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Review Article

Pharmacokinetics and Pharmacodynamics Provide Insights into the Possible Action Mechanism of Ginkgo Flavonoids in Cardiovascular Disease Treatment

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Abstract

The prevalence of cardiovascular disease continues to increase. *Ginkgo biloba* is used as a therapeutic agent for cardiovascular disease. Flavonoids are considered bioactive components and quality control indicators in *G. biloba* leaf extract. They are closely related to cardiovascular and cerebrovascular pharmacodynamics and display the ability to dilate blood vessels, regulate blood lipid levels, antagonize platelet-activating factors and protect against ischemic damage. This review will help better understand the cardioprotective effects of *G. biloba* flavonoids. *Ginkgo biloba* has a positive safety profile but has some limitations. High-quality clinical trials are needed to confirm the effectiveness of ginkgo extracts. Furthermore, most studies on *G. biloba* use have focused on its flavonoid aglycones, whereas the effects of other flavonoid glycosides remain largely unknown. Finally, the action mechanism of *G. biloba* flavonoids can be more scientifically revealed only by comprehensively considering the relationship among intestinal flora, host and *G. biloba* flavonoids.

Key words: Cardiovascular disease, flavonoid, *Ginkgo biloba*, pharmacodynamics and pharmacokinetics

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INTRODUCTION

Cardiovascular Disease (CVD) is a leading cause of death worldwide. This condition includes heart failure, atherosclerosis and hypertension[1]. The quality of life of patients with CVD is severely impaired and it imposes an immense financial burden on their families. Herbs, such as ginkgo (*Ginkgo biloba* L.), can be used to treat CVD[2].

In the 1960s, German scientists discovered for the first time that the ginkgo leaf contains medicinal ingredients that can be used to treat CVD. Since then, ginkgo leaf extract has been gaining attention worldwide. Several epidemiological studies have shown that dietary flavonoids and flavonoid-rich food products correlate with the decreased likelihood of CVD development[3]. Flavonoids, which are known to have biological activities, can dilate blood vessels[4], regulate blood lipid levels[5], control blood sugar levels[6] and inhibit apoptosis and vascular rupture[7-9].

As shown in Fig. 1, we analyze the pharmacodynamics and pharmacokinetics of *G. biloba* flavonoids, thereby providing valuable information on the cardioprotective effects of this plant and laying a foundation for its clinical application.

Medication history: Ginkgo is the oldest (existed for more than 200 million years) and most valuable medicinal plant and is often termed a "living fossil"[10,11]. This tall and strong plant species is native to China. The earliest publications on the medicinal use of *G. biloba* can be traced back to 1505 AD. *Ginkgo biloba* possesses various medicinal and therapeutic properties, which have been widely recognized in recent decades[12].

Ginkgo biloba extract was first used in 1965 by the German physician and pharmacist W. Schwabe. It is found to improve peripheral and central blood circulation and can be used to treat CVD[13-15]. In 1974, *G. biloba* extract 761 (EGb761) became the first commercially available *G. biloba* extract for human use and it is one of the best-selling herbal extracts in the world. Since then, *G. biloba* extracts have garnered increasing attention from researchers worldwide.

In the United States, more than $250 million is spent annually on *G. biloba*-containing products[16,17]. In Europe, patients were recommended a daily standardized ginkgo extract dose of 120-240 mg for 8 weeks to prevent cardiovascular disease[18]. Leaf extract of *G. biloba* is one of the most widely used herbal medicines in the US and Europe.

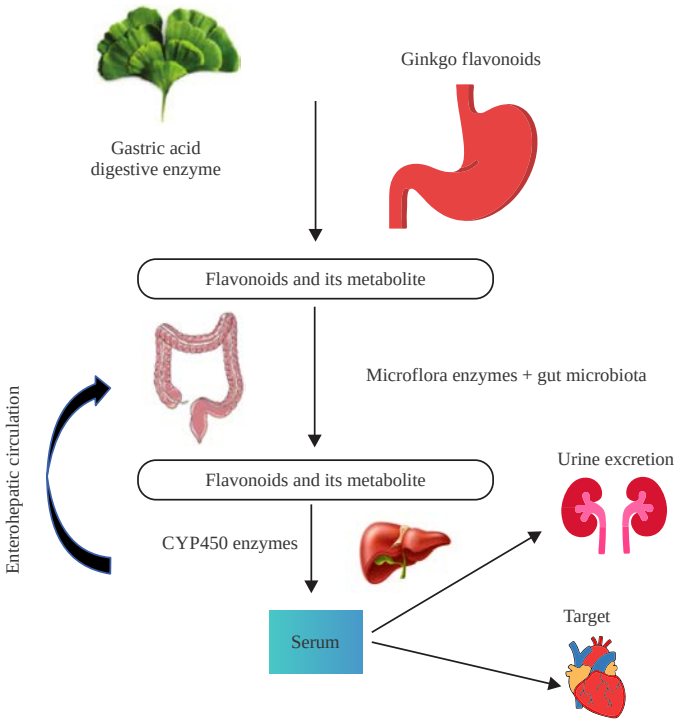


Fig. 1: Metabolism, pharmacokinetics and metabolomics of *Ginkgo biloba* flavonoids

Adapted from 'Pharmacokinetic, Metabolism and Metabolomic Strategies Provide Deep Insight Into the Underlying Mechanism of *Ginkgo biloba* Flavonoids in the Treatment of Cardiovascular Disease' by Tao *et al.*[68]. Retrieved from https://doi.org/10.3389/fnut.2022.857370. Copyright (2022) by Frontiers

Flavonoids are crucial compounds in *G. biloba* leaf extracts, with concentrations of up to 24%. Leaf extracts can be divided into two types: Whole-leaf and standardized. Whole-leaf extracts are typically obtained using alcohol and contain alcohol-soluble components, whereas standardized extracts are prepared via a multistep process[11]. Some compounds (terpene trione and flavonoids) are concentrated, whereas others (biflavones and ginkgolic acid) are removed. The standardized extracts include 6% terpene trione, 24% flavonoid glycosides and less than 5 ppm ginkgolic acid[19]. Finally, stable quality can be achieved by mixing batches of high- and low-concentration flavonol glycosides with terpene trione[20].

The medicinal value of *G. biloba* extract is gaining increasing recognition and researchers are attempting to standardize it by incorporating it into the pharmacopeia of several European countries. Meanwhile, the regulatory authorities in these countries have promoted the standardization of extracts into doses that are safe for human use as herbal medicines and have taken corresponding safety measures.

Chemical constituents: Phytochemicals are the main sources of new drugs and *G. biloba* leaves have been extensively studied as a source of beneficial medicinal components. A range of promising compounds, including flavonoids (quercetin, kaempferol and isorhamnetin) and terpenoids (bilobalide and ginkgolides), have been identified in this plant[21]. More than 110 compounds related to the aforementioned compounds have been isolated from the leaves of ginkgo plants[22]. Due to the presence of these compounds, *G. biloba* extracts are widely used to treat CVD[23].

Among the compounds found in *G. biloba*, trilactones, which are unique to *G. biloba*, have received the most attention, whereas flavonoids have received less attention. To date, over 70 flavonoids have been isolated from ginkgo leaves[24]. Most of these compounds can be categorized into four groups: Flavones, flavonols, biflavonoids and catechins[25]. Ginkgo flavones mainly include apigenin, luteolin and chrysin. Ginkgo flavonols contain quercetin, kaempferol and isorhamnetin. Biflavonoids are a class of compounds formed by the polymerization of two flavonoid cores through C-C bonds. A total of 13 biflavonoids, including bilobetin, amentoflavone, sequoiaflavone, sciadopitysin and podocarpusflavone A, have been identified in *G. biloba*. The catechins in *G. biloba* can be classified into four types: Catechins, epicatechin, gallic acid catechins and epigallic acid catechins and their corresponding dimers.

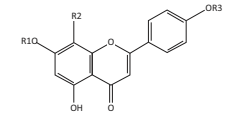
Most flavonoids are present in *G. biloba* leaves in the form of glycosides (such as isorhamnetin glycoside and quercetin glycoside) and several bioflavonoids[26]. The abundance of flavonol glycosides in the leaves is higher than that of other flavonoids. Isorhamnetin, quercetin and kaempferol are the major sources of flavonol glycosides and are linked via rhamnose and glucose in various forms. The mono-, di- and tri-glycanic moieties are at the C-3 position[27]. Other aglycones, such as syringetin and dihydroxy methyl flavone, have also been discovered. Based on the number of glycosyl units and species, flavonol glycosides can be classified into the following four groups: Mono-glucose, double glycosides, triple glycosides and acylated glycosides. These structures of *G. biloba* flavonoid ingredients are shown in Fig. 2.

Pharmacological activities of flavonoids: There are several clinical studies on *G. biloba* preparation in treating CVD and its effects are remarkable. A systematic evaluation study in patients with coronary heart disease found that the combination of *G. biloba* preparation and Western medicine could substantially improve the symptoms of angina pectoris and reduce adverse reactions[28]. Another study involving 1201 clinical patients systematically evaluated the ability of *G. biloba* dropping pills to improve blood rheology and lipid levels and found that the pills could considerably improve blood lipid levels and viscosity[29]. The cardioprotective mechanisms of *G. biloba* have been summarized in Table 1[30-42].

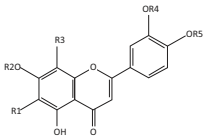
Luteolin and apigenin are the two main flavones found in *G. biloba* extracts[12]. Numerous epidemiological studies have also shown that luteolin can reduce the risk of CVD by reducing oxidative stress, protecting vascular endothelial cells, dilating coronary arteries and blood vessels, scavenging free radicals, stimulating blood lipid production, exerting antihypertensive and antioxidant effects and protecting against ischemic damage[43-47].

Owing to its interactions with the signal transducer and the activator of the transcription 3 factor, luteolin can reduce the severity of oxidized low-density lipoprotein-induced inflammation[48]. Luteolin stimulates NO production by reducing oxidative stress[49] and protects venous endothelial cells. It increases the expression of B-Cell Lymphoma 2 (Bcl-2) and decreases the pro-apoptotic activity of Bcl-2-associated X protein when combined with peroxidase II. Luteolin also ameliorates ischemia/reperfusion injury effects in patients[50].

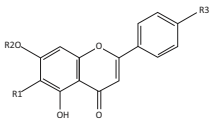
Flavone and its glycosides



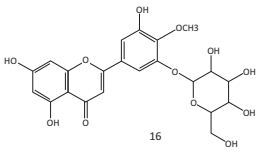
	R_1	R_2	R_3
1	H	H	H
2	H	H	CH_3
3	Glc	H	H
4	H	H	Glc
5	H	Glc	H



	R_1	R_2	R_3	R_4	R_5
6	H	H	H	H	H
7	H	H	H	H	Glc
8	H	Glc	H	H	H
9	H	H	H	Glc	H
10	H	H	Glc	H	H
11	Glc	H	H	H	H
12	H	Glc	H	Glc	H



	R_1	R_2	R_3
13	OH	H	H
14	H	CH_3	OH



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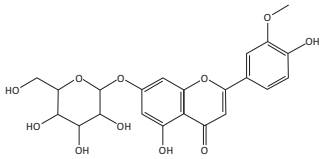

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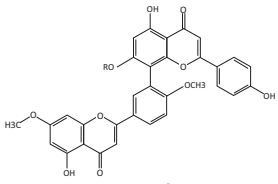
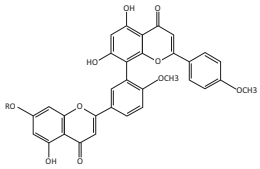
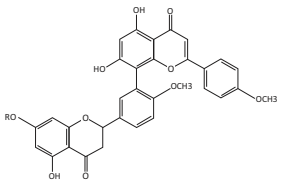
Biflavonoids



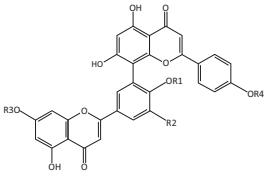
	R
52	H
53	Glc



	R
54	H
55	Glc

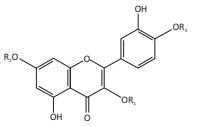


	R
56	H
57	CH_3

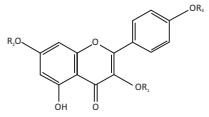


	R_1	R_2	R_3	R_4
58	H	OH	H	H
59	CH_3	H	H	H
60	H	H	CH_3	H
61	H	H	H	CH_3
62	H	OH	CH_3	H
63	CH_3	OCH_3	H	H
64	CH_3	H	CH_3	CH_3

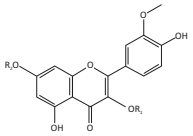
Flavonol and its glycosides



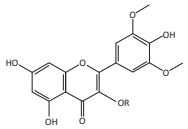
	R_1	R_2	R_3
17	H	H	H
18	Rha	H	H
19	Glc	H	H
20	Gal	H	H
21	Rha-{1→6}-Glc	H	H
22	Glc-{1→2}-Rha	H	H
23	4-O-Rha-Glc	H	H
24	Glc	H	Glc



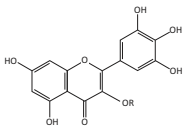
	R_1	R_2	R_3
25	H	H	H
26	Rha	H	H
27	Glc	H	H
28	H	Glc	H
29	Rha-{1→6}-Glc	H	H
30	Glc	Rha	H
31	Rha-{1→2}-Glc	H	H
32	Glc	Glc	H
33	H	Glc	Glc



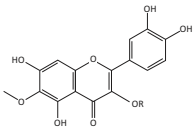
	R_1	R_2
34	H	H
35	Glc	H
36	H	Glc
37	Rha-{1→6}-Glc	H
38	Rha-{1→2}-Glc	H



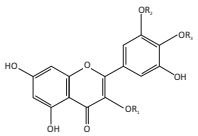
	R
39	H
40	Rha-{1→6}-Glc
41	2-O-Rha-Glc



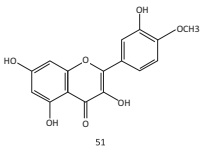
	R
42	H
43	Glc
44	Rha-{1→6}-Glc



	R
45	Rha-{1→6}-Glc
46	Rha-{1→2}-Glc



	R_1	R_2	R_3
47	H	CH_3	H
48	Rha-{1→6}-Glc	CH_3	H
49	Rha-{1→6}-Glc	H	CH_3
50	Rha-{1→6}-Glc	CH_3	CH_3



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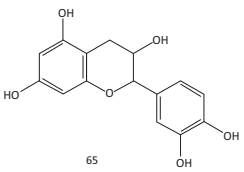
Catechins



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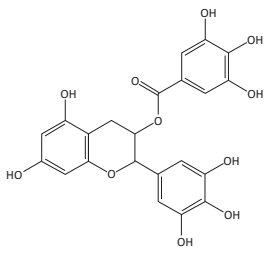

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Fig. 2: Chemical structures of flavonol and its glycosides

Table 1: *In vitro* and *in vivo* studies on the therapeutic effects of *Ginkgo biloba*

Field of study	Bioactive	Subjects	Dosage	Outcome	References
Antioxidant effect	EGb761	Young rat (6 months) Old rat (22-24 months)	100 mg/kg/day p.o. for 90 days	Alleviated ultrastructural-morphometric hypoxic damage Decreased SOD and MDA levels	Mozet *et al.*[30]
Treatment of cardiovascular diseases	GBE	Mice fed a high-fat diet	300 mg/kg/day p.o. for 28 days	Decreased final body weight and fat accumulation Decreased epididymal adipose tissue mass and adipocyte size Increased serum high-density lipoprotein cholesterol levels	Kang[31]
Antilipidemic effects	GBE	Human patients (n = 664)	Various doses	Increased triglyceride, total cholesterol, low-density lipoprotein cholesterol and high-density lipoprotein cholesterol levels	Fan *et al.*[32]
Antihypertensive effects	GBE	Rats	100 mg/kg/day p.o. for 28 days	Decreased systolic, diastolic and mean arterial blood pressures Decreased renal tissue MDA and nitrite levels TNF-α, IL-6 and IL-1β levels decreased Increased glutathione level Increased endothelial nitric oxide synthase activity Decreased protein expression of inducible NO synthase, TNF-α, IL-6 and IL-1β in kidney tissues EGb761 enhanced the effects of losartan with simvastatin	Abdel-Zaher *et al.*[33]
Anti-myocardial ischemia reperfusion	GBE50	Rats	50, 100 and 200 mg/kg/day p.o. for 15 days	Improved the levels of antioxidants and other related health benefits in ischemia reperfusion rats	Lu *et al.*[34]
Anti-myocardial ischemia reperfusion	EGb761	Rabbits	100 mg/kg A single dose, IV	Reduced the generation of free radicals and boosted the levels of antioxidants in the heart Decreased SOD and MDA levels	Xiao *et al.*[35]
Anti-myocardial ischemia reperfusion	EGb761	Diabetic and non-diabetic rats	25 and 50 mg/kg/day p.o. for 10 days	Reduced the amount of free radicals Improved the cardiac function of the rats after ischemia	Tosaki *et al.*[36]
Anti-cerebral ischemia reperfusion	EGb761	Rats	50 mg/kg/day i.p. for 14 days	Protected the rats from ischemia reperfusion injury	Erbil *et al.*[37]
Anti-cerebral ischemia reperfusion	GBE	Rats	50 mg/kg/day p.o. for 7 days	Reduced the expression of certain genes that are involved in stroke	Loh *et al.*[38]
Anti-cerebral ischemia reperfusion	EGb761	Rats	40 mg/kg/day p.o. for 7 days	Enhanced the antioxidant levels	Uríková *et al.*[39]
Anti-cerebral ischemia reperfusion	EGb761	Gerbils	25, 50 and 100 mg/kg/day p.o. for 7 days	Protected the neurons from experiencing neuronal death caused by glutamate and ischemia Inhibited the generation of free radicals	Chandrasekaran *et al.*[40]
Treatment of cardiovascular diseases	GBE	The study randomly assigned patients with CAD to receive either GBE (n = 42) or to a control group (n = 38)	p.o. for 14 days	Re-established the equilibrium between NO and endothelin-1	Wu *et al.*[41]
Treatment of cardiovascular diseases	EGb761	Patients with a high risk of cardiovascular disease (n = 8)	240 mg/day p.o. for 14 days	Upregulated radical-scavenging enzyme activities	Rodríguez *et al.*[42]

EGb761: Extract of *Ginkgo biloba* 761, p.o.: Per os (orally), SOD: Superoxide dismutase, MDA: Malondialdehyde, TNF-α: Tumor necrosis factor alpha, IL-6: Interleukin-6, IL-1β: Interleukin-1β, NO: Nitric oxide, IV: Intravenous, i.p.: Intraperitoneal, CAD: Coronary artery disease and GBE: *Ginkgo biloba* extract

Apigenin can prevent strokes[51]. In addition, using apigenin can improve heart function by reducing myocardial infarction size[52]. It also decreases the number of apoptotic cells in coronary blood flow[53]. Apigenin prevents the activation of proinflammatory chemicals in H9 myoblasts and protects the myoblasts from the effects of hypoxia and ischemia. However, in healthy individuals, consuming large amounts of this substance does not affect hemostatic variables or platelet aggregation[54].

Flavonols from *G. biloba* exhibit antibacterial, neuroprotective and antioxidant properties[9]. They can also improve blood circulation. These properties can be attributed to their ability to promote the growth and development of certain tissues and cells, such as vascular-type scaffolds[55]. In a previous study, *G. biloba* flavonol exhibited a concentration-dependent vasodilation effect. Rats with an enlarged aortic annulus reportedly showed strong contraction after being administered 5 μM norepinephrine but exhibited a significant decrease in contraction after administering *G. biloba* flavonoids[56]. Kaempferol improves the growth and development of vascular cells and binds to the vascular growth factor[57].

Only 5-10% of the total dietary flavonoid intake can be absorbed in the small intestine and the remaining flavonoids (90-95% of the total dietary flavonoid intake) will be enriched in the lower gastrointestinal lumen and reach the millimolar level. Flavonoid glycosides and metabolic conjugates excreted into the intestinal lumen through bile are affected by the intestinal microbial community[58].

Therefore, in addition to the above-mentioned effects on the systemic circulatory system, such as lowering blood pressure and reducing blood lipid, blood glucose and cholesterol levels, alterations in the composition and functions of intestinal microorganisms play a key role in the pathological development of CVD[59]. Gut bacteria produce specific metabolites, such as short-chain fatty acids, branched-chain fatty acids, aromatic acids, trimethylamine/trimethylamine-N-oxide and secondary bile acids, which are considered to be closely related to the occurrence of CVD[60].

By comparing oral and intravenous administrations of kaempferol in mice with collagen arthritis, it has been found that the concentration of kaempferol and its metabolites in the intestinal cavity after oral administration is considerably higher than that in the blood circulation. The high level of kaempferol in the intestine can regulate intestinal flora and microbial metabolism, which may be related to the anti-arthritis activity of kaempferol[48,61]. Therefore, the gut microbiota may be an important potential target for exploring the efficacy of flavonoids in the treatment of CVD.

Meanwhile, although *G. biloba* has been shown to have a positive safety profile in numerous randomized clinical trials, some issues remain to be considered. For example, the effects of certain plant components on the development of CVD are not well known. Most studies on the use of *G. biloba* have focused on its flavonoids, such as quercetin, kaempferol and luteolin. However, the effects of other bioactive compounds remain unknown.

Pharmacokinetics of flavonoids: Human pharmacokinetic studies of *G. biloba* flavonoid components are limited. Ude *et al*.[62] conducted a preliminary study on two healthy volunteers to explore the plasma pharmacokinetics of *G. biloba* flavonoids orally administered at 50, 100 and 300 mg concentrations. The time of maximum concentration (T_{max}) of flavonoids was 2-2.5 hrs and the elimination half-life was 2-4 hrs. No flavonoids were detected in the plasma after 24 hrs. Guan *et al*.[63] analyzed and compared the pharmacokinetic characteristics of *G. biloba* glycosides in three different *G. biloba* extract preparations (drops, capsules and tablets). The T_{max} of each flavonoid was approximately 2 hrs and the flavonoids were completely undetectable 24 hrs after a single oral administration of *G. biloba* extract, showing a large distribution volume. The half-life in the plasma (approximately 3 hrs) and the *in vivo* clearance rate (approximately 39-53 L/hrs) indicate the rapid elimination of flavonoids from the blood. In a study, 10 volunteers, all aged 28 years, were administered a single dose of *G. biloba* extract. The levels of various substances in plasma, such as quercetin and kaempferol, were analyzed using reverse-phase liquid chromatography. The results revealed that the substances found in the extract were mainly excreted in urine[32,64].

The absorption mechanism of flavonoid glycosides can be divided into two types. Firstly, the absorption of flavonol glucoside can be mediated by lactase-phlorizin hydrolase (LPH) or Sodium-Dependent Glucose Transporter (SGLT1) in the small intestine. Second, non-glucosides are poorly absorbed in the small intestine and most of them migrate to the colon, where they are hydrolyzed to a glycogen by colonic microbes and then absorbed into the body.

The ability of P-glycoprotein to transport various compounds from the interior of the cell to the extracellular space is mediated by adenosine triphosphate (ATP)-dependent efflux pumps. The P-glycoprotein-mediated efflux has been identified as one of the factors contributing to the low levels of flavonoids[65]. However, a previous study also found that certain substances, such as isorhamnetin, kaempferol and quercetin, inhibit the efflux-mediated activity of P-glycoproteins[66]. Meanwhile, fermentation by the small

intestinal flora allows the hydrolysis and glucuronidation of flavonoids, resulting in the formation of aglycones[52]. These compounds are metabolized by the liver enzyme UDP-Glucuronosyltransferase 1A9 (UGT1A9). Flavonoids have low bioavailability because of first-pass effects and are primarily absorbed as aglycones or in the form of glucuronate or sulfate[67].

The interactions between efflux pumps and intestinal metabolites can also contribute to the development of a connecting barrier, which prevents the absorption of flavonoid aglycones[68]. The first-pass effect of the flavonoids from *G. biloba* extract is believed to be responsible for the low concentrations of these compounds in the oral form. The complex components of *G. biloba* extract make it difficult to determine the exact effects of a single compound on different components. Meanwhile, the various components of *G. biloba* extract can interact with each other to cause changes in its pharmacokinetics. Therefore, it is necessary to strengthen the clinical pharmacokinetic study of *G. biloba* flavonoids.

The multi-component pharmacokinetic study of *G. biloba* should be based on the clinical human pharmacokinetics study in healthy people. However, limited by ethical issues, human pharmacokinetics experiments alone cannot fully reflect the *in vivo* exposure and processes of *G. biloba* extracts. Therefore, *in vitro* supplement experiments and humanized animal experiments should be combined. Based on reducing the differences in species as much as possible, it is necessary to objectively reflect the material basis and pharmacokinetic characteristics of the curative effect of traditional Chinese medicine to better guide the clinical medication.

Some questions remain unanswered in the pharmacokinetic study of *G. biloba* flavonoids. What is the level and form of systemic exposure in the human body after administering *G. biloba* extract preparations? Is it possible to reach the concentration at which *in vitro* pharmacodynamic activity can be achieved? For gut microbiota-related targets in the treatment of CVD, what substances can contact and act on the gut microbiota? In light of these knowledge gaps, further studies are required to identify the interactions between the bioactive components of plants and conventional herbal products.

CONCLUSION

As we all know, according to the evidence available in the literature, the use of *G. biloba* extract can potentially help improve the symptoms of CVD. However, high-quality clinical trials are necessary to confirm the effectiveness of *G. biloba* extracts. This process can help researchers and physicians make informed decisions regarding product use. At the same time, the transformation of flavonoids caused by intestinal flora and the host and the targets of flavonoids for intestinal flora and the host should be considered. The mechanism of action of *G. biloba* flavonoids can be more scientifically revealed only by comprehensively considering the relationship among intestinal flora, host and *G. biloba* flavonoids.

SIGNIFICANCE STATEMENT

This review analyzes the clinical pharmacodynamics and pharmacokinetics of *Ginkgo biloba* flavonoids, thereby providing valuable information on the cardioprotective effects of this plant and laying a foundation for its clinical application. It points out that the current study cannot scientifically elucidate how *G. biloba* flavonoids exert their cardiovascular therapeutic effects. The mechanism of action of *G. biloba* flavonoids can be more scientifically revealed only by comprehensively considering the relationship among intestinal flora, host and *G. biloba* flavonoids.

REFERENCES

1. Andersson, C. and R.S. Vasan, 2018. Epidemiology of cardiovascular disease in young individuals. Nat. Rev. Cardiol., 15: 230-240.
2. Yang, Y., Y. Li , J. Wang, K. Sun and W. Tao *et al*., 2017. Systematic investigation of *Ginkgo biloba* leaves for treating cardio-cerebrovascular diseases in an animal model. ACS Chem. Biol., 12: 1363-1372.
3. Peterson, J.J., J.T. Dwyer, P.F. Jacques and M.L. McCullough, 2012. Associations between flavonoids and cardiovascular disease incidence or mortality in European and US populations. Nutr. Rev., 70: 491-508.
4. Satoh, H. and S. Nishida, 2004. Electropharmacological actions of *Ginkgo biloba* extract on vascular smooth and heart muscles. Clin. Chim. Acta, 342: 13-22.
5. Huang, X.F., S.Z. Zhang, Y.Y. You, N. Zhang, H. Lu, A. Daugherty and X.J. Xie, 2019. *Ginkgo biloba* extracts prevent aortic rupture in angiotensin II-infused hypercholesterolemic mice. Acta Pharmacol. Sin., 40: 192-198.
6. Du, Y., B.J. Xu, X. Deng, X.W. Wu and Y.J. Li *et al*., 2019. Predictive metabolic signatures for the occurrence and development of diabetic nephropathy and the intervention of *Ginkgo biloba* leaves extract based on gas or liquid chromatography with mass spectrometry. J. Pharm. Biomed. Anal., 166: 30-39.

7. Li, T., Y. Zhang, J. Tian, L. Yang and J. Wang, 2019. *Ginkgo biloba* pretreatment attenuates myocardial ischemia-reperfusion injury via mitoBK$_{Ca}$. Am. J. Chin. Med., 47: 1057-1073.
8. Trumbeckaite, S., J. Bernatoniene, D. Majiene, V. Jakštas, A. Savickas and A. Toleikis, 2007. Effect of *Ginkgo biloba* extract on the rat heart mitochondrial function. J. Ethnopharmacol., 111: 512-516.
9. Zheng, X., Q. Gao, S. Liang, G. Zhu, D. Wang and Y. Feng, 2021. Cardioprotective properties of *Ginkgo biloba* extract 80 *via* the activation of AKT/GSK3β/β-catenin signaling pathway. Front. Mol. Biosci., Vol. 8. 10.3389/fmolb.2021.771208.
10. Major, R.T., 1967. The ginkgo, the most ancient living tree: The resistance of *Ginkgo biloba* L. to pests accounts in part for the longevity of this species. Science, 157: 1270-1273.
11. van Beek, T.A. and P. Montoro, 2009. Chemical analysis and quality control of *Ginkgo biloba* leaves, extracts and phytopharmaceuticals. J. Chromatogr. A, 1216: 2002-2032.
12. Singh, B., P. Kaur, R.D. Singh and P.S. Ahuja, 2008. Biology and chemistry of *Ginkgo biloba*. Fitoterapia, 79: 401-418.
13. Gertz, H.J. and M. Kiefer, 2004. Review about *Ginkgo biloba* special extract EGb 761 (Ginkgo). Curr. Pharm. Des., 10: 261-264.
14. do Socorro S. Chagas, M., M.D. Behrens, C.J. Moragas-Tellis, G.X.M. Penedo, A.R. Silva and C.F. Gonçalves-de-Albuquerque, 2022. Flavonols and flavones as potential anti-inflammatory, antioxidant, and antibacterial compounds. Oxid. Med. Cell. Longevity, Vol. 2022. 10.1155/2022/9966750.
15. Kato-Noguchi, H., 2024. Isolation and identification of allelochemicals and their activities and functions. J. Pestic. Sci., 49: 1-14.
16. DeKosky, S.T. and C.D. Furberg, 2008. Turning over a new leaf: *Ginkgo biloba* in prevention of dementia? Neurology, 70: 1730-1731.
17. Suroowan, S. and M.F. Mahomoodally, 2019. Herbal medicine of the 21st century: A focus on the chemistry, pharmacokinetics and toxicity of five widely advocated phytotherapies. Curr. Top. Med. Chem., 19: 2718-2738.
18. Isah, T., 2015. Rethinking *Ginkgo biloba* L.: Medicinal uses and conservation. Pharmacogn. Rev., 9: 140-148.
19. Pan, S., A. Neeraj, K.S. Srivastava, P. Kishore, M.K. Danquah and I.P. Sarethy, 2013. A proposal for a quality system for herbal products. J. Pharm. Sci., 102: 4230-4241.
20. van Beek, T.A., 2002. Chemical analysis of *Ginkgo biloba* leaves and extracts. J. Chromatogr. A., 967: 21-55.
21. Liu, L., Y. Wang, J. Zhang and S. Wang, 2021. Advances in the chemical constituents and chemical analysis of *Ginkgo biloba* leaf, extract, and phytopharmaceuticals. J. Pharm. Biomed. Anal., Vol. 193. 10.1016/j.jpba.2020.113704.
22. Liu, X.G., S.Q. Wu, P. Li and H. Yang, 2015. Advancement in the chemical analysis and quality control of flavonoid in *Ginkgo biloba*. J. Pharm. Biomed. Anal., 113: 212-225.
23. Strømgaard, K. and K. Nakanishi, 2004. Chemistry and biology of terpene trilactones from *Ginkgo biloba*. Angew. Chem. Int. Edn., 43: 1640-1658.
24. Lin, Y., R. Shi, X. Wang and H.M. Shen, 2008. Luteolin, a flavonoid with potential for cancer prevention and therapy. Curr. Cancer Drug Targets, 8: 634-646.
25. Singh, D., W.C. Cho and G. Upadhyay, 2016. Drug-induced liver toxicity and prevention by herbal antioxidants: An overview. Front. Physiol., Vol. 6. 10.3389/fphys.2015.00363.
26. Ude, C., A. Paulke, M. Schubert-Zsilavecz and M. Wurglics, 2009. Chemistry, pharmacokinetics and metabolism of *Ginkgo* extract. One of the best-studied phytopharmaceuticals. Pharm. Unserer Zeit, 38: 418-423.
27. Kakigi, Y., T. Hakamatsuka, T. Icho, Y. Goda and N. Mochizuki, 2012. Comprehensive analysis of flavonols in *Ginkgo biloba* products by ultra-high-performance liquid chromatography coupled with ultra-violet detection and time-of-flight mass spectrometry. Biosci. Biotechnol. Biochem., 76: 1003-1007.
28. Sun, T., X. Wang and H. Xu, 2015. *Ginkgo biloba* extract for angina pectoris: A systematic review. Chin. J. Integr. Med., 21: 542-550.
29. Chen, H., C. Zhou, M. Yu, S. Feng and Y. Ma *et al*., 2019. The effect of *Ginkgo biloba* dropping pills on hemorheology and blood lipid: A systematic review of randomized trials. Evidence-Based Complementary Altern. Med., Vol. 2019. 10.1155/2019/2609625.
30. Mozet, C., R. Martin, K. Welt and G. Fitzl, 2009. Cardioprotective effect of EGb 761 on myocardial ultrastructure of young and old rat heart and antioxidant status during acute hypoxia. Aging Clin. Exp. Res., 21: 14-21.
31. Kang, H., 2017. Hypocholesterolemic effect of *Ginkgo biloba* seeds extract from high fat diet mice. Biomed. Sci. Lett., 23: 138-143.
32. Fan, Y., X. Jin, C. Man and D. Gong, 2018. Does adjuvant treatment with *Ginkgo biloba* to statins have additional benefits in patients with dyslipidemia? Front. Pharmacol., Vol. 9. 10.3389/fphar.2018.00659.
33. Abdel-Zaher, A.O., H.S.M. Farghaly, A.E.M. El-Refaiy and A.M. Abd-Eldayem, 2018. Protective effect of the standardized leaf extract of *Ginkgo biloba* (EGb761) against hypertension-induced renal injury in rats. Clin. Exp. Hypertens., 40: 703-714.
34. Lu, S., X. Guo and P. Zhao, 2011. Effect of *Ginkgo biloba* extract 50 on immunity and antioxidant enzyme activities in ischemia reperfusion rats. Molecules, 16: 9194-9206.
35. Xiao, G., M. Lyu, Y. Wang, S. He and X. Liu *et al*., 2019. Ginkgo flavonol glycosides or ginkgolides tend to differentially protect myocardial or cerebral ischemia-reperfusion injury via regulation of TWEAK-Fn14 signaling in heart and brain. Front. Pharmacol., Vol. 10. 10.3389/fphar.2019.00735.

36. Tosaki, A., T. Pali and M.T. Droy-Lefaix, 1996. Effects of *Ginkgo biloba* extract and preconditioning on the diabetic rat myocardium. Diabetologia, 39: 1255-1262.
37. Erbil, G., S. Ozbal, U. Sonmez, C. Pekcetin, K. Tugyan, A. Bagriyanik and C. Ozogul, 2008. Neuroprotective effects of selenium and *Ginkgo biloba* extract (EGb761) against ischemia and reperfusion injury in rat brain. Neurosci. J., 13: 233-238.
38. Loh, K.P., L.S. Low, W.H. Wong, S. Zhou and S.H. Huang *et al*., 2006. A comparison study of cerebral protection using *Ginkgo biloba* extract and Losartan on stroked rats. Neurosci. Lett., 398: 28-33.
39. Uríková, A., E. Babušíková, D. Dobrota, A. Drgová, P. Kaplán, Z. Tatarková and J. Lehotský, 2006. Impact of *Ginkgo biloba* extract *EGb 761* on ischemia/reperfusion-Induced oxidative stress products formation in rat forebrain. Cell. Mol. Neurobiol., 26: 1341-1351.
40. Chandrasekaran, K., Z. Mehrabian, B. Spinnewyn, C. Chinopoulos, K. Drieu and G. Fiskum, 2003. Neuroprotective effects of bilobalide, a component of *Ginkgo biloba* extract (EGb 761®) in global brain ischemia and in excitotoxicity-induced neuronal death. Pharmacopsychiatry, 36: 89-94.
41. Wu, Y.Z., S.Q. Li, X.G. Zu, J. Du and F.F. Wang, 2008. *Ginkgo biloba* extract improves coronary artery circulation in patients with coronary artery disease: Contribution of plasma nitric oxide and endothelin 1. Phytother. Res., 22: 734-739.
42. Rodríguez, M., L. Ringstad, P. Schäfer, S. Just, H.W. Hofer, M. Malmsten and G. Siegel, 2007. Reduction of atherosclerotic nanoplaque formation and size by *Ginkgo biloba* (EGb 761) in cardiovascular high-risk patients. Atherosclerosis, 192: 438-444.
43. Clayton, Z.S., D.A. Hutton, V.E. Brunt, N.S. VanDongen and B.P. Ziemba *et al*., 2021. Apigenin restores endothelial function by ameliorating oxidative stress, reverses aortic stiffening, and mitigates vascular inflammation with aging. Am. J. Physiol. Heart Circ. Physiol., 321: H185-H196.
44. Kuller, L.H., D.G. Ives, A.L. Fitzpatrick, M.C. Carlson and C. Mercado *et al*., 2010. Does *Ginkgo biloba* reduce the risk of cardiovascular events? Circ.: Cardiovasc. Qual. Outcomes, 3: 41-47.
45. Lin, L.Z., P. Chen, M. Ozcan and J.M. Harnly, 2008. Chromatographic profiles and ıdentification of new phenolic components of *Ginkgo biloba* leaves and selected products. J. Agric. Food Chem., 56: 6671-6679.
46. Sarikçioğlu, S.B., G. Öner and E. Tercan, 2004. Antioxidant effect of EGb 761 on hydrogen peroxide-induced lipoperoxidation of G-6-PD de cient erythrocytes. Phytother. Res., 18: 837-840.
47. Shaito, A., D.T.B. Thuan, H.T. Phu, T.H.D. Nguyen and H. Hasan *et al*., 2020. Herbal medicine for cardiovascular diseases: Efficacy, mechanisms, and safety. Front. Pharmacol., Vol. 11. 10.3389/fphar.2020.00422.
48. Ding, X., L. Zheng, B. Yang, X. Wang and Y. Ying, 2019. Luteolin attenuates atherosclerosis via modulating signal transducer and activator of transcription 3-mediated inflammatory response. Drug Des. Dev. Ther., 13: 3899-3911.
49. Qian, L.B., H.P. Wang, Y. Chen, F.X. Chen, Y.Y. Ma, I.C. Bruce and Q. Xia, 2010. Luteolin reduces high glucose-mediated impairment of endothelium-dependent relaxation in rat aorta by reducing oxidative stress. Pharmacol. Res., 61: 281-287.
50. Chen, H.I., W.S. Hu, M.Y. Hung, H.C. Ou and S.H. Huang *et al*., 2020. Protective effects of luteolin against oxidative stress and mitochondrial dysfunction in endothelial cells. Nutr. Metab. Cardiovasc. Dis., 30: 1032-1043.
51. Li, W., L. Chen and Y. Xiao, 2020. Apigenin protects against ischemia-/hypoxia-induced myocardial injury by mediating pyroptosis and apoptosis. *In vitro* Cell. Dev. Biol. Anim., 56: 307-312.
52. Zhou, Z., Y. Zhang, L. Lin and J. Zhou, 2018. Apigenin suppresses the apoptosis of H9C2 rat cardiomyocytes subjected to myocardial ischemia-reperfusion injury via upregulation of the PI3K/Akt pathway. Mol. Med. Rep., 18: 1560-1570.
53. Du, H., J. Hao, F. Liu, J. Lu and X. Yang, 2015. Apigenin attenuates acute myocardial infarction of rats via the inhibitions of matrix metalloprotease-9 andinflammatory reactions. Int. J. Clin. Exp. Med., 8: 8854-8859.
54. McEwen, B.J., 2015. The influence of herbal medicine on platelet function and coagulation: A narrative review. Semin. Thromb. Hemost., 41: 300-314.
55. Lan, W.J. and X.X. Zheng, 2006. Activity of *Ginkgo biloba* extract and quercetin on thrombomodulin expression and tissue-type plasminogen activator secretion by human umbilical vein endothelial cells. Biomed. Environ. Sci., 19: 249-253.
56. Nishida, S. and H. Satoh, 2004. Comparative vasodilating actions among terpenoids and flavonoids contained in *Ginkgo biloba* extract. Clin. Chim. Acta, 339: 129-133.
57. Hu, W.H., H.Y. Wang, Y.T. Xia, D.K. Dai and Q.P. Xiong *et al*., 2020. Kaempferol, a major flavonoid in ginkgo folium, potentiates angiogenic functions in cultured endothelial cells by binding to vascular endothelial growth factor. Front. Pharmacol., Vol. 11. 10.3389/fphar.2020.00526.
58. Edmands, W.M.B., P. Ferrari, J.A. Rothwell, S. Rinaldi and N. Slimani *et al*., 2015. Polyphenol metabolome in human urine and its association with intake of polyphenol-rich foods across European countries. Am. J. Clin. Nutr., 102: 905-913.
59. Walther, G., P. Obert, F. Dutheil, R. Chapier and B. Lesourd *et al*., 2015. Metabolic syndrome individuals with and without type 2 diabetes mellitus present generalized vascular dysfunction: Cross-sectional study. Arterioscler. Thromb Vasc. Biol., 35: 1022-1029.

60. Jacqueline, B., J.C. Aristizabal, C.N. Blesso, J.S. Volek and F.M. Luz, 2012. Grape polyphenols reduce blood pressure and increase flow-mediated vasodilation in men with metabolic syndrome. J. Nutr., 142: 1626-1632.
61. Aa, L.X., F. Fei, Q. Qi, R.B. Sun and S.H. Gu *et al*., 2020. Rebalancing of the gut flora and microbial metabolism is responsible for the anti-arthritis effect of kaempferol. Acta Pharmacol. Sin., 41: 73-81.
62. Ude, C., M. Schubert-Zsilavecz and M. Wurglics, 2013. *Ginkgo biloba* extracts: A review of the pharmacokinetics of the active ingredients. Clin. Pharmacokinet., 52: 727-749.
63. Guan, H., D. Qian, H. Ren, W. Zhang, H. Nie, E. Shang and J. Duan, 2014. Interactions of pharmacokinetic profile of different parts from *Ginkgo biloba* extract in rats. J. Ethnopharmacol., 155: 758-768.
64. Wang, F.M., T.W. Yao and S. Zeng, 2003. Disposition of quercetin and kaempferol in human following an oral administration of *Ginkgo biloba* extract tablets. Eur. J. Drug Metab. Pharmacokinet., 28: 173-177.
65. Wang, Y., J. Cao and S. Zeng, 2005. Involvement of P-glycoprotein in regulating cellular levels of Ginkgo flavonols: Quercetin, kaempferol, and isorhamnetin. J. Pharm. Pharmacol., 57: 751-758.
66. Etheridge, A.S., S.R. Black, P.R. Patel, J. So and J.M. Mathews, 2007. An *in vitro* evaluation of cytochrome p450 inhibition and p-glycoprotein interaction with goldenseal, *Ginkgo biloba*, grape seed, milk thistle, and ginseng extracts and their constituents. Planta Med., 73: 731-741.
67. Dabeek, W.M. and M.V. Marra, 2019. Dietary quercetin and kaempferol: Bioavailability and potential cardiovascular-related bioactivity in humans. Nutrients, Vol. 11. 10.3390/nu11102288.
68. Tao, Y., F. Zhu, M. Pan, Q. Liu and P. Wang, 2022. Pharmacokinetic, metabolism, and metabolomic strategies provide deep insight into the underlying mechanism of *Ginkgo biloba* flavonoids in the treatment of cardiovascular disease. Front. Nutr., Vol. 9. 10.3389/fnut.2022.857370.