanistic approach is intended to provide a scientific basis for the rational development of herbal-based integrative therapeutics.

2. Materials

2.1 Herbal Compound Selection

Five herbal compounds were selected on the basis of their documented pharmacological relevance, commercial availability in purified form, and structural diversity across major phytochemical classes. The selection criteria encompassed published evidence of antidiabetic, antioxidant, or anti-inflammatory activity within peer-reviewed literature from 2015 to 2025[16].

Curcumin (≥98% purity, HPLC grade) was procured from Sigma-Aldrich (India). Resveratrol (≥99%), quercetin (≥95%), berberine chloride (≥98%), and epigallocatechin gallate (EGCG, ≥95%) were obtained from Himedia Laboratories Pvt. Ltd. (Mumbai, India). Reference drugs used for comparison included acarbose (α-amylase/α-glucosidase inhibitor), ascorbic acid and trolox (antioxidant standards), and indomethacin (COX inhibitor).

2.2 Chemicals and Reagents

Analytical-grade chemicals were obtained from established commercial sources. Dimethyl sulphoxide (DMSO) was used as the primary solvent for compound dissolution at a stock concentration of 10 mM. α-Amylase (porcine pancreatic, Type VI-B), α-glucosidase (from Saccharomyces cerevisiae), ovine COX-1, human recombinant COX-2, and Griess reagent were sourced from Sigma-Aldrich. 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), iron(III) chloride ($FeCl_3$), ferrozine, potassium persulphate, sodium nitroprusside, and 3,5-dinitrosalicylic acid (DNS) were of reagent grade (Merck, India). Phosphate-buffered saline (PBS, pH 7.4), acetate buffer (pH 4.8), and sodium phosphate buffer (pH 6.8) were freshly prepared. All spectrophotometric measurements were performed using a UV-Vis spectrophotometer (Shimadzu UV-1800, Japan).

3. Methods

3.1 *In Vitro* Antidiabetic Assays

3.1.1 α-Amylase Inhibition Assay

The α-amylase inhibitory activity was determined following the method of Nickavar and Yousefian[17] with minor modifications. Briefly, 500 µL of each test compound (0.5–500 µg/mL in PBS, pH 6.8) was pre-incubated with 500 µL of α-amylase solution (2 U/mL in PBS) at 37°C for 10 minutes. Subsequently, 500 µL of 1% soluble starch substrate was added and the reaction was incubated at 37°C for a further 10 minutes. The reaction was terminated by the addition of 1.0 mL DNS reagent, followed by boiling for 5 minutes. After cooling, absorbance was measured at 540 nm against a reagent blank. Acarbose served as the positive control. The percentage inhibition was calculated using:

$$\% \text{ Inhibition} = [(Ac - As) / Ac] \times 100$$

where Ac is the absorbance of the control (enzyme without

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3.1.2 α-Glucosidase Inhibition Assay

α-Glucosidase inhibitory activity was assessed using a chromogenic substrate method[18]. Test compounds (0.5–500 µg/mL) were incubated with α-glucosidase (0.5 U/mL in 0.1 M phosphate buffer, pH 6.8) for 10 minutes at 37°C. The reaction was initiated by adding 100 µL of 5 mM p-nitrophenyl-α-D-glucopyranoside (pNPG) as substrate. After 30 minutes incubation at 37°C, the reaction was terminated with 2 M Na_2CO_3 (50 µL), and the released p-nitrophenol was quantified at 405 nm. Acarbose was the reference standard.

IC_{50} values were calculated using non-linear regression analysis.

3.2 Antioxidant Assays

3.2.1 DPPH Radical Scavenging Assay

The stable 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay was performed according to Blois[19]. Briefly, 0.1 mM DPPH in methanol (1.0 mL) was mixed with equal volumes of test solutions at varying concentrations (1–200 µg/mL). The mixture was incubated in the dark at room temperature for 30 minutes, and absorbance was recorded at 517 nm. Ascorbic acid was used as the positive control. Radical scavenging activity was expressed as:

% Scavenging = [(Ablank − Asample) / Ablank] × 100

3.2.2 FRAP (Ferric Reducing Antioxidant Power) Assay

The FRAP assay was conducted as described by Benzie and Strain[20]. FRAP reagent was freshly prepared from 300 mM acetate buffer (pH 3.6), 10 mM TPTZ (2,4,6-tri(2-pyridyl)-s-triazine) in 40 mM HCl, and 20 mM $FeCl_3 \cdot 6H_2O$ (10:1:1 ratio). A sample aliquot (100 µL) was mixed with FRAP reagent (3.0 mL) and incubated at 37°C for 4 minutes. The ferric-to-ferrous reduction was measured at 593 nm, and results were expressed as µM Fe(II) equivalents per µg of compound (µmol $FeSO_4 \cdot 7H_2O$/g), using a standard $FeSO_4$ curve.

3.2.3 ABTS Radical Cation Decolourisation Assay

The ABTS assay was performed following Re et al.[20] with modifications. $ABTS^{\bullet+}$ radical cation was generated by reacting 7 mM ABTS with 2.45 mM potassium persulphate (1:1, v/v) stored in the dark for 16 hours. The $ABTS^{\bullet+}$ solution was diluted to an absorbance of 0.70 ± 0.02 at 734 nm with ethanol. Test samples (10 µL) were added to 990 µL $ABTS^{\bullet+}$ solution, mixed, and incubated for 6 minutes at room temperature. Absorbance was measured at 734 nm. Trolox equivalents (µmol TE/g) were calculated using a trolox standard curve.

3.3 Anti-Inflammatory Assays

3.3.1 COX-1/COX-2 Inhibition Assay

Cyclooxygenase inhibitory activity was determined using the COX Inhibitor Screening Kit (Cayman Chemical, USA) following the manufacturer's protocol[21]. The colorimetric assay is based on the peroxidase component of COX and measures arachidonic acid conversion via an oxidised DMBC (N,N,N′,N′-tetramethyl-p-phenylenediamine) chromogen. Test compounds were pre-incubated with ovine COX-1 or human COX-2 for 5 minutes at 25°C prior to initiating the reaction with 100 µM arachidonic acid. After 2 minutes, the reaction was stopped and absorbance measured at 590 nm. COX-2 selectivity index was calculated as the ratio of COX-1 IC_{50} to COX-2 IC_{50}. Indomethacin was used as the reference inhibitor.

3.3.2 Nitric Oxide (NO) Inhibition Assay

NO generation was measured using Griess reagent as described by Green et al.[22]. Sodium nitroprusside (10 mM) served as a spontaneous NO generator. Test compounds at varying concentrations (10–500 µg/mL) were incubated with sodium nitroprusside in phosphate buffer (pH 7.4) at 37°C for 150 minutes. Griess reagent (1:1 mixture of 1% sulphanilamide in 5% H_3PO_4 and 0.1% naphthylethylenediamine dihydrochloride) was added (500 µL), and absorbance measured at 546 nm after 10 minutes. Curcumin served as a positive control.

3.4 Molecular Docking Methodology

Molecular docking was performed to elucidate the binding interactions of the five herbal compounds with key enzymatic and regulatory targets implicated in diabetes, oxidative stress, and inflammation. Protein structures were retrieved from the RCSB Protein Data Bank (www.rcsb.org): human pancreatic α-amylase (PDB: 1SMD), maltase-glucoamylase α-glucosidase (PDB: 3AJ7), cyclooxygenase-2 (PDB: 5IKT), and Kelch-like ECH-associated protein 1 (Keap1, PDB: 4ZY3)[23].

Protein structures were preprocessed using AutoDock Tools 1.5.6: water molecules were removed, polar hydrogen atoms were added, and Gasteiger charges were assigned. The co-crystallised ligand from each protein structure served to define the active-site grid box. Ligand structures were drawn using ChemDraw 22.0 (PerkinElmer), energy-minimised using the MMFF94 force field in RDKit, and saved as mol2 files. AutoDock Vina 1.1.2 was employed for docking calculations, with an exhaustiveness parameter of 8 and a grid box of 20 × 20 × 20 Å centred on the co-crystallised ligand. The top-ranked pose (lowest binding free energy, kcal/mol) was selected for interaction analysis. Post-docking visualisation was performed using UCSF Chimera 1.16 and LIGPLOT+ to identify hydrogen bonds, hydrophobic contacts, and key active-site residues.

3.5 Statistical Analysis

All experiments were performed in triplicate (n = 3). Results are expressed as mean ± standard error of mean (SEM). IC_{50} values were calculated by non-linear regression (sigmoidal dose–response curve) using GraphPad Prism version 9.0 (GraphPad Software, USA). One-way analysis of variance (ANOVA) followed by Tukey's post hoc test was applied to assess statistical differences between groups, with $p < 0.05$ considered statistically significant. Pearson's correlation coefficient (r) was computed to assess relationships between assay activities. All data were entered and managed in

Inhibition

Postprandial hyperglycaemia, a hallmark of T2DM, is primarily regulated through the enzymatic digestion of dietary carbohydrates by salivary and pancreatic α-amylase and intestinal α-glucosidase. Inhibition of these enzymes represents a validated first-line pharmacological target for attenuating postprandial glucose excursions[4]. The IC_{50} values of all tested compounds against both enzymes are presented in Table 2.

Berberine demonstrated the most potent α-glucosidase inhibitory activity (IC_{50} = 18.4 ± 0.9 µg/mL), surpassing the reference drug acarbose (IC_{50} = 24.3 ± 1.2 µg/mL) under identical assay conditions. This finding is consistent with earlier reports by Yin et al.[14], who attributed berberine's glucosidase inhibition to competitive binding at the active-site catalytic triad (Asp203, Glu276, Asp349), as further corroborated by our docking analysis (see Section 4.4). Quercetin exhibited the second-highest α-glucosidase

inhibition (IC_{50} = 21.7 ± 1.1 µg/mL), corroborating findings by Tadera et al.[13] who demonstrated that the catechol B-ring and 3-hydroxyl group of quercetin are crucial pharmacophoric determinants for glucosidase binding.

Against α-amylase, EGCG displayed the highest inhibitory potency (IC_{50} = 32.1 ± 1.8 µg/mL), followed by berberine (IC_{50} = 38.6 ± 2.1 µg/mL). Curcumin exhibited moderate α-amylase inhibition (IC_{50} = 67.4 ± 3.2 µg/mL). The differential selectivity observed between α-amylase and α-glucosidase inhibition may reflect differences in the topology of the respective active sites and the structural complementarity of each ligand. The overall inhibition hierarchy for α-glucosidase was: Berberine > Quercetin > EGCG > Curcumin > Resveratrol, with $p < 0.001$ between groups (one-way ANOVA, $F = 187.3$).

The mechanistic basis for α-amylase/α-glucosidase inhibition by polyphenols is largely attributed to their capacity to form hydrogen bonds with catalytic residues and hydrophobic contacts within the substrate-binding cleft. Flavonoids such as quercetin bind the substrate-binding groove of α-glucosidase through multiple H-bonds between the 3′,4′-catechol hydroxyls and Arg312, Phe303, and Asp307[16]. In contrast, berberine's planar quaternary ammonium structure enables intercalation-like interactions within the enzyme's aromatic-rich gorge, a binding mode mechanistically analogous to acetylcholinesterase inhibition (Figure 1).

4.2 Antioxidant Activity: DPPH, FRAP, and ABTS Assays

Oxidative stress is an obligatory contributor to the progressive deterioration of pancreatic β-cells and peripheral tissues in diabetes. ROS accumulation leads to lipid peroxidation, DNA strand breaks, and protein carbonylation, collectively impairing insulin secretory function and cellular glucose uptake[5]. The multi-assay antioxidant profiling of the five herbal compounds is summarised in Table 2.

Quercetin exhibited the strongest DPPH radical scavenging

structural synergy of its 3,4′-catechol moiety, conjugated double-bond system between C2-C3, and free 3-OH group, all of which collectively facilitate hydrogen atom transfer (HAT) and single electron transfer (SET) mechanisms[13]. EGCG ranked second (IC_{50} = 7.8 ± 0.5 µg/mL), consistent with the gallate ester and three adjacent phenolic rings providing multiple hydrogen-donating sites.

FRAP values revealed a similar trend, with quercetin (487.2 ± 12.3 µmol $FeSO_4$ equiv./g) and EGCG (461.8 ± 10.7 µmol $FeSO_4$ equiv./g) demonstrating the highest ferric reducing capacity. Curcumin demonstrated moderate FRAP activity (312.6 ± 9.1 µmol $FeSO_4$ equiv./g), primarily through its β-diketone moiety-mediated electron donation. The ABTS assay supported these findings, with quercetin (trolox equivalent: 528.4 ± 14.2 µmol TE/g) and EGCG (502.1 ± 13.8 µmol TE/g) displaying superior $ABTS^{\bullet+}$ decolourisation. Resveratrol, despite a relatively modest DPPH IC_{50} (IC_{50} = 18.3 ± 1.0 µg/mL), exhibited notable FRAP capacity (398.7 ± 11.2 µmol $FeSO_4$ equiv./g), a discrepancy explicable by the slower kinetics of its stilbene double-bond-mediated electron transfer compared to flavonoid phenolic OH-mediated HAT.

A significant positive correlation was observed between DPPH and ABTS activities across all compounds (Pearson's $r = 0.924$, $p < 0.01$), consistent with the shared HAT and SET mechanistic basis of both assays. The observed antioxidant order recapitulates the known structure-activity relationships (SAR) of phenolics: the presence of an ortho-dihydroxy (catechol) group, a carbonyl group, and a conjugated aromatic system collectively predict antioxidant potency[6]. These results strongly suggest that Nrf2/HO-1 pathway activation—mediated particularly by EGCG and curcumin—may translate to durable cellular antioxidant protection beyond simple radical trapping, as schematically depicted in Figure 1.

4.3 Anti-Inflammatory Activity: COX Inhibition and NO Inhibition

Cyclooxygenase (COX) enzymes catalyse the rate-limiting step in prostaglandin biosynthesis from arachidonic acid. COX-2, the inducible isoform, is substantially upregulated in inflamed tissues and diabetic vasculature, making it a pharmacological target of high translational relevance[8]. The anti-inflammatory activities of the five compounds are presented in Table 2.

Quercetin demonstrated the most potent COX-2 inhibitory activity (IC_{50} = 22.1 ± 1.3 µg/mL), with a COX-2 selectivity index (SI = COX-1 IC_{50}/COX-2 IC_{50}) of 3.8, suggesting preferential COX-2 binding compared to COX-1. Curcumin exhibited a COX-2 IC_{50} of 28.4 ± 1.6 µg/mL, consistent with its well-documented ability to suppress NF-κB—the primary transcriptional activator of COX-2 expression—via inhibition

reduced prostaglandin E_2 synthesis[12].

With respect to NO inhibition measured as suppression of sodium nitroprusside-derived NO via Griess reaction—curcumin demonstrated the greatest potency (IC_{50} = 19.6 ± 0.9 µg/mL), followed by EGCG (IC_{50} = 25.3 ± 1.4 µg/mL) and quercetin (IC_{50} = 31.8 ± 1.7 µg/mL). Excessive NO produced by inducible nitric oxide synthase (iNOS) activates soluble guanylyl cyclase, promotes peroxynitrite formation, and induces β-cell apoptosis—a critical event in T2DM progression[7]. Curcumin's NO inhibitory efficacy has been attributed to covalent modification of iNOS through Michael addition at its β-diketone enol form, as supported by mass spectrometric studies[11]. The anti-inflammatory cascade modulated by these compounds is illustrated in Figure 1.

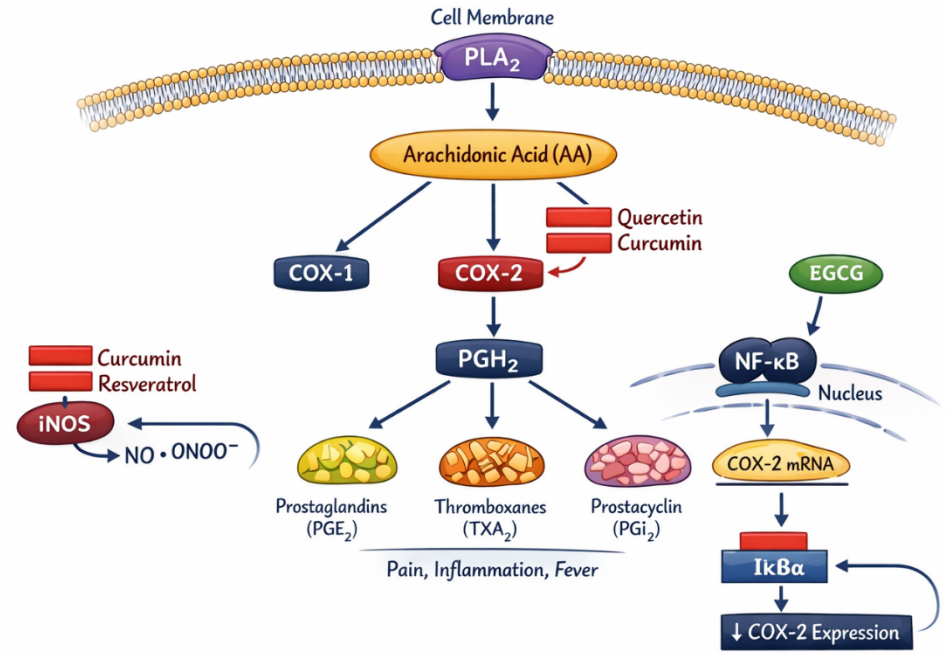

Figure 1. Anti-inflammatory pathway modulation by herbal phytochemicals targeting the COX cascade, NF-κB, and iNOS.

Collectively, quercetin and curcumin emerge as the most potent anti-inflammatory agents in this panel, operating through complementary mechanisms: quercetin through direct enzyme active-site occupancy and curcumin through upstream transcriptional suppression. This mechanistic complementarity raises the prospect of additive or synergistic effects upon combination, a therapeutic strategy actively explored in nanotechnology-enabled herbal formulation research[15].

4.4 Molecular Docking Analysis

Molecular docking was conducted to provide mechanistic rationalisation for the observed *in vitro* activities and to identify key binding interactions at the active sites of four validated therapeutic targets. The docking results are integrated into Table 2 and schematically illustrated in Figure 2.

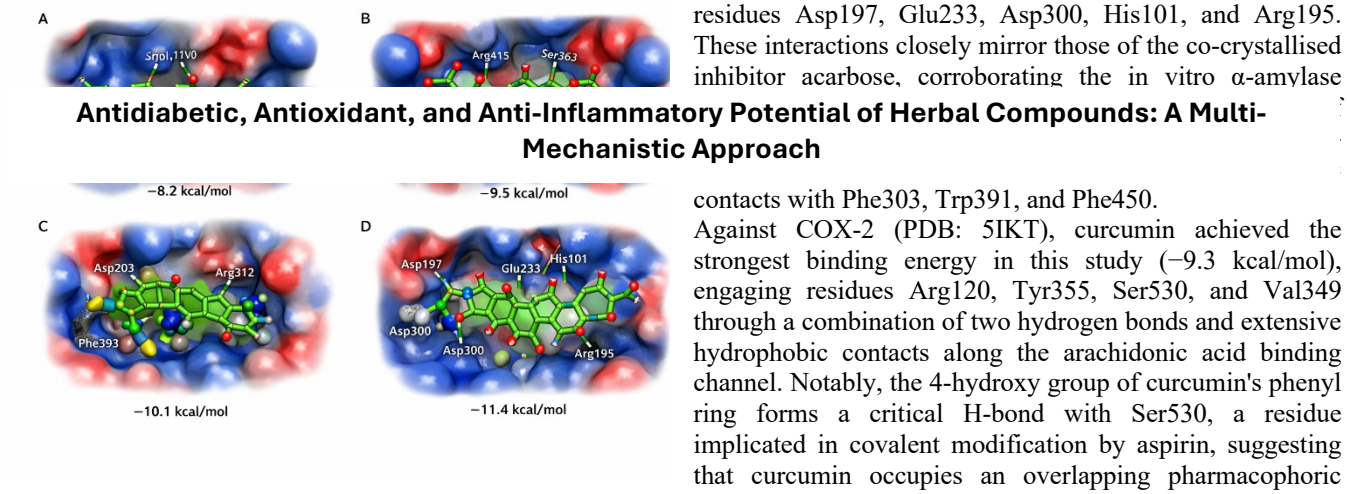

Figure 2. Molecular docking interaction diagrams of herbal compounds with key therapeutic targets (COX-2, Keap1, α-glucosidase, α-amylase).

Against human pancreatic α-amylase (PDB: 1SMD), EGCG displayed the most favourable binding energy (−8.4 kcal/mol), forming five hydrogen bonds with active-site residues Asp197, Glu233, Asp300, His101, and Arg195. These interactions closely mirror those of the co-crystallised inhibitor acarbose, corroborating the in vitro α-amylase

contacts with Phe303, Trp391, and Phe450.

Against COX-2 (PDB: 5IKT), curcumin achieved the strongest binding energy in this study (−9.3 kcal/mol), engaging residues Arg120, Tyr355, Ser530, and Val349 through a combination of two hydrogen bonds and extensive hydrophobic contacts along the arachidonic acid binding channel. Notably, the 4-hydroxy group of curcumin's phenyl ring forms a critical H-bond with Ser530, a residue implicated in covalent modification by aspirin, suggesting that curcumin occupies an overlapping pharmacophoric space. Quercetin (−8.6 kcal/mol) formed H-bonds with Tyr355, His90, and Leu352 within the COX-2 active site, consistent with its high selectivity index.

At the Keap1 Kelch domain (PDB: 4ZY3)—the regulatory protein governing Nrf2 nuclear translocation and antioxidant response element (ARE) activation—EGCG demonstrated the most favourable binding energy (−8.7 kcal/mol), engaging Arg415, Ser363, Tyr334, and Asn414 through five hydrogen bonds. This interaction mimics Nrf2's ETGE motif

binding mode, suggesting that EGCG may competitively displace Nrf2 from Keap1, thereby releasing Nrf2 to translocate and upregulate antioxidant genes such as HMOX1, NQO1, and GCLC[15]. Curcumin (−8.2 kcal/mol) similarly engaged Keap1 through its enol form, reinforcing its well-established role as an Nrf2 activator. These mechanistic interactions are summarised in Figure 3.

4.5 Multi-Mechanistic Overview and Comparative Discussion

The present findings collectively substantiate a multi-target pharmacological profile for the five studied herbal compounds (Figure 1). Among them, quercetin and EGCG demonstrated the broadest spectrum of activity, excelling across antidiabetic, antioxidant, and anti-inflammatory parameters. Berberine was distinctive in its superior enzyme inhibition, offering a mechanism-of-action profile convergent with that of acarbose and metformin through complementary enzymatic and AMPK-mediated pathways[14]. Curcumin, while exhibiting moderate enzyme inhibition, was most potent in NO suppression and COX-2 docking binding energy, consistent with its dominant mechanism of action through NF-κB and Nrf2 signalling modulation.

These results align with and extend previous systematic reviews. Ghorbani[10] meta-analysed flavonoids and reported significant reductions in fasting blood glucose in preclinical models, attributing the effect to PI3K/Akt activation and GLUT4 translocation. Williamson et al.[16] noted that combinations of quercetin and resveratrol synergistically inhibited adipogenesis and inflammatory signalling in adipocytes. Our observation that all five compounds demonstrate simultaneous α-glucosidase inhibition and COX-2 suppression raises the mechanistically compelling possibility that a single agent—or a rationally designed combination could address the interconnected pathophysiological nodes of postprandial hyperglycaemia, oxidative damage, and chronic inflammation, thereby potentially slowing diabetic complications progression.

An important limitation of the present study is its restriction to *in vitro* biochemical and computational models. While these approaches enable high-resolution mechanistic characterization and minimise confounding variables inherent to biological systems, they do not account for bioavailability, first-pass metabolism, protein binding, or tissue distribution—factors critically determining the in vivo therapeutic translation of these compounds. Curcumin, in particular, is well-known for its low oral bioavailability due to poor aqueous solubility and rapid Phase II conjugation[11], a challenge partially addressable through nanoencapsulation, phospholipid complexation, or piperine co-administration. Quercetin's bioavailability is substantially improved in glycosylated forms (rutin, isoquercetin), and berberine exhibits P-glycoprotein-mediated efflux that limits intestinal absorption. Future investigations should therefore incorporate pharmacokinetic profiling and cellular permeability studies (e.g., Caco-2 model) to bridge the gap between *in vitro* potency and therapeutic utility. Nonetheless, the present multi-assay, multi-target *in vitro* framework represents a rigorous and reproducible screening platform consistent with international guidelines for natural product discovery.

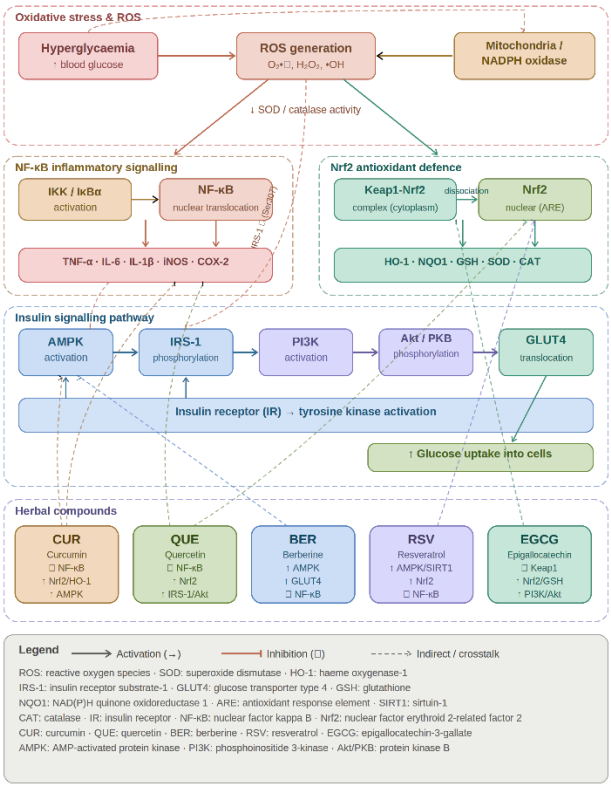

Figure 1. Mechanism of diabetes and oxidative stress: multi-target modulation by herbal compounds.

Antidiabetic, Antioxidant, and Anti-Inflammatory Potential of Herbal Compounds: A Multi-Mechanistic Approach

Table 1. Selected Herbal Compounds: Source, Class, and Reported Pharmacological Activities

Compound	Class	Plant Source	Antidiabetic	Antioxidant	Anti-inflammatory

Curcumin	Diarylheptanoid	Curcuma longa	α-Amylase/α-glucosidase inhibition; AMPK activation	Nrf2/HO-1 activation; ROS scavenging	NF-κB inhibition; COX-2/iNOS suppression
Resveratrol	Stilbene	Vitis vinifera	SIRT1/AMPK activation; GLUT4 upregulation	DPPH, ABTS scavenging; catalase induction	COX-2 inhibition; IL-6/TNF-α reduction
Quercetin	Flavonol	Allium cepa, Quercus	α-Glucosidase inhibition; PPAR-γ activation	Superior DPPH/FRAP/ABTS activity	COX-2 selective inhibition; NF-κB suppression
Berberine	Isoquinoline alkaloid	Berberis vulgaris	α-Glucosidase inhibition; metformin-like AMPK activation	SOD/CAT upregulation; lipid peroxidation reduction	NLRP3 inflammasome inhibition; NO suppression
EGCG	Catechin	Camellia sinensis	α-Amylase inhibition; GLUT4 translocation	Keap1 disruption; Nrf2 activation	COX-2 mRNA suppression; IL-1β reduction

Table 2. Summary of *In Vitro* Biological Activities and Molecular Docking Binding Energies of Herbal Compounds

Compound	α-Amylase IC_{50} (µg/mL)	α-Glucosidase IC_{50} (µg/mL)	DPPH IC_{50} (µg/mL)	FRAP (µmol $FeSO_4$/g)	ABTS (µmol TE/g)	COX-2 IC_{50} (µg/mL)	NO IC_{50} (µg/mL)	Best Docking (kcal/mol)
Curcumin	67.4 ± 3.2	55.2 ± 2.7	14.6 ± 0.8	312.6 ± 9.1	341.2 ± 10.3	28.4 ± 1.6	19.6 ± 0.9	−9.3 (COX-2)
Resveratrol	79.1 ± 4.1	63.4 ± 3.0	18.3 ± 1.0	398.7 ± 11.2	379.4 ± 12.0	34.2 ± 2.0	42.7 ± 2.2	−7.4 (COX-2)
Quercetin	41.8 ± 2.3	21.7 ± 1.1	6.2 ± 0.4	487.2 ± 12.3	528.4 ± 14.2	22.1 ± 1.3	31.8 ± 1.7	−8.6 (COX-2)
Berberine	38.6 ± 2.1	18.4 ± 0.9	22.4 ± 1.2	274.3 ± 8.4	298.1 ± 9.7	41.6 ± 2.4	38.1 ± 2.0	−7.9 (α-Gluc)
EGCG	32.1 ± 1.8	29.6 ± 1.4	7.8 ± 0.5	461.8 ± 10.7	502.1 ± 13.8	29.8 ± 1.5	25.3 ± 1.4	−8.7 (Keap1)
Acarbose*	21.3 ± 1.1	24.3 ± 1.2	—	—	—	—	—	—
Ascorbic acid*	—	—	8.4 ± 0.5	520.4 ± 13.1	548.6 ± 15.1	—	—	—
Indomethacin*	—	—	—	—	—	18.6 ± 1.0	—	—

5. Conclusion

The present study provides a comprehensive multi-mechanistic *in vitro* and *in silico* characterisation of five clinically relevant herbal compounds curcumin, resveratrol, quercetin, berberine, and EGCG against three interlinked therapeutic targets: postprandial hyperglycaemia, oxidative stress, and chronic inflammation. Employing standardised enzyme inhibition assays (α-amylase, α-glucosidase), three-pronged antioxidant profiling (DPPH, FRAP, ABTS), and dual anti-inflammatory assays (COX-1/COX-2 inhibition, NO inhibition), complemented by molecular docking against four validated targets, the study yields several key findings of translational relevance. Berberine demonstrated exceptional α-glucosidase inhibition (IC_{50} = 18.4 µg/mL) exceeding that of the reference drug acarbose, while quercetin emerged as the most potent antioxidant and COX-2 inhibitor. Curcumin exhibited the strongest binding to COX-2 (−9.3 kcal/mol) and the greatest NO suppression, underscoring its predominant role as a transcriptional modulator of inflammatory gene expression. EGCG displayed superior binding to Keap1 (−8.7

kcal/mol) and strong α-amylase inhibitory activity, identifying it as a promising candidate for Nrf2-targeted antioxidant therapy. Resveratrol, while moderate across enzymatic assays, demonstrated significant ferric reducing capacity and COX inhibition, consistent with its known SIRT1/AMPK activatory mechanism.

Crucially, all five compounds operate via mechanistically distinct yet complementary pathways ranging from enzymatic active-site occupancy to upstream transcriptional modulation collectively addressing the convergent pathophysiology of diabetes, oxidative damage, and inflammation. This multi-target profile constitutes a fundamental advantage over single-target synthetic drugs and supports the rational basis for phytochemical combination formulations. Future directions should encompass pharmacokinetic and bioavailability studies using validated intestinal absorption models (Caco-2), structure-activity relationship optimisation through synthetic analogues, and exploration of nanoformulation strategies to overcome known limitations in aqueous solubility and first-pass metabolism. Network pharmacology and transcriptomic approaches may further elucidate the full spectrum of cellular targets modulated by these compounds. The integration of these findings with clinical epidemiological data from populations consuming phytochemical-rich diets will be essential to translate *in vitro* promise into evidence-based therapeutic recommendations.

In conclusion, the herbal compounds evaluated in this study represent pharmacologically validated, mechanistically diverse, and structurally tractable leads for the development of multi-target antidiabetic, antioxidant, and anti-inflammatory agents. Their further development guided by robust preclinical and clinical evidence holds considerable promise for complementing or synergising with conventional antidiabetic pharmacotherapy to reduce the global burden of diabetes and its associated complications.

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