© 2024 Journal of Pharmacy & Pharmacognosy Research, 12 (3), 487-513, 2024
ISSN 0719-4250
https://jppres.com

DOI: https://doi.org/10.56499/jppres23.1841_12.3.487

Review

Andrographis paniculata: A potential supplementary therapy for cardiovascular diseases - A systematic review of its effects and molecular actions

[*Andrographis paniculata*: Una terapia suplementaria potencial para las enfermedades cardiovasculares - Una revisión sistemática de sus efectos y acciones moleculares]

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Abstract

Context: Cardiovascular diseases claim the lives of an estimated 17.9 million people worldwide (report by the World Health Organization), yet the drug pipeline compared to some other life-threatening diseases, including cancer and neurological disorders, is low.

Aims: To investigate the potential of *Andrographis paniculata* as a supplementary therapy for cardiovascular diseases based on recent *in vivo* animal studies.

Methods: This study adopted a systematic review approach to analyze preclinical evidence from *in vivo* animal studies. Three databases (PubMed, Scopus, and Embase) were searched using the keywords "*Andrographis paniculata*", "cardiovascular disease", "CVD", "heart disease", "cardioprotective", "cardio*", "inflammation", "oxidative stress", "obesity", "lipopolysaccharide", "hypertension", "arrhythmia" and "aortic disease". The search period was from April 20th, 2023, to April 26th, 2023, and included studies published from 2013 to 2023. Only *in vivo* animal studies were appraised. In contrast, clinical studies, *in vitro* studies, *in silico* studies, and review papers were excluded. SYRCLE's risk of bias tool was used to assess the risk of bias.

Results: Sixteen eligible *in vivo* animal studies showed that *Andrographis paniculata* extracts and isolated bioactive compounds have strong anti-inflammatory and antioxidant effects on cardiovascular diseases. These effects lead to lowering the risk of coronary artery disease and myocardial infarction, easing the effects of bad cardiac remodeling, stopping cardiac hypertrophy, and improving diabetic cardiomyopathy. Although SYRCLE's tool detected some bias, the studies were included since they met the inclusion criteria and had no conflicts of interest.

Conclusions: *Andrographis paniculata* may have the potential to be used as a supplementary therapy for cardiovascular diseases, but more animal studies and clinical trials should be done to establish these findings.

Keywords: *Andrographis paniculata*; animal models; cardiovascular diseases; herbal medicine; inflammation; oxidative stress.

Resumen

Contexto: Se calcula que las enfermedades cardiovasculares se cobran la vida de 17,9 millones de personas en todo el mundo (informe de la Organización Mundial de la Salud) y, sin embargo, el número de fármacos disponibles es bajo en comparación con otras enfermedades potencialmente mortales, como el cáncer y los trastornos neurológicos.

Objetivos: Investigar el potencial de *Andrographis paniculata* como terapia complementaria para enfermedades cardiovasculares basándonos en estudios recientes *in vivo* en animales.

Métodos: Este estudio adoptó un enfoque de revisión sistemática para analizar la evidencia preclínica de estudios *in vivo* en animales. Se realizaron búsquedas en tres bases de datos (PubMed, Scopus y Embase) utilizando las palabras clave "*Andrographis paniculata*", "cardiovascular disease", "CVD", "heart disease", "cardioprotective", "cardio*", "inflammation", "oxidative stress", "obesity", "lipopolysaccharide", "hypertension", "arrhythmia" y "aortic disease". El periodo de búsqueda fue del 20 de abril de 2023 al 26 de abril de 2023, e incluyó estudios publicados entre 2013 y 2023. Sólo se valoraron estudios *in vivo* en animales. Por el contrario, se excluyeron los estudios clínicos, los estudios *in vitro*, los estudios *in silico* y los artículos de revisión. Se utilizó la herramienta de riesgo de sesgo de SYRCLE para evaluar el riesgo de sesgo.

Resultados: Dieciséis estudios *in vivo* con animales demostraron que los extractos de *Andrographis paniculata* y los compuestos bioactivos aislados tienen potentes efectos antiinflamatorios y antioxidantes sobre las enfermedades cardiovasculares. Estos efectos conducen a la reducción del riesgo de enfermedad coronaria e infarto de miocardio, el alivio de los efectos de la mala remodelación cardiaca, la detención de la hipertrofia cardiaca y la mejora de la cardiomiopatía diabética. Aunque la herramienta SYRCLE detectó cierto sesgo, los estudios se incluyeron ya que cumplían los criterios de inclusión y no presentaban conflictos de intereses.

Conclusiones: *Andrographis paniculata* puede tener potencial para ser utilizada como terapia complementaria en enfermedades cardiovasculares, pero deben realizarse más estudios en animales y ensayos clínicos para establecer estos hallazgos.

Palabras Clave: *Andrographis paniculata*; modelos animales; enfermedades cardiovasculares; fitoterapia; inflamación; estrés oxidativo.

ARTICLE INFO
Received: October 11, 2023.
Accepted: December 30, 2023.
Available Online: January 30, 2024.

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Abbreviations: •OH: Hydroxyl radical; $^1O_2$: Singlet oxygen; 3-NT: 3-Nitrotyrosine; 4-HNE: 4-Hydroxynonenal; AKT: Protein kinase B (PKB); ALP: Alkaline phosphatase; AND: Andrographolide; ANP: Atrial natriuretic peptide; AP: *Andrographis paniculata;* AST: Aspartate aminotransferase; AVC: Aortic valve calcification; Bad: BCL-2-associated agonist of cell death; Bak: BCL-2-antagonist; BAT: Brown adipose tissue; BNP: B-type natriuretic peptide; BW: Body weight; CAD: Coronary artery disease; CAT: Catalase; CD45, CD68: Cluster of differentiation; CDER: Center for Drug Evaluation and Research; CHD: Coronary heart disease; CK: Creatine kinase; CK-MB: Creatine kinase-myoglobin binding; COX-2: Cyclooxygenase 2; CRP: c-Reactive protein; CTGF: Connective tissue growth factor; cTnI: Cardiac troponin 1; CVD: Cardiovascular disease; Cyt C: Cytochrome c; deAND: 14-deoxy-11,12-didehydroandrographolide; DOX: Doxorubicin; eNOS: Endothelial NO synthase; ET-1. Endothelin-1; FADD: Fas and Fas-associated via death domain; FAS: Fatty-acid synthase; FN: Fibronectin; GPx: Glutathione peroxidase; GSH: Glutathione; H_2O_2: Hydrogen peroxide; HDL-C: HDL-cholesterol; HFD: High-fat diet; HMGCR: 3-Hydroxy-3-methylglutaryl-coenzyme A reductase; HO-1: Heme oxygenase-1; HOM-IR: Homeostatic Model Assessment of Insulin Resistance; HW/BW: Heart weight to body weight; HW/TL: Heart weight to tibial length; IκBα: Inhibitor of *kappa* B alpha; ICAM-1: Intercellular cell adhesion molecule-1; IDLs: Intermediate-density lipoproteins; IFN-γ: Interferon-gamma; IGF-1R: Insulin-like growth factor 1 receptor; IL-10: Interleukin-10; IL-17: Interleukin-17; IL-1β: Interleukin-1β; IL-2: Interleukin-2; IL-6: Interleukin 6; iNOS: Inducible nitric oxide synthase; IRS: Insulin receptor substrate; LAD: Left anterior descending coronary artery; LDH: Lactate dehydrogenase; LDL: Low-density lipoproteins; LDL-C: LDL-cholesterol; LVEF: Left ventricular ejection fraction; LVESD: Left ventricular end-systolic diameter; LW/BW: Lung weight to body weight; MCP-1: Monocyte chemoattractant protein-1; MDA: Malondialdehyde; MGLL: Monoacylglycerol lipase; MHC-β: β-Myosin heavy chain; MI: Myocardial infarction; NFκB: Nuclear factor *kappa* B; NHS: National Health Service; NO: Nitric oxide; NOD: Non-obese diabetic; NOX2: NADPH oxidase 2; NOX4: NADPH oxidase 4; NQO1: Quinone oxidoreductase 1; Nrf2: Nuclear factor erythroid 2-related factor 2; O_2•−: Superoxide radical; ONOO-: Peroxynitrite; P13K: Phosphatidylinositol 3-kinase; PGI_2: Prostaglandin I_2; PIP3: Phosphatidylinositol (3,4,5)-triphosphate; PPAR-α: Peroxisome proliferator-activated receptor-alpha; PKB: protein kinase B (AKT); PRISMA: Preferred Reporting Items for Systematic Reviews and Meta-Analyses; ROS: Reactive oxygen species; SCD-1: Stearoyl-coenzyme A desaturase-1; α-SMA: α-Smooth muscle actin; SOD: Superoxide dismutase; SREBP-1, SREBP-2: Sterol regulatory element binding protein-1 and -2; T1D: Type 1 diabetes; T2D: Type 2 diabetes; TAX2: Thromboxane A2; TC: Total cholesterol; TG: Triglycerides; TGF-β1: Transforming growth factor-beta 1; Th1/Th2/Th17: T-helper 1, 2, and 17; TNF-α: Tumor necrosis factor-alpha; VCAM-1: Vascular cell adhesion molecule-1; VLDL: Very low-density lipoproteins; WHO: World Health Organization.

INTRODUCTION

Cardiovascular disease (CVD) refers to all conditions that negatively impact the heart and blood vessels, causing a decline in the normal functioning of the circulatory system. According to the National Health Service (NHS) of the United Kingdom, the four main CVDs are coronary heart disease, strokes, peripheral arterial disease, and aortic disease (Nhs.uk, 2022). The World Health Organization (WHO) has reported that CVDs claim the lives of an estimated 17.9 million people a year, posing a worldwide socio-economic burden and a global emergency (WHO, n.d.). Moreover, data from the WHO further highlights the severity of this issue in Indonesia, where CVDs are the leading cause of death among both males and females, with stroke and ischemic heart disease being the most prevalent forms (WHO, 2023). Some of the risk factors for CVDs, as highlighted by the WHO and NHS, are unhealthy diets, high blood pressure, physical inactivity, smoking, alcohol misuse, high cholesterol, diabetes, obesity, family history of CVDs, and ethnic background (Nhs.uk, 2022; WHO, n.d.).

The drug pipeline for cardiovascular diseases, compared to other diseases, such as cancer and neurologic disorders, is generally low (Mullard, 2020; Stern and Lebowitz, 2010). The number of new medications for CVDs introduced in the United States was relatively low in 2008 and 2009, with one and two new medications, respectively. In contrast, seven new oncology medications were introduced in 2009 (Stern and Lebowitz, 2010). Fast forward to 2019, and the FDA's Center for Drug Evaluation and Research (CDER) approved 48 new drugs. 23% of drugs were approved to treat cancer, 19% to treat neurological conditions, and only 2% to treat CVDs (Mullard, 2020). In 2022, only 6% of the 37 drugs approved by CDER were for the treatment of CVDs (Al-Madhagi, 2023). These statistics unveil the need for the discovery of new drugs to treat CVDs (at least supplementary treatment), particularly medications from plant sources, as those are relatively safer (with lesser side effects) than the conventional ones (Karimi et al., 2015).

Plants, particularly herbal plants, have been used for centuries to make medicines either in the form of plant extracts or as isolated, pure compounds (Fabricant and Farnsworth, 2001). Medications from plant sources have been seen to possess antioxidants and anti-inflammatory effects and are used for the supplementary treatment of CVDs such as atherosclerosis and hypertension (Shaito et al., 2020).

Andrographis paniculata (AP), a plant from the *Acanthaceae* family, is an herb largely found in regions of Southeast Asia, China, and America (Lattoo et al., 2006). AP is known by many names in different languages, including King of Bitters (English), Sambiloto (Bahasa Indonesia), Kalmegh (Bengali), Mahatikta (Sanskrit), Mahatita (Hindi), and Fai Talai Jone (Thai) (Okhuarobo et al., 2014). AP whole plant, aerial portion, and roots have a long history of use in traditional Asian medicine for several ailments such as stomachaches, inflammation, fever, intermittent fever, hypertension, malaria, cancer, diabetes, respiratory infections, influenza, and hepatitis (Chturvedi et al., 1983; Okhuarobo et al., 2014; Lattoo et al., 2006). AP contains several bioactive compounds that influence

its pharmacological activities (Salim et al., 2021). Andrographolide (AND), the commonly studied bioactive compound of AP, belongs to the diterpene lactone group. Some other members of this group include dehydroandrographolide, deoxyandrgraopholide, and neoandrographolide (Okhuarobo et al., 2014; Salim et al., 2021). Flavonoids including flavone-1, 2′-O-glucoside and 5-hydroxy-7, 8, 2′, 5′-tetramethoxyflavone and xanthones such as 1, 8-dihydroxy-3,7-dimethoxy-xanthone, 4,8-dihydroxy-2,7-dimethoxy-xanthone, 1,2-dihydroxy-6,8-dimethoxy-xanthone, and 3,7,8-trimethoxy-1-hydroxy-xanthone have been identified in AP (Okhuarobo et al., 2014).

Because it fights free radicals and inflammation, AP is one of the herbs being researched as a way to treat heart diseases like diabetic cardiomyopathy, atherosclerosis, coronary artery disease (CAD), cardiac hypertrophy, myocardial infarction, and many more (Al Batran et al., 2014; Hsieh et al., 2016; Liang et al., 2018; Shu et al., 2020; Xie et al., 2020). Although a review by Wong et al. (2021) reported the molecular mechanisms underlying the protective effects of andrographolide and AP against myocardial injury, this review fills a crucial gap by focusing on individual cardiovascular diseases and showcasing the specific effects and protective molecular actions of AP against each of them, drawing upon evidence from *in vivo* animal models. Furthermore, this review explored the interconnections between certain cardiovascular diseases, highlighting the potential of AP as an antagonist. It also focused solely on recent research and subjected each included study to a quality assessment. This review, however, consolidates and updates several other reports, including the report by Wong et al. (2021) on the protective effect of AP against cardiovascular diseases and encourages the application of AP as a supplementary treatment in clinical trials. A thorough database search was conducted in this study to compile reports that focused on AP's effects and molecular actions against cardiovascular-related diseases in animal models. The sole purpose of this study was to identify substantial evidence from recent reports supporting the use of AP as a supplementary therapy for treating CVDs.

MATERIAL AND METHODS

This systematic review was conducted and reported by the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (Page et al., 2021), but it is not yet registered in any international register.

Eligibility criteria

As population groups, this review looked at *in vivo* animal models (rodents and zebrafish) with heart diseases like high blood pressure, arrhythmia, cardiac hypertrophy, coronary artery diseases, myocardial infarction, and diabetic cardiomyopathy. Other animal models of cardiac injuries, such as doxorubicin-induced cardiotoxicity, transverse aortic constriction models, and left anterior descending coronary artery (LAD)-ligated models, were also included. Similarly, animal models with conditions that could lead to cardiovascular disease, such as a high-fat diet, obesity, and diabetes, were included. All non-cardiovascular-related disease models or animal studies with no focus on cardiovascular diseases were excluded. Animal models were used in this review because there are few or no clinical studies regarding this subject. Rodents (mainly) and zebrafish were included in the population because they possess characteristics similar to humans.

AP extracts and bioactive compounds constituted the intervention groups, including AND and 14-deoxy-11,12-didehydroandrographolide. Thus, all the included studies employed any of these interventions for the aforementioned population group. Studies utilizing AP in combination with other agents were excluded, as this review focused solely on the independent effects and molecular actions of AP and its bioactive compounds.

As a result, any papers that showed how AP or its bioactive compounds affected heart diseases were considered. The most preferred were reports that demonstrated both molecular actions and effects. Only studies reported in English and published within the last 10 years were included. *In vitro* studies, *in silico* studies, clinical studies, review papers, meta-analyses, and irrelevant study designs were excluded. Further, reports with multiple studies were included, but only the *in vivo* animal studies were appraised. Additionally, reports with multiple studies in which the mechanism of action was centered on the other studies and animal studies in which insufficient data was extracted were excluded.

Information sources

On April 20, 2023, OME searched three electronic databases: PubMed, Scopus, and Embase. The results of each search and search term used were sent to six reviewers (WA, SIW, SD, N, MS, and ML) to validate the search term and identify studies. On April 26, 2023, the reviewers reached a consensus and checked the databases for the last time, but narrowed the search to reports in the English language, published within the last 10 years, and animal studies, as shown in Table 1. Boolean operators (OR and AND) were utilized to broaden and refine search results, ensuring comprehensive coverage of relevant literature.

Table 1. The search term used for this study.

Database	Search term	Filters
PubMed	(Andrographis paniculata OR Andrographolide) AND (Cardiovascular disease OR CVD OR Heart disease OR Cardioprotective OR Cardio* OR Inflammation OR Oxidative stress OR Obesity OR Lipopolysaccharide OR hypertension OR Arrhythmia OR Aortic disease)	10 years; English language; other animals
1 Scopus	(Andrographis paniculata OR Andrographolide) AND (Cardiovascular disease OR CVD OR Heart disease OR Cardioprotective OR Cardio* OR Inflammation OR Oxidative stress OR Obesity OR Lipopolysaccharide OR hypertension OR Arrhythmia OR Aortic disease)	2013-2022; English language
Embase	(Andrographis paniculata OR Andrographolide) AND (Cardiovascular disease OR CVD OR Heart disease OR Cardioprotective OR Cardio* OR Inflammation OR Oxidative stress OR Obesity OR Lipopolysaccharide OR hypertension OR Arrhythmia OR Aortic disease)	2014-2023; *In vivo* study; Animal model

Search strategy

AP and cardiovascular diseases were searched for on Google. Candidate search terms were identified from the search results. A search strategy draft was developed using these search terms, and additional search terms were further discovered. The authors approved the search strategy. A thorough search of three databases—PubMed, Scopus, and Embase—was performed. The search terms used for all three databases were the same. The search terms were: "*Andrographis paniculata*", "cardiovascular disease", "CVD", "heart disease", "cardioprotective", "cardio*", "inflammation", "oxidative stress", "obesity", "lipopolysaccharide", "hypertension", "arrhythmia" and "aortic disease". The search was concluded on April 26, 2023. Filters such as "10 years", "English language," and "other animals" were applied to the PubMed search. In addition, filters such as "2013-2022" and "English language" were applied to the Scopus search. Further, "2014-2023", "*in vivo* study", and "animal model" were the filters used for Embase searches. More details on the search strategy can be found in Table 1. The bibliographies of all manuscripts relevant to this study were checked to identify publications that may have been missed.

Selection process

Three independent review authors (OME, WA, and SIW) reviewed the titles of the first 340 records and removed duplicate records (n = 16). In this same review, the authors further reviewed the abstracts of 324 records. Arising discrepancies, including a lack of coherence with the study selection criteria, were resolved by intensive discussions with the other authors (SD, N, MS, and ML) until a consensus was reached. Articles that did not meet the inclusion criteria of this study were removed (n = 301). Next, OME, WA, and SIW independently screened articles in full text (n = 23). Again, disagreements were resolved by discussions with SD, N, MS, and ML. The selection process ended with the inclusion of 16 articles.

Data collection process and the items

A data collection form was designed with Microsoft Word, modeled by Louisa et al. (2022), and employed to extract data from the *in vivo* animal studies. OME, WA, and ML independently extracted data from eligible studies using this form. Comparisons were made between the data extracted by the three authors, and discrepancies were resolved through discussions. Briefly, a complete review of each article was included after screening was concluded, and the following data were extracted: The first author's name, publication date, type of extract or main bioactive compounds, methods of extraction or bioactive preparation, yield of preparation from raw material or number of bioactive compounds in extracts, *in vivo* models used, treatment or duration, effect, molecular action, and toxicity. A summary of the data extracted is presented in Table 2. Additional data extracted included the countries where the experiments took place, funding sources, and evidence of potential conflicts of interest (Table S3). The effects and molecular actions of AP against cardiovascular diseases were the primary outcomes of this study.

Study risk of bias assessment

The risk of bias in the included studies was assessed using SYRCLE's risk of bias tool for animal studies (Hooijmans et al., 2014). This tool addresses ten specific domains: (1) sequence generation; (2) baseline characteristics; (3) allocation concealment; (4) random housing; (5) blinding (Caregiver/Investigator blinding); (6) random outcome assessment; (7) blinding (outcome assessor blinding); (8) incomplete outcome data; (9) selective outcome reporting; (10) other sources of bias.

Table 2. Summary of extracted data.

References	Types of extracts/main bioactive compound	Methods of extraction/bioactive preparation	The yield of preparation from raw material/amount of bioactive compounds in extracts	*In vivo* models used	Treatment/ duration	Molecular action	Effects	Toxicity
(Xie et al., 2020)	Andrographolide	Not reported	Not mentioned	C57/BL6 mice with LAD ligation	25mg/kg/day orally for 14 days	Reduced ANP, BNP, and β-MHC levels; reduced TGFβ and p-smad3; reduced the expression of Collagen I, Collagen III, and CTGF; decreased the amount of CD45 and CD68; reduced the expressions of p-IκBα and p-P65; decreased the expressions of TNF-α, IL-1β, IL-6, and MCP-1; reduced 4-HNE in cardiac tissue; increased the expression of SOD2 and decreased expression of Gp91; upregulated NQO1, Gpx, and SOD2; downregulated P67 phox, Gp91 and NOX4 in heart tissue; enhanced Nrf2 translocation to the nucleus; increased HO-1 expression	Antioxidant; anti-inflammation; downregulating cardiac hypertrophy and cardiac fibrosis; increased LVEF and decreased LVESD	Not reported
(Chen et al., 2020)	Ethanolic extract of *Andrographis paniculata* (AP extract); 14-Deoxy-11,12-didehydroandrographolide (dAND), Andrographolide (AND)	Methods described by lin et al., 2019 for AP extract and AND	dAND = 5.45 mg/g dried AP	HFD-induced obese C57BL/6JNarl mice	AP extract = 30 mg/kg BW/day for 16 weeks; AND = 55 mg/kg BW/day for 16 weeks; deAND = 30 mg/kg BW/day for 16 weeks	Blood levels of TNF-α were reduced; increased phosphorylation of IRS1, AKT, and AS160 in epididymal adipose tissue; enhanced translocation of GLUT4 Membrane in the epididymal adipose tissue	Anti-inflammation	None recorded
(Hsieh et al., 2016)	Water extract of *Andrographis paniculata*	Methods described by Chen et al., 2013	Not mentioned	HFD-induced pathological cardiac hypertrophy and apoptosis in C57/BL6 mice	2 g/kg/day for 1 week	Reduced COX2 levels; reduced p-IkB and NFκB; reduced ANP and BNP expressions in heart; deactivated pMRK5 and STAT3; decreased p-MRK1/2 and p-JNK; reduced collagen accumulation; Fas ligand, Fas, FADD. Caspase-8, and caspase-3; decreased accumulation of Bad, Bak and cytochrome c; increased IGFR protein level	Anti-inflammation; anti-hypertrophy; anti-apoptosis	Not reported

Table 2. Summary of extracted data (continued...)

References	Types of extracts/main bioactive compound	Methods of extraction/bioactive preparation	The yield of preparation from raw material/amount of bioactive compounds in extracts	*In vivo* models used	Treatment/ duration	Molecular action	Effects	Toxicity
(Tian et al., 2023)	Andrographolide	Not mentioned	Not stated	Transverse aortic constriction (TAC) Male C57 mice	100 and 200 mg/kg/day for 14 days.	Decreased plasma BNP and Ang II levels; reduced fibrosis in cardiac tissue; reduced tunnel-positive cells in heart tissues	Anti-apoptosis; Effect on hypertrophy and endoplasmic stress; LVESD and LVEDD were decreased; enhanced EF and FS of the left ventricle	Not reported
(Lin et al., 2020)	Andrographolide	Not mentioned	Not stated	HFD-induced obese 4-week-old C57/BL6 mice	25 mg/kg BW/ day and 50 mg/kg BW/day for 1 week	Suppressed elevated levels of Fas, FADD, Bak, and cytochrome c proteins; Increased IGF-1R in cardiac tissues	Anti-apoptosis	Not reported
(Wang et al., 2021)	Andrographolide	Not mentioned	Not stated	HFD-induced aortic valve calcific male ApoE-/- mice	10 mg/kg BW for 8 weeks	Decrease of MGLL and ALP	Amelioration of aortic valve calcification	Not reported
(Liang et al., 2018)	Andrographolide	Not mentioned	Not stated	Streptozotocin induced diabetes in 8-week-old C57/BL6J mice	1, 10, or 20 mg/kg/day for 12 weeks	Reduced collagen deposition in heart tissue; reduced the expression of collagen I, collagen III, TGF-β1, and fibronectin (FN) in the myocardium; downregulated ANP and BNP; downregulated the expressions of IL-6, ICAM-1, VCAM-1 in heart tissues; downregulated the expression of IL-1β, IL-6, and TNF-α; reduced SOD activity; reduced MDA and 4-HNE in the diabetic myocardium; suppressed diabetes-induced accumulation of 3-NT; downregulated the expression of iNOS in the diabetic myocardium; mitigated the expression of NADPH oxidases (NOX2, NOX4, and p47phox); increased Nrf2 and HO-1 levels increased phosphorylation of IκBα and p65NF-κB; decreased COX-2, gel shift assay confirmed the inhibition of NF-κB activation in the diabetic myocardium	Anti-cardiac inflammation, anti-cardiac oxidative stress inhibition; anti-cardiac apoptosis; Improved LVEF, FS, and E/A ratios	None recorded

Table 2. Summary of extracted data (continued...)

References	Types of extracts/main bioactive compound	Methods of extraction/bioactive preparation	The yield of preparation from raw material/amount of bioactive compounds in extracts	*In vivo* models used	Treatment/ duration	Molecular action	Effects	Toxicity
(Akhtar et al., 2016)	*Andrographis paniculata* water extract	Described by Muhammad et al, 2016	The extract was composed of Andrographolide (2.4 mg/25 mg of extract), dehydroandrographolide (2.1 mg/25 mg of extract), deoxyandrographolide (2.65 mg/25 mg of extract), Neoandrographolide (1.88 mg/25 mg of extract)	Non-genetic out-bred male Sprague-Dawley rat model	50 and 200 mg/kg BW	Normalized the metabolic profile (glucose, creatinine, lactate, allantoin, choline, taurine, formate, citrate, 2-oxoglutarate, dimethylamine, acetate, acetoacetate, succinate, and hippurate) of obese or obese diabetic rats in urine	Anti-diabetic and anti-obesity	None recorded
(Shu et al., 2020)	Andrographolide	Not mentioned	Not stated	Vitamin D3 induced High fat-diet model of male C57BL/6 mouse	10 and 50 mg/kg BW	Improved lipid profiles; downregulation of ET-1 and TAX2 serum levels; upregulation of NO and PGI_2 serum levels; elevated t-PA and lowered PAI-1 in serum; downregulated the expression of serum CRP, IL-1β TNF-α, MCP-1; Improved caspase-3 levels; up regulating PPARα and downregulating p65 and IκBα (regulation of NF-κB and PPAR signaling pathways)	Gradual repair of myocardial tissue; anti-myocardial inflammation; anti-cardiac apoptosis	Not reported
(Ding et al., 2014)	Andrographolide	Not mentioned	Not stated	C57BL/6 mice model of HFD-induced obesity	100 and 50 mg/kg per day	Improved lipid parameters in adipose tissue, liver, and blood; alleviated glucose and insulin resistance; Inhibited the pathway of Sterol Regulatory Element-Binding Protein (SREBP-1 and SREBP-2) (reduced expression of SREBP-2, HMGCR, FDPS, FDFT1, DHCR7 DHCR24, LSS, convertase subtilisin/kexin type 9, mevalonate kinase, sterol-C4-methyl oxidase-like, squalene epoxidase, proprotein and LDLR in liver tissue; reduced the expression of SREBP-1, ACC-1, FAS, SCD-1, ACL, acyl-CoA synthetase, FADS-1, GAPT, and FADS-2 in liver tissue); uncoupling protein 2 expression was enhanced in adipose tissue; the expressions of FAS, SCD-1, and HMGCR were reduced in adipose tissue	Reduced the accumulation of fat in the liver or adipose tissues; anti-dyslipidemia	Not reported

Table 2. Summary of extracted data (continued...)

References	Types of extracts/main bioactive compound	Methods of extraction/bioactive preparation	The yield of preparation from raw material/amount of bioactive compounds in extracts	*In vivo* models used	Treatment/ duration	Molecular action	Effects	Toxicity
(Zhang et al., 2015)	Andrographolide	Not mentioned	Not stated	LPS-induced myocardial malfunction in Male BALB/c mice model	10 mg/kg/day for 7 days	Decreased the over expression of TNF-α and IL-1β in heart; decreased cardiac NO levels; inhibited the activation of IκB; significantly suppressed myocardial apoptosis (decreased apoptotic cells and caspase-3/7 activities in the heart)	Anti-apoptosis; anti-inflammation; Improved left ventricular ejection fraction (EF) and fractional shortening;	Not reported
(Zhang et al., 2013)	Andrographolide	Not mentioned	Not stated	NOD mice model	50, 100, and 150 mg/kg body weight for four weeks and 22 weeks	Decreased IL-2 and IFN-γ production; increased TGF-β, IL-10, and reduced IL-17; elevated the mRNA expression of GATA3; decreased RORγt and T-bet and mRNA expression	Maintaining the homeostasis of Th1/Th2/Th17; Inhibited insulitis and suppressed diabetes development in NOD mice	Not reported
(Elasoru et al., 2021)	Andrographolide	Not mentioned	Not stated	Isoproterenol-induced myocardial infarction in male Wistar rats	20 mg/kg/day for 21 days	Reduced the high levels of CK, LDH, CK-MB, cTnI, and AST; increased GPx and catalase activities; improved cardiac histological alteration; preserved intracellular Ca^{2+} handling; prevented MI-associated APD changes in cardiomyocytes; prevented the elevation of $I_{Ca,L}$; increased conductance of the I_{to}	Antioxidant; prevents ventricular stress	Not reported
(Al Batran and Al-Bayaty, 2014)	Andrographolide	Not mentioned	Not stated	*Porphyromonas gingivalis* induced atherosclerosis in White New Zealand rabbits	10 and 20 mg/kg BW for over 12 weeks	Reversed liver and renal biochemical changes; improved serum lipid profile; increased GSH, GPx, SOD, and CAT levels in serum; decreased MDA levels; reduced serum levels of TNF-α; decreased levels of VCAM-1 and ICAM-1; increased levels of nitrotyrosine and decreased levels of MCP1; reduced intimal thickening plaque	Antioxidants and anti-inflammation	Not reported

Table 2. Summary of extracted data (continued...)

References	Types of extracts/main bioactive compound	Methods of extraction/bioactive preparation	The yield of preparation from raw material/amount of bioactive compounds in extracts	*In vivo* models used	Treatment/ duration	Molecular action	Effects	Toxicity
(Al Batran et al., 2014)	Andrographolide	Not mentioned	Not stated	*Porphyromonas gingivalis* induced atherosclerosis in White New Zealand rabbits	10 and 20 mg/kg BW for over 12 weeks	Improved lipid profile; reduced CRP serum levels; reduced levels of IL-1β and IL-6; reduced atherosclerotic lesions; reduced intimal layer and foam cells; elevated α-SMA protein expression; inhibited CD36	Anti-inflammation	No toxicity observed
(Wang et al., 2022)	Aqueous extract of *Andrographis paniculata*	Dissolved in 10 mL of Sterile Milli Q water, heated for 1 h (100°C). Centrifuged cooled boiled extract for 10 minutes at 5000 rpm.	Not stated	Doxorubicin induced toxicity in zebrafish	Doses of 5, 10, and 15 µg/mL	Improved SOD, glutathione, and catalase activities; decreased nitric oxide levels; decreased creatinine kinase and lactate dehydrogenase activity; impaired neutrophil infiltration	Antioxidants, anti-inflammation	Not reported

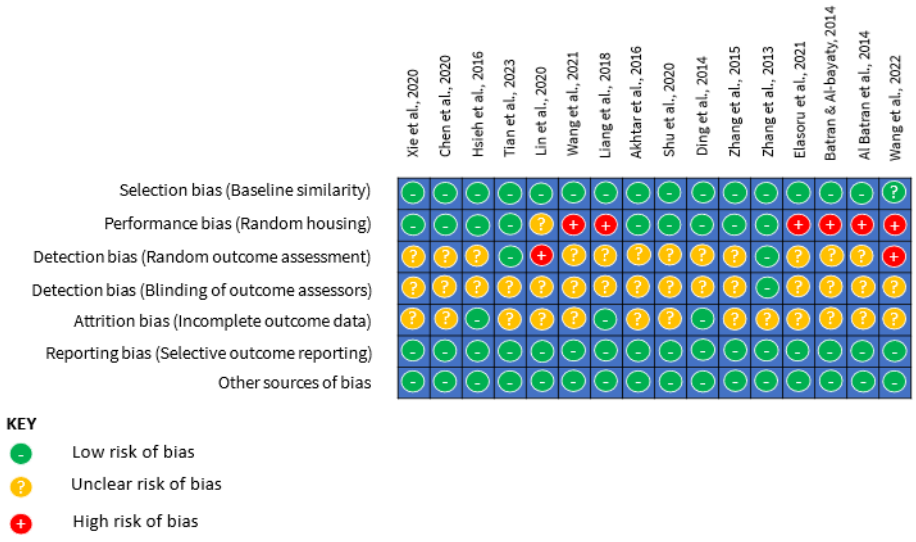

Figure 1. Summary of risk of bias.

This risk of bias tool comprehensively assesses several domains: baseline similarity, random housing, random outcome assessment, blinding of outcome assessors, incomplete outcome data, selective outcome reporting, and other sources of bias.

This tool formulates one question for every domain to allow reviewers to easily conclude the risk of bias for each domain by commenting yes, no, or unclear. OME, WA, and ML employed the tool independently to evaluate each of the included studies. Discussions were made to justify the judgments made in every study, and a more intense discussion resolved any discrepancies in judgments. This data is presented in the Table S2. In addition, a summary of the risk of bias covering the six types of bias, including selection bias, performance bias, detection bias, attrition bias, reporting bias, and other biases, is presented in Fig. 1. This summary was made using SYRCLE's tool and presented in a figure to allow readers to easily access the risk of bias within the included studies. Following discussions among OME, WA, and ML, a conclusion was made to include seven domains: selection bias (baseline similarity), performance bias (random housing), detection bias (random outcome assessment), detection bias (blinding of outcome assessors), attrition bias (incomplete outcome data), reporting bias (selective outcome reporting), and other sources of bias. Determining baseline similarity involved clearly defining the characteristics of the experimental and control populations. These criteria were as follows: (1) All animals in both groups must belong to the same species; (2) they must be within the same age group; and (3) their weights must be similar. Meanwhile, random housing, random outcome assessment, blinding of common assessors, and incomplete outcome data were assessed based on their mention in the studies. If not mentioned, they were considered "unclear" or high risk of bias based on the review authors collective decision. After thoroughly reading the manuscripts, the review authors independently determined selective outcomes and other biases. The risk of bias in each domain was categorized as low, unclear, or high.

Effective measures, synthesis methods, reporting bias assessment, certainty of evidence

Data from the populations, interventions, and outcomes were tabulated using Microsoft Word. This software was chosen because it is user-friendly. The effective measures were not performed due to the differences in each outcome in the studies. Thus, no pooled analysis of the cardiovascular effects was done. Whether each included study was free from selective outcome reporting was determined by comparing the methods and results sections. The certainty of the evidence was not achieved because there was no follow-up with meta-analysis in this study. To ensure the quality of the literature included in this study, the journals in which the articles were published were assessed for their prestige within the relevant field.

Data analysis

Four variables were evaluated: (1) *Andrographis paniculata* (with indicators including AP extract, andrographolide, and 14-deoxy-11,12-didehydroandrographolide), (2) cardiovascular diseases (with indicators including cardiac hypertrophy, aortic valve calcification, myocardial infarction, post-myocardial infarction atherosclerosis, cardiotoxicity, myocardial malfunctions, obesity, diabetes, and dyslipidemia), (3) effects (including anti-inflammation, antioxidant, anti-apoptosis, anti-fibrosis, anti-endothelial dysfunc-

tion, anti-cardiac hypertrophy, anti-dyslipidemia, anti-diabetes, anti-obesity, and reduced ventricular stress), and (4) molecular actions (encompassing all molecular interactions leading to desired effects such as anti-inflammation, anti-oxidative stress, and reduced ventricular stress). The operationalization of these variables is presented in Table S1.

RESULTS

Study selection and characteristics

The search of three databases, such as PubMed, Scopus, and Embase, identified 340 articles. 16 duplicates were detected and removed, and a total of 301 irrelevant studies to the review were excluded after being screened based on the abstracts. 23 studies were thoroughly reviewed for eligibility, and 8 studies were excluded. The study selection process yielded only 16 articles, as shown in Fig. 2. A summary of the effects of AP extract or its bioactive compounds on cardiovascular diseases was made from 16 articles appraising only *in vivo* studies.

Table 2 presents each included study. Many of the studies used bioactive compounds (mainly andrographolide) rather than plant extracts (Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014; Ding et al., 2014; Elasoru et al., 2021; Liang et al., 2018; Lin et al., 2020; Shu et al., 2020; Tian et al., 2023; Wang et al., 2021; Xie et al., 2020; Zhang et al., 2013; 2015). A study included the use of 14-deoxy-11,12-didehydroandrographolide (deAND) (Chen et al., 2020). Only two studies stated or cited the methods used to extract these bioactive compounds (Chen et al., 2020; Xie et al., 2020). Next, four manuscripts reported the use of AP plant extracts, including water extracts, ethanolic extracts, and aqueous extracts (Akhtar et al., 2016; Chen et al., 2020; Hsieh et al., 2016; Wang et al., 2022). The methods for preparing plant extracts in these four studies were described and well-cited. Although most of the papers reviewed reported the purity of the preparation, only three manuscripts reported the exact yields of the preparation or the number of bioactive compounds in the extracts (Akhtar et al., 2016; Chen et al., 2020).

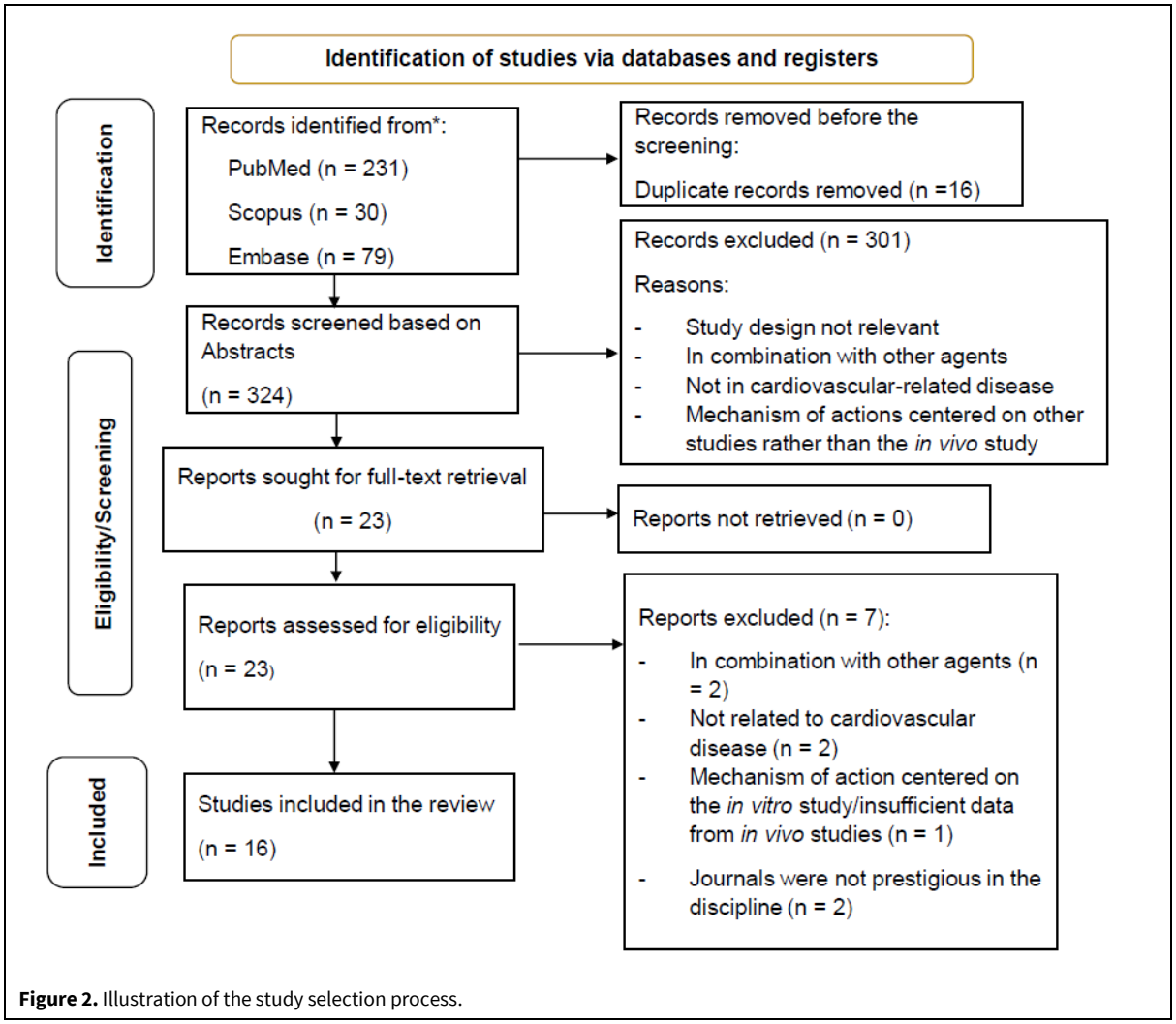

Figure 2. Illustration of the study selection process.

Risk of bias in studies and reporting bias

The baseline characteristics of the control and experimental populations were similar across all studies. Six studies had a high risk of bias in the performance bias domain (random sampling). Additionally, most studies had an unclear risk of bias in the random outcome assessment, blinding of outcome assessors, and incomplete outcome data domains. Notably, all included studies were free from selective outcome reporting and other sources of bias (Fig. 1 and Table S2). Despite some biases found in the animal studies, their inclusion in this review was justifiable due to their compliance with the established inclusion criteria and the absence of competing interests in the majority of studies. In addition, some of these studies were published in reputable journals, so their findings may be considered. The quality assessment was conducted to facilitate reader evaluation of the data sources utilized in this review, fostering a comprehensive understanding of their integrity and reliability.

Results of individual studies

The individual studies' results are presented in Table 2. These studies were further categorized below to provide context regarding the protective effect of AP against specific cardiovascular diseases or conditions that could lead to cardiovascular disease.

Effect of AP on hyperglycemia, insulin deficiency/resistance

In this study, some of the papers reviewed were on the effect of AP on diabetic cardiomyopathy, diabetes, obesity, hyperglycemia, insulin deficiency, and insulin resistance (Akhtar et al., 2016; Chen et al., 2020; Ding et al., 2014; Liang et al., 2018; Zhang et al., 2013). One of the studies revealed that AP extract and deAND significantly attenuated high-fat diet-induced hyperglycemia and insulin resistance. AP extract and deAND significantly restored the normal Homeostatic Model Assessment of Insulin Resistance (HOM-IR) values that were initially increased by the high-fat diet (HFD). AP and deAND positively affected the insulin signaling pathway in epididymal adipose tissue of mice fed with HFD by increasing the phosphorylation of insulin receptor substrate 1 (IRS1), protein kinase B (PKB/AKT), and AS160 (AKT substrate of 160 kDa), which enhanced the insulin stimulation of GLUT4 membrane translocation in the epididymal adipose tissue that was initially impeded by the HFD (Chen et al., 2020).

Another study reported that AND ameliorated diabetic cardiomyopathy by preventing myocardial remodeling induced by diabetes, alleviating nuclear factor *kappa* B (NFκB)-mediated inflammatory response in the myocardium of diabetic rats, and alleviating oxidative stress in the myocardium induced by diabetes. The prevention of myocardial remodeling induced by diabetes was characterized by a decrease in collagen deposition in the interstitial regions of the myocardium, decreased expression of markers of fibrosis such as fibronectin (FN), transforming growth factor-beta 1 (TGF-β1), collagen I, and collagen III, and a reduction in the expression of cardiac hypertrophy markers, including atrial natriuretic peptide (ANP) and B-type natriuretic peptide (BNP). In addition, the effect of AND in alleviating nuclear factor *kappa*-light-chain-enhancer of activated B cells (NFκB)-mediated inflammatory response in the myocardium of diabetic rats was evidenced by a decrease in phosphorylation of p65NFκB and nuclear factor kappa light polypeptide gene enhancer in B-cells inhibitor, alpha (IκBα); a decrease of cyclooxygenase 2 (COX-2); inhibition of pro-inflammatory cytokines including interleukin 6 (IL-6), interleukin-1β (IL-1β), and tumor necrosis factor-alpha (TNF-α); and inhibition of vascular cell adhesion molecule-1 (VCAM-1) and intercellular cell adhesion molecule-1 (ICAM-1). In addition, the inducible nitric oxide synthase (iNOS) expressions were inhibited. Additionally, suppressing the buildup of 3-nitrotyrosine (3-NT), increasing superoxide dismutase (SOD) activity, lowering the levels of lipid peroxidation markers like malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE), reducing the expression of NADPH oxidases (NOX2 and NOX4), and normalizing nuclear factor erythroid 2-related factor 2 (Nrf2) and heme oxygenase-1 (HO-1) were all indicators of an alleviated oxidative stress (Liang et al., 2018).

AND displayed several notable therapeutic activities in an autoimmune non-obese diabetic (NOD) mouse model. It effectively reduced insulitis significantly, delayed the onset of diabetes, and reduced its severity. These effects were correlated with the following molecular actions: reduced production of pro-inflammatory cytokines such as interferon-gamma (IFN-γ), interleukin-2 (IL-2), and interleukin-17 (IL-17); increased production of interleukin-10 (IL-10), an anti-inflammatory cytokine, and TGF-β in plasma; increased expression of the GATA3 gene but decreased the expression of the T-bet and RORγt genes in the pancreatic lymphatic nodes of the rats. It was shown that keeping T-helper 1, 2, and 17 (Th1/Th2/Th17) levels stable was important for stopping type 1 diabetes (T1D) (Zhang et al., 2013).

In a high-fat diet-induced obese mouse model, AND decreased the levels of Fas and Fas-associated

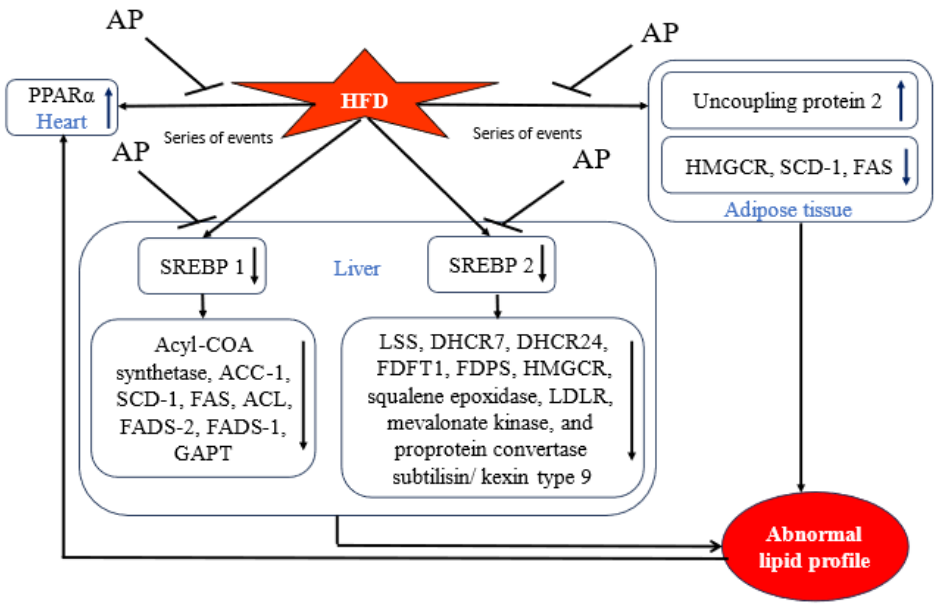

Figure 3. Proposed molecular mechanism of action of *Andrographis paniculata* (AP) on dyslipidemia.

In cardiac tissue, AP raises the levels of PPARα and uncoupling protein 2, which control lipid metabolism. In adipose tissues, AP lowers the levels of HMGCR, SCD-1, and FAS, which make fat. It also decreases the expressions of SREBP 1 and SREBP 2 (in the liver), which regulate fatty acid and cholesterol metabolism, respectively. In addition, AP decreases the target proteins of SREBP 1 and SREBP 2. These molecular actions are proof that AP decreases lipid synthesis and promotes the oxidation of fatty acids, thereby alleviating dyslipidemia.

via death domain (FADD) proteins (associated with the extrinsic apoptosis pathway) and decreased BCL-2-antagonist (Bak) and cytochrome c (associated with the intrinsic apoptosis pathway). The mice also had more of the insulin-like growth factor 1 receptor (IGF-1R) survival mechanism in their heart tissue, which helped them deal with and avoid HFD. AND treatment solidified cardioprotection by further elevating the IGF-1R survival mechanism (Lin et al., 2020).

In addition, AP extract normalized the metabolic profile of obese or obese-diabetic rats. More data on this can be found in Table 2 (Akhtar et al., 2016).

Effect of AP on dyslipidemia

In this study, some of the papers reviewed reported the effect of AP on abnormal lipid levels. AP extracts or AND alleviated dyslipidemia and restored the normal lipid profile level in the animal models (Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014; Shu et al., 2020).

AND treatment increased the expression of peroxisome proliferator-activated receptor-alpha (PPAR-α) in a high-fat-induced coronary heart disease (CHD) mouse model and enhanced the regulation of lipid metabolism (Shu et al., 2020).

In a C57BL/6 mouse model of HFD-induced obesity, treatment with AND decreased the expression of hepatic sterol regulatory element binding protein-2 (SREBP-2), which decreased the expression of other genes involved in cholesterol biosynthesis regulated by SREBP-2. Meanwhile, AND also decreased the expression of sterol regulatory element binding protein-1 (SREBP-1) and that of its target genes involved in the metabolism of fatty acids (Ding et al., 2014), as shown in Fig. 3. In the brown adipose tissue (BAT), AND treatment increased the expression of uncoupling protein 2 and decreased seropositivity for 3-hydroxy-3-methylglutaryl-coenzyme A reductase (HMGCR), stearoyl-coenzyme A desaturase-1 (SCD-1), and fatty-acid synthase (FAS) expressions, demonstrating that lipid synthesis was decreased, and fatty acid oxidation was promoted (Ding et al., 2014).

Effect of AP on coronary artery diseases

In a coronary heart disease mouse model, AND protected against cardiac injury and restored a normal lipid profile; decreased levels of TNF-α, IL-1β, monocyte chemoattractant protein-1 (MCP-1), and c-reactive protein (CRP); and diminished endothelial dysfunction by reducing blood levels of thromboxane A2 (TXA2), endothelin-1 (ET-1), and elevating prostaglandin I_2 (PGI_2) and nitric oxide (NO) levels (Shu et

al., 2020). AND greatly improved the blood lipid profile and lowered MCP-1 levels in the aortic homogenate of a rabbit model of atherosclerosis caused by *Porphyromonas gingivalis* (Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014). The atherogenic rabbits experienced liver and renal impairment, which was characterized by the elevation of liver enzymes, creatinine, and urea, respectively, in serum, which was normalized after AND treatment. Meanwhile, AND lowered MDA levels, increased glutathione (GSH) levels, and decreased ICAM-1, VCAM-1, and TNF-α serum levels. AND was also effective in elevating antioxidant enzyme levels such as SOD, catalase (CAT), and glutathione peroxidase (GPx) (Al Batran and Al-Bayaty, 2014). In addition, AND treatment reduced the expression of CRP and pro-inflammatory cytokines in the serum, reduced the expression of cluster of differentiation (CD36) in the aortic homogenate of the atherogenic rabbits, and revealed a potential to inhibit macrophages and the accumulation of lipids in rabbits with atherosclerosis (Al Batran et al., 2014).

An article reviewed reported that AND significantly reduced the expression of alkaline phosphatase (ALP) and monoacylglycerol lipase (MGLL) in a high-fat diet-induced aortic valve calcification mouse model (Wang et al., 2021).

Effect of AP on myocardial infarction (MI)/adverse cardiac remodeling (post-MI)

In a rat model of isoproterenol-induced myocardial infarction, AND reduced the elevated level of the cardiac injury systemic biomarkers, including creatine kinase (CK), creatine kinase-myoglobin binding (CK-MB), lactate dehydrogenase (LDH), aspartate aminotransferase (AST), cardiac troponin 1 (cTnI), and increased antioxidant activities such as catalase and GPx in cardiac tissue, ultimately offering cardiac protection (Elasoru et al., 2021). Another study reported that AND treatment shielded against post-myocardial infarction (adverse cardiac remodeling) by improving echocardiographic indices such as left ventricular ejection fraction (LVEF) and left ventricular end-systolic diameter (LVESD); lowering the levels of related profibrotic proteins such as p-smad-3 and TGF-β; and decreasing the expression of connective tissue growth factor (CTGF), collagen I, and collagen III. In addition, AND treatment reduced the positive inflammatory cardiomyocyte numbers such as cluster of differentiation (CD45) and cluster of differentiation (CD68), inhibited p-P65 and p-IκB, and reduced the expression of inflammatory markers including IL-6, IL-1β, TNF-α, and MCP-1, showing myocardial protection against post-myocardial inflammation. Also, AND increased the levels of SOD2, GPx, and quinone oxidoreductase 1 (NQO1) while decreasing the levels of NOX4, P67 phox, and GP91 transcription in the heart. Moreover, AND enhanced the Nrf2/HO-1 route in mice and ultimately suppressed oxidative stress induced by postmyocardial infarction (Xie et al., 2020).

Effect of AP on cardiac hypertrophy

Studies reported that AND or AP extract decreased evidence of hypertrophy by restoring normal ratios of heart weight to tibial length (HW/TL), heart weight to body weight (HW/BW), and lung weight to body weight (LW/BW) and reducing the expressions of markers of cardiac hypertrophy, including ANP, BNP, and β-myosin heavy chain (MHC-β) (Hsieh et al., 2016; Xie et al., 2020). A study reported that AP extracts reduced the expression of markers of inflammation such as COX-2, p-IκBα, and NFκB in HFD-fed mice and reduced collagen formation in cardiac tissues. Furthermore, AP extract exerted anti-apoptotic effects, which were characterized by a reduction in the protein levels of Fas-ligand, FADD, Fas, caspase-3, and caspase-8 in the heart tissue of HFD-fed mice. Moreover, AP extract reduced the expression of pro-apoptotic proteins Bak and BCL-2-associated agonist of cell death (Bad) and decreased cytochrome c (Cyt C) in HFD-fed mouse cardiac tissues (Hsieh et al., 2016). In another report, AND reduced hypertrophy in a transverse aortic constriction (TAC) mouse model: improved echocardiographic indices, decreased BNP and Ang II levels, reduced cardiac fibrosis, and decreased cardiac cell apoptosis (Tian et al., 2023).

Antioxidative effect of AP

In this review, AP significantly improved antioxidant enzyme activities in the heart, including CAT, SOD, and GPx, and offered myocardial protection against oxidative damage (Al Batran and Al-Bayaty, 2014; Liang et al., 2018). AP also reduced lipid peroxidation, a process that is associated with oxidative damage, by reducing its end products, including MDA and 4-HNE, in the heart (Liang et al., 2018). In addition, AP enhanced Nfr2 expression, leading to the upregulation of proteins such as HO-1, NQO1, GPx, and SOD, and ultimately suppressing oxidative stress induced by myocardial infarction (Xie et al., 2020).

Anti-inflammatory effect of AP

In this review, cardiovascular complications brought on by HFD, diabetes, and other agents led to an increase in inflammatory markers like NF-κB, IκB-α, COX-2, VCAM-1, ICAM, and pro-inflammatory cytokines like IL-6, IL-1 β, and TNF-α in cardiac tissue and plasma. Meanwhile, the expression of IL-10 (an

anti-inflammatory cytokine) was reduced. However, AP treatment increased the expression of IL-10 and reduced the expression of all other inflammatory markers, thereby demonstrating an anti-inflammatory effect (Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014; Chen et al., 2020; Liang et al., 2018).

In response to inflammatory and danger signals induced by diabetes in rats, serum IFN-γ and IL-2 expressions were elevated, whereas the expression of IL-10 and TGF-β was reduced. However, AP treatment improved the expression of IL-10 and suppressed the expression of IFN-γ and IL-2 (Zhang et al., 2013). AP reduced blood CRP levels in a coronary heart disease mouse model, demonstrating a cardiac protective effect (Shu et al., 2020). CD36 was increased in atherogenic rabbits, and AP administration reversed it, demonstrating a cardio-protective effect against atherosclerosis (Al Batran et al., 2014). The expressions of CD45 and CD68 increased in response to an inflammatory signal; however, AP revealed a tendency to decrease these proteins in an inflamed cardiomyocyte, thereby reducing myocardial inflammation (Xie et al., 2020).

Due to T1D and inflammation, the expressions of GATA3 were reduced, whereas T-bet and RORγt expressions in rats were increased in the pancreatic lymphatic node. However, AP treatment reversed these trends, demonstrating an anti-inflammatory effect (Zhang et al., 2013).

Agents of cardiovascular diseases in animal models

Most of the reviewed manuscripts used substances to induce cardiovascular complications in this study. Some of these substances are streptozotocin (Akhtar et al., 2016; Liang et al., 2018), lipopolysaccharide (Zhang et al., 2015), high-fat diets (Akhtar et al., 2016; Chen et al., 2020; Ding et al., 2014; Hsieh et al., 2016; Lin et al., 2020; Wang et al., 2021), streptozotocin (Akhtar et al., 2016; Liang et al., 2018), and isoproterenol (Elasoru et al., 2021). Out of the 16 manuscripts reviewed, seven reported the use of HFD to induce cardiovascular complications. Most of these studies recorded that HFD induced obesity, diabetes, and dyslipidemia in the experimental animals, which are risk factors for cardiovascular diseases (Zhu, 2017). In addition, they revealed the progression of cardiovascular disease using various indices for cardiac function.

Anthracyclines, including doxorubicin (DOX), which is used as a chemotherapy treatment, cause heart damage. A study reviewed showed the protective effects of AP extract on DOX-induced cardiomyopathy. Briefly, AP extract improved antioxidant enzyme activities such as catalase and SOD and also increased glutathione levels. In addition, creatinine kinase and lactate dehydrogenase activities were reduced by AP extract, thereby decreasing cellular damage (Wang et al., 2022).

Interestingly, AP and its bioactive compounds showed cardioprotective effects against cardiovascular complications induced by streptozotocin, *Porphyromonas gingivalis*, lipopolysaccharide, and isoproterenol (Akhtar et al., 2016; Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014; Elasoru et al., 2021; Liang et al., 2018; Zhang et al., 2015).

Toxicity

None of the studies reviewed reported any toxicity to AP or its bioactive compounds, including AND and deAND. One of the studies performed a short-term toxicity test (two weeks) of AP water extract on Sprague-Dawley rats. The doses administered were 5, 50, and 300 kg/BW. At the end of the study period, no deaths, toxicity, or pathological abnormalities in organs or blood chemistry were observed (Akhtar et al., 2016).

DISCUSSION

Hyperglycemia, in the case of diabetes, is caused either by insulin deficiency or insulin resistance (Akhtar et al., 2016). Insulin deficiency may occur as a result of the destruction of insulin-producing β-cells in the pancreas, which has been linked to several causes, including autoimmune conditions (Yoon and Jun, 2005). Insulin deficiency leads to T1D (DiMeglio et al., 2018). Insulin resistance, in contrast, is characterized by the body's inability to effectively make use of insulin. In the initial stage, there is an increase in blood levels of insulin (hyperinsulinemia). Over time, pancreatic β-cells become unable to produce sufficient insulin to meet the body's needs, resulting in hyperglycemia and T2D (Ahmad et al., 2022). Diabetes, particularly T2D, causes diabetic cardiomyopathy (Boudina and Abel, 2010); therefore, the need to study diabetes or its related conditions as they affect the heart is key to finding a treatment.

When blood glucose levels rise, insulin is released from the pancreatic β-cells. Insulin, through a series of events, stimulates the translocation of GLUT4 to the cell membrane, which transports glucose into the cell, thereby maintaining glucose homeostasis (Bryant et al., 2002; Wang et al., 2020). The series of events that take place in response to glucose level rise are as follows: binding of insulin to its receptor (a tetramer having two alpha-subunits and two beta-subunits linked by disulfide bonds) on the cell membrane; autophosphorylation by the beta subunits of insulin receptor containing tyrosine kinase domain; recruit-

ing and phosphorylating IRS by the phosphorylated beta-subunits; binding and activation of phosphatidylinositol 3-kinase (P13K) by the phosphorylated IRS; recruiting of PI3K to the plasma membrane to convert PIP2 to PIP3 (phosphatidylinositol (3,4,5)-triphosphate) and activate PIP3 dependent protein kinase; phosphorylation and activation of AKT by PIP3-dependent protein kinase; and vesicle fusion which results from AKT activation that leads to GLUT4 containing vesicle translocation to the plasma membrane from intracellular compartments (Wang et al., 2020). From the molecular events above, it is evident that IRS and AKT are significantly involved in the translocation of GLUT4 to the plasma cell membrane. AP and deAND positively affected the insulin signaling pathway in epididymal adipose tissue of mice fed with HFD by increasing the phosphorylation of IRS1, AKT, and AS160 (an AKT substrate of 160 kDa), which enhanced the insulin stimulation of GLUT4 membrane translocation in the epididymal adipose tissue that was initially impeded by the HFD (Chen et al., 2020).

Th1, Th2, and Th17 cells are regulated by Tbet, GATA3, and RORγt transcription factors, respectively (Zhu, 2017). Th1 cells secrete cytokines such as TNF-α, IL-1β, IFN-γ, and IL-2. Th2 and Th17 cytokines include IL-10 and IL-17, respectively (Lee et al., 2019; Shaw et al., 2018). Any change in the levels of these master transcription factors (Tbet, GATA3, and RORγt) could impact how Th1, Th2, and Th17 cells work and grow (Zhu, 2017). AP ameliorated pancreatic lymphatic node inflammation by mediating Th1/Th2/Th17 balance and reduced systemic inflammation in an autoimmune NOD mouse model (Zhang et al., 2013). Zhang et al. (2013) concluded that the "*AP protective effect against TID might be attributed to Th1/Th2/Th17 balance, which could prevent beta cell death and T cell infiltration into the pancreatic islets.*" In a diabetic cardiomyopathy mouse model, cardiac inflammation, oxidative stress, apoptosis, and fibrosis were alleviated by AP (Liang et al., 2018).

As demonstrated in Fig. 4, this review proposes, based on findings, that one of AP's protective mechanisms in diabetic animal models is by alleviating inflammation in the pancreatic lymphatic nodes, which enhances insulin production in the pancreas. Insulins produced in response to elevated blood sugar levels stimulate GLUT4 membrane translocation to the cell membrane in adipose tissues, allowing glucose uptake into fat cells and normalizing blood glucose levels. The alleviation of hyperglycemia inhibits diabetes and other conditions that lead to diabetic cardiomyopathy, such as myocardial inflammation, oxidative stress, and apoptosis.

Although lipids play very significant roles within the body system, including storage roles (fatty acids, triglycerides, and sterols) and structural roles (glycolipids, ceramides, and phospholipids), when at abnormal levels (dyslipidemia), they increase the risk of CVDs, particularly coronary heart diseases (Bhargava et al., 2022; Gervois et al., 2000; Ni et al., 2015). Lipoprotein (a), low-density lipoproteins (LDL), very low-density lipoproteins (VLDL), intermediate-density lipoproteins (IDLs), triglycerides, cholesterols, trans fatty acids, lysophosphatidic acid, and phosphatidylcholine are some of the lipids that can make CVDs more likely (Bhargava et al., 2022). Dyslipidemia is primarily evaluated by conducting a fasting lipid profile test, which primarily measures LDL-cholesterol (LDL-C), HDL-cholesterol (HDL-C), triglycerides (TG), and total cholesterol (TC) (Ni et al., 2015). This review found that AP halted the progress of the molecular events that may lead to dyslipidemia.

PPARα is a transcription factor primarily expressed in tissues with high levels of fatty acid catabolism, including the heart, muscle, and liver. It plays a significant role in regulating lipid metabolism in cells and tissues and controls the activities of numerous genes that are important in lipid metabolism (Gervois et al., 2000; Yoon, 2009). AP increased the expression of PPARα in a high-fat-induced CHD mouse model, demonstrating a beneficial effect on lipid metabolism (Shu et al., 2020). SREBPs play a significant role in lipid homeostasis by controlling the expression of important downstream target genes that are involved in maintaining healthy levels of lipids in the body. Shreds of evidence suggest that fatty acid metabolism is regulated by SREBP 1, whereas SREBP 2 primarily regulates cholesterol metabolism (Horton et al., 2002, 2003).

AP treatment decreased the expressions of SREBP-1 and SREBP-2, along with their target genes. In addition, AP increased the expression of uncoupling protein 2 (involved in regulating lipid metabolism) and decreased HMGCR, SCD-1, and FAS expressions (involved in lipid synthesis) in a C57BL/6 mouse model of HFD-induced obesity (Ding et al., 2014).

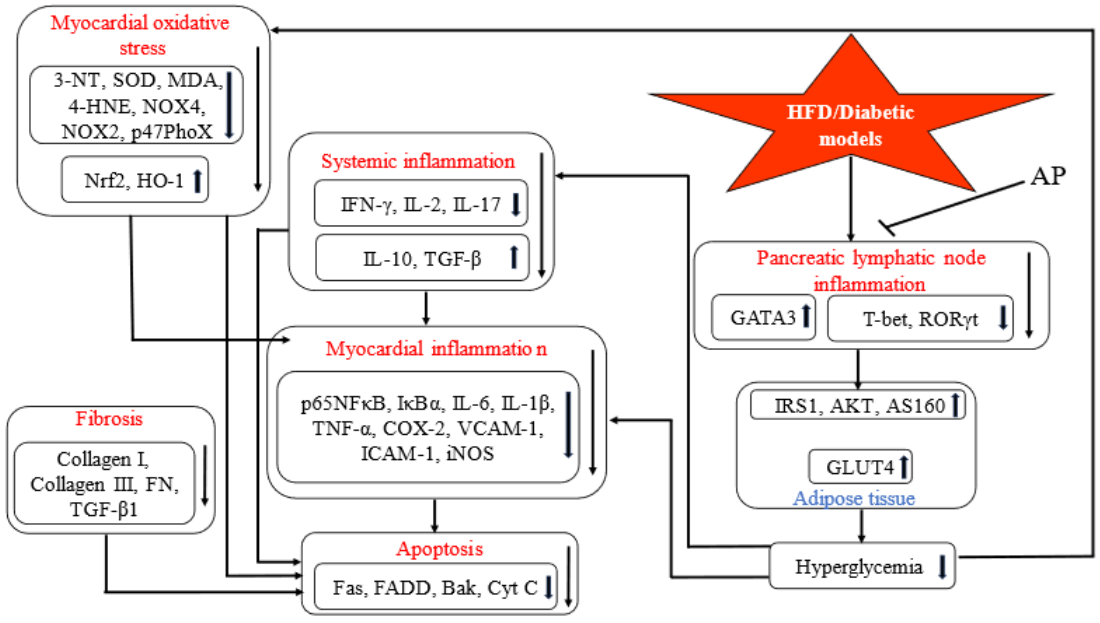

Figure 4. Proposed mechanism of action of *Andrographis paniculata* (AP) on hyperglycemia.

Andrographis paniculata stops inflammation in pancreatic lymphatic nodes by increasing GATA 3 and decreasing T-bet and RORγt. This results in the upregulation of IRS1, AKT, and AS160, which enhances insulin stimulation of GLUT4 membrane translocation in the adipose tissue. The stimulation of GLUT4 membrane translocation alleviates hyperglycemia. Systemic inflammation, myocardial inflammation, and myocardial oxidative stress induced by hyperglycemia are inhibited. Apoptosis and fibrosis that result from inflammation and oxidative stress are also inhibited.

This review proposes, based on findings as demonstrated in Fig. 3, that AP restores a normal lipid profile (Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014; Shu et al., 2020) by regulating genes involved in lipid synthesis and fatty acid oxidation. This positive effect of AP alleviates oxidative stress, inflammation, and endothelial dysfunction and suppresses coronary artery diseases and arteriosclerosis (Al Batran and Al-Bayaty, 2014; Shu et al., 2020). In addition, it might also inhibit myocardial infarction, as arteriosclerosis is the main cause of myocardial infarction (Palasubramaniam et al., 2019; Samaniego and Moguel-Ancheita, 2021). Since arteriosclerosis is impeded, myocardial infarction would also be inhibited.

CAD is a condition in which atherosclerotic plaques block the coronary arteries, preventing blood flow to the myocardium. Obesity, smoking, genetics, gender, age, and abnormal lipid levels are just a few of the causes of CAD (Assmann et al., 1999; Hajar, 2017). Aortic valve calcification (AVC), on the other hand, is closely related to atherosclerosis and also serves as an indication of coronary artery disease (Hajar, 2017). A study reported that AP suppressed AVC by reducing the expression of ALP and MGLL, which play key roles in the progression of AVC (Wang et al., 2021). However, the protective mechanism of AP in this *in vivo* study was not fully elucidated; hence, more studies are needed in this area. The MCP-1 plays a role in the development of atherosclerosis. MCP-1 recruits monocytes and macrophages to the arterial walls, contributing to atherosclerosis formation (Ikeda et al., 2002). In two different models of coronary disease—a mouse model and a rabbit model—AP lowered MCP-1 (Al Batran et al., 2014; Shu et al., 2020). Some vasodilators, such as NO, and vasoconstrictors, like ET-1 and TXA2, are released by the endothelium. These help to control the tone of the blood vessels. Vasodilators relax the vessels. Vasodilator production is primarily what causes endothelial dysfunction, which can result in atherosclerosis and other cardiovascular diseases (Godo and Shimokawa, 2017; Matsuzawa and Lerman, 2014). Oxidative stress, through several mechanisms, is one of the primary influencers of endothelial dysfunction. Briefly, in one of the mechanisms, superoxide anions react with NO to make peroxynitrite (ONOO-), which lowers the amount of NO that is available. Additionally, the activity of endothelial NO synthase (eNOS) (an enzyme that catalyzes the production of NO) can be inhibited by ONOO-, causing a drastic decline in NO concentration (Matsuzawa and Lerman, 2014). In addition, a body of evidence also suggests that inflammation may trigger endothelial dysfunction (Galle et al., 2003; Theofilis et al., 2021). In a coronary heart disease mouse model, AP ameliorated endothelial dysfunction by reducing blood levels of TAX2 and ET-1 and elevating PGI_2 and NO levels. In addition, inflammation was also reduced (Shu et al., 2020). Fur-

thermore, in a rabbit model of *Porphyromonas gingivalis*-induced atherosclerosis, AP alleviated systemic inflammation and oxidative stress. Similarly, inflammation in the aortic homogenate was also reduced (Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014).

As demonstrated in Fig. 5, based on findings, this review proposes that AP's protective effect against endothelial dysfunction (which is implicated in coronary artery diseases) is by suppressing oxidative stress and inflammation. This effect protects other organs, such as the kidney and liver, from damage induced by coronary artery disease (Al Batran and Al-Bayaty, 2014; Al Batran et al., 2014).

Myocardial infarction is a heart attack characterized by a decrease in blood flow to a particular portion of the myocardium, which leads to the death of the cells in that portion as well as in other areas. Myocardial infarction proceeds with cardiac ischemia (Thygesen et al., 2007). Risk factors for myocardial infarction include, but are not limited to, an abnormal lipid profile, obesity, diabetes, and smoking (Yusuf et al., 2004). Immediately after myocardial infarction, the heart undergoes significant changes, known as cardiac remodeling, which involves the building up of fibrous tissue around the damaged heart muscle. This leads to structural distortion of the heart and heart stiffness and thereafter progresses to inevitable heart failure if no timely solution is proffered (Frantz et al., 2022; Garza, 2015). In myocardial-infarcted animal models, AP reduced cardiac injury systemic biomarkers, pro-inflammatory cytokines and increased antioxidant activities (Elasoru et al., 2021; Xie et al., 2020). In a post-myocardial infarcted animal model, AP reduced inflammation, oxidative stress, and fibrosis in heart tissues (Xie et al., 2020). The underlying mechanism of action of AP on the myocardial infarcted heart is via an anti-inflammation and antioxidant mechanism that suppresses post-myocardial infarction/adverse cardiac remodeling and reduces the formation of excessive fibrotic tissues (Xie et al., 2020).

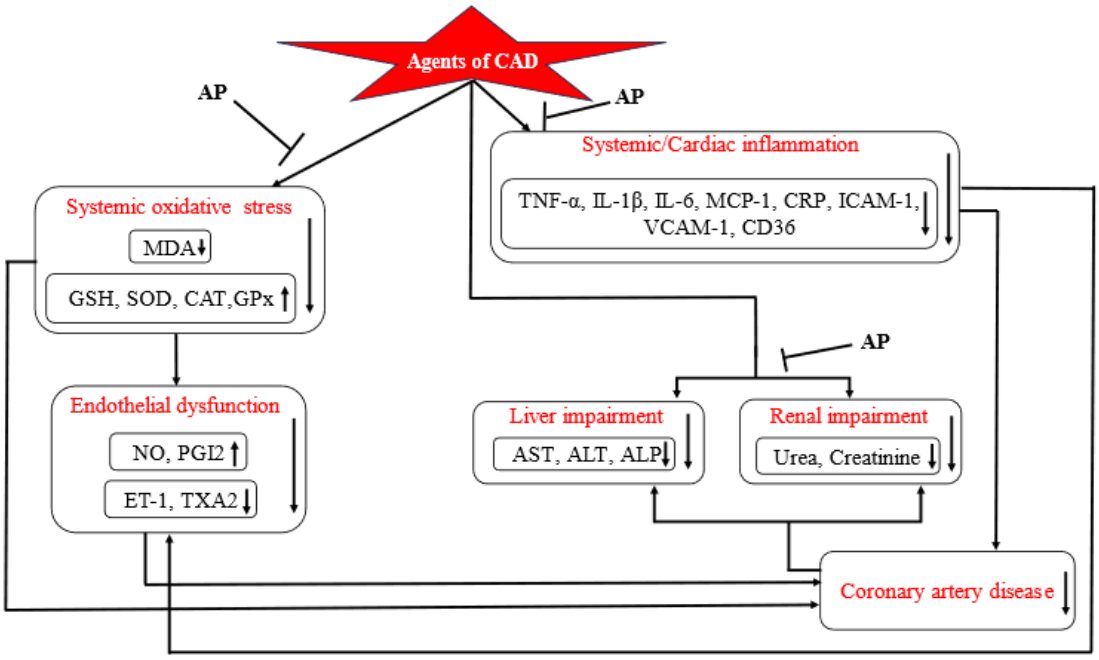

Figure 5. Proposed mechanism of action of *Andrographis paniculata* (AP) on coronary artery disease.

AP protects against coronary artery disease by suppressing endothelial dysfunction and reducing oxidative stress and inflammation, which potentially protects other organs, such as the kidneys and liver, from damage.

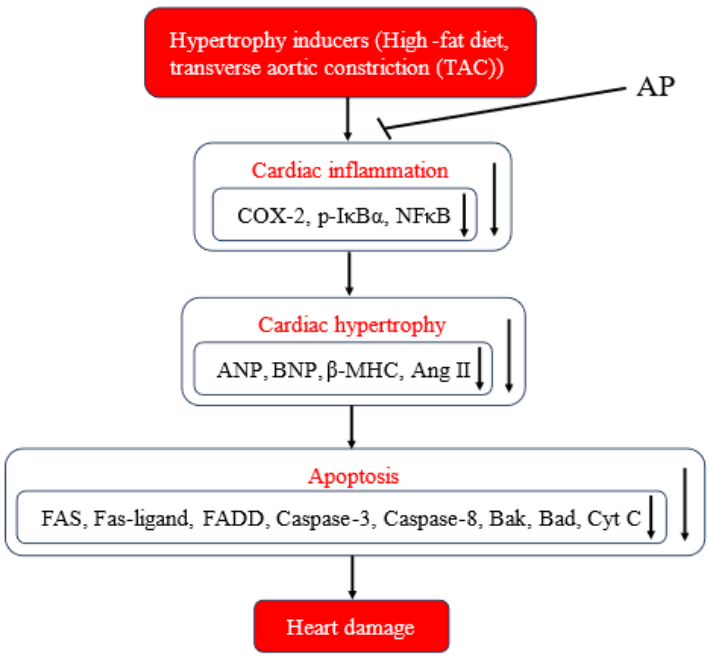

Figure 6. Proposed mechanism of action of *Andrographis paniculata* (AP) on cardiac hypertrophy.

AP inhibits markers of cardiac inflammation such as COX-2, p-IκBα, and NFκB. The inhibition of cardiac inflammation results in the inhibition of cardiac hypertrophy markers, including ANP, BNP, β-MHC, Ang II, and apoptotic markers, including Fas, Fas-ligand, FADD, caspase-3, caspase 8, Bak, Bad, and Cyt C, thereby halting cardiac hypertrophy-induced heart damage.

Myocardial hypertrophy is a condition that occurs as a result of an increased workload in the myocardium, which increases the mass of ventricular heart muscles. Importantly, pressure/volume overload or myocardial infarction triggers the activation of pro-hypertrophic molecular pathways, which most often lead to heart failure (Heinzel et al., 2015). In the articles reviewed, HFD-fed animal models and transverse aortic constriction models were used for cardiac hypertrophy studies. Cardiac inflammatory markers were elevated, which led to an increase in cardiac hypertrophy markers including ANP, BNP, β-MHC, and Ang II, and ultimately resulted in apoptosis. Interestingly, AP reversed this trend with its anti-inflammatory effect, as demonstrated in Fig. 6 (Hsieh et al., 2016; Tian et al., 2023).

Reactive oxygen species (ROS) are a group of molecules that are generated as byproducts of cellular metabolism. Common ROS include superoxide radicals (O_2•−), hydrogen peroxide (H_2O_2), hydroxyl radicals (•OH), and singlet oxygen ($^1O_2$) (Pizzino et al., 2017). ROS are vital for a variety of cellular processes, including the activation of transcription factors, immunity, phosphorylation, differentiation, and apoptosis. However, when ROS are higher than normal, they become dangerous to the cell, leading to a condition known as oxidative stress (Pizzino et al., 2017; Rajendran et al., 2014). Therefore, it is important to balance ROS production and ROS clearance. Interestingly, the cell has an antioxidant mechanism for preventing oxidative stress and keeping ROS at a normal level (Pizzino et al., 2017; Rajendran et al., 2014). Inflammation, in contrast, is a natural response by the body's system to a variety of factors, including harmful stimuli, pathogens, and damaged cells (Zhao et al., 2021). Heart diseases like coronary artery disease, myocardial infarction, cardiac hypertrophy, and diabetic cardiomyopathy are thought to be caused by inflammation and oxidative stress (Al Batran et al., 2014; Hsieh et al., 2016; Liang et al., 2018; Shu et al., 2020; Xie et al., 2020). Figs. 7 and 8 show the protective molecular actions of AP against oxidative stress and inflammation, respectively.

Tang et al. (2021) constructed a cardiovascular disease network (progCDN) to map the pathways along which cardiovascular diseases progress. In the study, acute heart failure, chronic heart failure, cardiomyopathy, aortic valve disorder, and some other cardiovascular diseases were grouped under diseases that could lead to further complications. Coronary arteriosclerosis was grouped under diseases that could result from many diverse pathways (Tang et al., 2021). As one cardiovascular disease condition could lead to another, understanding the protective mechanism of action of AP becomes imperative to establishing it as a cardiovascular disease supplementary therapy.

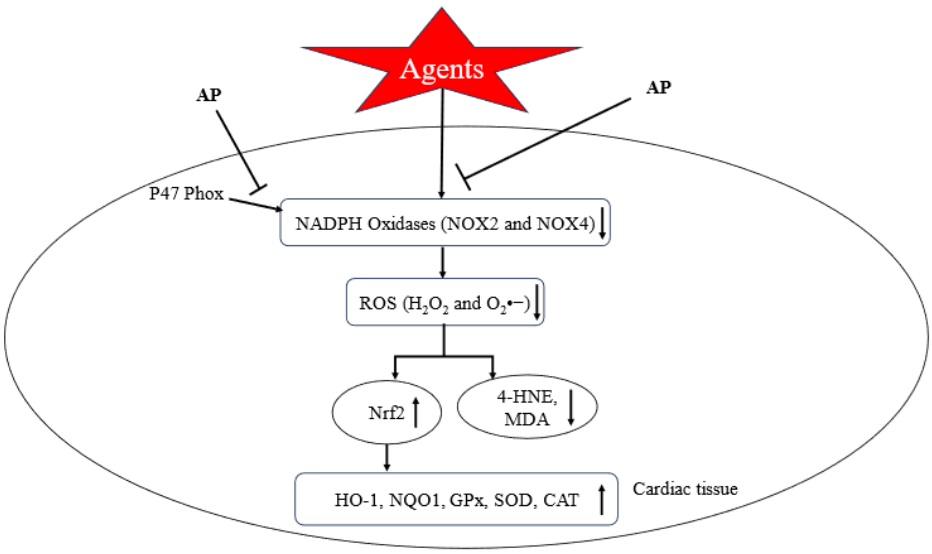

Figure 7. Proposed mechanism of action of *Andrographis paniculata* (AP) on oxidative stress in cardiac tissue.

AP inhibits the upregulation of NADPH oxidases, including NOX2 and NOX4. AP inhibits NOX2 by inhibiting its primary regulator, P47phox. NADPH oxidase inhibition results in a decrease in ROS, which leads to a reduction in the levels of lipid peroxidation markers such as 4-HNE and MDA. Nrf2 is upregulated and enhances the upregulation of HO-1 and antioxidants such as GPx, SOD, CAT, and NQO1. These molecular actions are proof that AP is an antioxidant.

Several studies, including *in vitro* studies, have reported the antioxidant and anti-inflammatory properties of AP. Sya'ban et al. (2023) examined the antioxidant and anti-inflammatory effects of AP methanolic extract and some of its bioactive compounds using lipopolysaccharide (LPS)-induced RAW 264.7 cells. The extract and bioactive compounds inhibited NO production in the cell, demonstrating their antioxidant and anti-inflammatory properties (Sya'ban et al., 2023). According to Sheeja et al. (2006), AP extract reduced inflammation and oxidative stress in the lab by blocking NO, lipid peroxidation, superoxide, and hydroxyl radicals. Another study (Feng et al., 2017) showed that andrographolide could reduce the levels of inflammatory cytokines IL-6 and TNF-α in rat myocardium microvascular endothelial cells. This made the cells less inflamed. In H9C2 cardiomyocytes induced with high glucose concentration, andrographolide reduced the expressions of p-IκBα and p-NFκB (inflammatory proteins), decreased ROS generation, downregulated NOX2, NOX4, $p47^{phox}$ (involved in ROS generation), promoted Nrf2 nuclear translocation, reduced the levels of ANP and BNP (cardiac hypertrophy markers), and reduced Bax/Bcl-2 ratio and caspase-3 activity (Liang et al., 2018).

AP reduces ROS accumulation by downregulating NOX2, NOX4, and $p47^{phox}$ and promoting the nuclear translocation of Nrf2, thereby preventing oxidative stress in cardiac cells that could result in several cardiovascular complications (Fig. 7). Furthermore, AP inhibits the nuclear translocation of a regulatory inflammatory protein, NFκB, thus regulating the expression of its downstream targets and thereby alleviating cardiac inflammation (Fig. 8). This review aligns with the findings of *in vitro* studies (Feng et al., 2017; Liang et al., 2018) and much other research, indicating that AP possesses potent antioxidant and anti-inflammatory properties, suggesting its potential as an effective therapy for cardiovascular diseases. Interestingly, AP also has a direct effect on genes that are involved in lipid synthesis and fatty acid oxidation, which normalizes the lipid profile and inhibits oxidative stress, inflammation, apoptosis, fibrosis, endothelial dysfunction, coronary artery disease, atherosclerosis, myocardial infarction, hypertrophy, and post-myocardial infarction (Al Batran et al., 2014; Ding et al., 2014; Hsieh et al., 2016; Liang et al., 2018; Lin et al., 2020; Shu et al., 2020; Tian et al., 2023; Xie et al., 2020; Zhang et al., 2013; 2015).

Limitations of the study and future perspectives

Given the lack of robust data on the effect and molecular actions of AP on AVC, further *in vivo* animal studies are needed to elucidate its effect through investigation of its molecular mechanism. It is worthwhile to note that some of the papers reviewed did not give a clear explanation of the method of preparation of plant extracts or extraction of bioactive compounds. A significant number of the research papers failed to report the total yield of the preparation. The

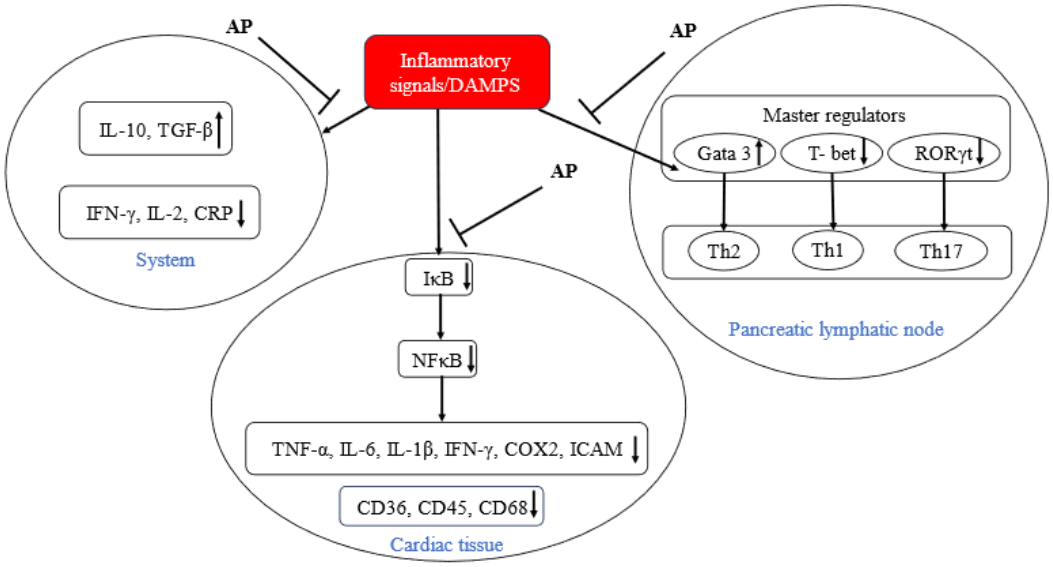

Figure 8. Proposed mechanism of action of *Andrographis paniculata* (AP) on inflammation.

AP enhances the upregulation of IL-10, TGF-β (systemic), and the downregulation of systemic IFN-γ, IL-2, and CRP. AP enhances the increase in the expression of Gata 3 (a Th2 cytokine regulator) and the decrease in the expression of T-bet and RORγt (regulators of Th1 and Th17, respectively) in the pancreatic lymphatic node. AP inhibits the phosphorylation of IκB in the heart tissue, which results in the downregulation of NFκB and other pro-inflammatory cytokines, including TNF-α, IL-6, IL-1β, IFN-γ, COX2, and ICAM. AP also inhibits CD36, CD45, and CD68 in cardiac tissues. These molecular actions confirm AP's anti-inflammatory activities.

reports of phytochemical analysis of plant extracts were lacking in some of the manuscripts. These limitations make the reproducibility of results difficult. Furthermore, most studies did not perform a proper toxicity test; hence, it is difficult to conclude the toxicity of AP from only a few studies.

While the review process involved the independent assessment of information by multiple review authors, it is important to acknowledge that a specific tool was not utilized for information synthesis, leaving a small margin for potential errors.

This review found that recently, andrographolide dominated the research landscape, with 12 out of 16 reviewed studies employing it (Table 2). This likely stems from the common conception that andrographolide is the major bioactive compound responsible for AP's pharmacological effects (Jayakumar et al., 2013). However, while andrographolide possesses established pharmacological activity, certain studies have demonstrated the superior efficacy of whole AP extracts (Chen et al., 2020; Sya'ban et al., 2023). This suggests that other phenolic components and compounds within AP, beyond just andrographolide and its derivatives, contribute synergistically to its therapeutic potential (Sya'ban et al., 2023). A compelling example is a study by Chen et al. (2020), where AP extract demonstrated significantly greater improvements in insulin resistance compared to pure andrographolide. These findings suggest that standardizing AP extracts, rather than focusing solely on andrographolide, may become a more promising approach for developing therapeutic agents, particularly for cardiovascular diseases.

These reviewed studies have shown favorable effects against cardiovascular diseases; however, clinical studies have to be done before the drug can be used effectively and safely in humans.

CONCLUSION

This systematic review found substantial evidence to support the use of *A. paniculata* as a supplementary therapy for cardiovascular disease. It should, however, be noted that some studies reported that *A. paniculata* effects on cardiovascular diseases were dose-dependent; hence, more studies are needed to ascertain the effective dose for the treatment of any named cardiovascular disease. In addition, low bioavailability is associated with most herbal medications, including *A. paniculata;* hence, the use of delivery vehicles can be an effective tool to improve bioavailability in future studies. Furthermore, most of the studies that have been conducted on *A. paniculata* effects against cardiovascular diseases are in animal experimentation and *in vitro* studies, so the efficacy and safety of *A. paniculata* are still questionable; further study is needed to explore the pharmacology effect of *A. paniculata* in humans. In addition, the safety profiles of *A. panic-*

ulata should also be confirmed in acute toxicity as well as chronic toxicity tests.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

ACKNOWLEDGMENTS

The authors thank Universitas Indonesia for the grant "Hibah Puti 2023", with the grant number NKB-694/UN2.RST/HKP.05.00/2023.

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AUTHOR CONTRIBUTION:

Contribution	Eziefule OM	Arozal W	Wanandi SI	Dewi S	Nafrialdi	Saraswati M	Louisa M
Concepts or ideas	x	x	x				
Design	x			x			x
Definition of intellectual content	x	x	x				
Literature search	x	x	x	x	x	x	x
Data acquisition	x	x					x
Data analysis	x	x					x
Manuscript preparation	x	x	x		x		x
Manuscript editing		x	x				
Manuscript review	x	x	x	x	x	x	x

Citation Format: Eziefule OM, Arozal W, Wanandi SI, Dewi S, Nafrialdi, Saraswati M, Louisa M (2024) *Andrographis paniculata*: A potential supplementary therapy for cardiovascular diseases - A systematic review of its effects and molecular actions. J Pharm Pharmacogn Res 12(3): 487–513. https://doi.org/10.56499/jppres23.1841_12.3.487

Supplementary data

Table S1. Operationalization of variables.

Variables	Conceptual definition	Operational definition	Indicators
Andrographis paniculata	Herbal plant from the *Acanthaceae* family	Potential cardiovascular disease supplementary therapy	*Andrographis paniculata* extracts, and bioactive compounds (andrographolide; 14-deoxy-11,12-didehydroandrographolide)
Cardiovascular diseases	Conditions that impact the heart negatively and reduce its functionality	Rodents and zebrafish models of cardiovascular diseases or conditions that could lead to cardiovascular diseases	Cardiac hypertrophy, aortic valve calcification, myocardial infarction, post-myocardial infarction atherosclerosis, cardiotoxicity, myocardial malfunctions, obesity, diabetes, dyslipidemia
Effects	Observable impacts of drug action	Therapeutic effects	Anti-inflammation, antioxidant, anti-apoptosis, anti-fibrosis, Anti-endothelial dysfunction, anti-cardiac hypertrophy, anti-dyslipidemia, anti-diabetes, anti-obesity, reduced ventricular stress
Molecular actions	Describes the interplay between the drug and its biological targets at the molecular level, leading to the observed effects (Swinney, 2011)	Protective molecular interaction	All molecular interactions that lead to desired effects (i.e. anti-inflammation, anti-oxidative stress, reduced ventricular stress, etc. (Table 1)

Table S2. Assessment of risk of bias in the animal studies (using SYRCLE's tool) (Hooijmans et al., 2014).

No.	Reference	Was the allocation sequence adequately generated and applied?	Were the groups similar at baseline or were they adjusted for confounders in the analysis?	Was the allocation adequately concealed?	Were the animals randomly housed during the experiment?	Were the caregivers and/or investigators blinded from knowledge of which intervention each animal received during the experiment?	Were animals selected at random for outcome assessment?	Was the outcome assessor blinded?	Were incomplete outcome data adequately addressed?	Are reports of the study free of selective outcome reporting?	Was the study apparently free of other problems that could result in high risk of bias?
1	(Xie et al., 2020)	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Unclear	Unclear	Yes	Yes
2	(Chen et al., 2020)	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Unclear	Unclear	Yes	Yes
3	(Hsieh et al., 2016)	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Unclear	Yes	Yes	Yes
4	(Tian et al., 2023)	Unclear	Yes	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Yes	Yes
5	(Lin et al., 2020)	No	Yes	Unclear	Unclear	Unclear	No	Unclear	Unclear	Yes	Yes
6	(Wang et al., 2021)	No	Yes	Unclear	No	Unclear	Unclear	Unclear	Unclear	Yes	Yes
7	(Liang et al., 2018)	No	Yes	Unclear	No	Unclear	Unclear	Unclear	Yes	Yes	Yes
8	(Akhtar et al., 2016)	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Unclear	Unclear	Yes	Yes
9	(Shu et al., 2020)	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Unclear	Unclear	Yes	Yes
10	(Ding et al., 2014)	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Unclear	Yes	Yes	Yes
11	(Zhang et al., 2015)	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Unclear	Unclear	Yes	Yes
12	(Zhang et al., 2013)	Unclear	Yes	Unclear	Yes	Unclear	Yes	Yes	Unclear	Yes	Yes
13	(Elasoru et al., 2021)	Unclear	Yes	Unclear	No	Unclear	Unclear	Unclear	Unclear	Yes	Yes
14	(Al Batran and Al-Bayaty, 2014)	Unclear	Yes	Unclear	No	Unclear	Unclear	Unclear	Unclear	Yes	Yes
15	(Al Batran et al., 2014)	Unclear	Yes	Unclear	No	Unclear	Unclear	Unclear	Unclear	Yes	Yes
16	(Wang et al., 2022)	Unclear	Unclear	Unclear	No	Unclear	No	Unclear	Unclear	Yes	Yes

Table S3. Conflict of interest and funding source.

No.	Reference/Country	Funding	Conflict of interest
1	(Xie et al., 2020) China	The National Natural Science Foundation of China and Xinjiang Science and Technology Supported with grants	None
2	(Chen et al., 2020) Taiwan	Dr. Chen received grants from Ministry of Science and Technology, China Medical University, and Asian University, Taiwan	None
3	(Hsieh et al., 2016) Taiwan	Partly sponsored by Taiwan Ministry of Health and Welfare Clinical Trial and Research Center of Excellence	None
4	(Tian et al., 2023) China	None	None
5	(Lin et al., 2020) Taiwan	Asia University and China Medical University funded this research	None
6	(Wang et al., 2021) China	National Key R&D Program of China, Hubei University of Chinese Medicine "Young Crops Program" Project funded this research	None
7	(Liang et al., 2018) China	Not stated	None
8	(Akhtar et al., 2016) Malaysia	Not stated	None
9	(Shu et al., 2020) China	None	None
10	(Ding et al., 2014) China	Not stated	Not stated
11	(Zhang et al., 2015) China	Not stated	None
12	(Zhang et al., 2013) China	Not stated	None
13	(Elasoru et al., 2021) Brazil	Received support from Coordenaçao de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) and Conselho Nacional de Desenvolvimento Científico e Tecnologico	None
14	(Al Batran and Al-Bayaty, 2014) Malaysia	Funds were received from Universiti Teknologi Mara	None
15	(Al Batran et al., 2014) Malaysia	Received funding from the Ministry of Higher Education (MOHE)	None
16	(Wang et al., 2022) China	Not stated	Not stated