

# *Review Article*

# Antidiabetic Potential of Phytochemicals Found in *Vernonia amygdalina*

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Type 2 diabetes mellitus (T2DM), or "insulin-independent diabetes mellitus," is a worldwide health concern. Diabetes affects roughly 415 million individuals worldwide, with 193 million undiagnosed cases. The number of people afflicted in the following decades is predicted to double. Although various synthetic medications are currently available to treat/manage T2DM, their side effects compel researchers to seek novel treatment options. Because of their affinity for biological receptors and broad bioactivity, nature has long been a source of innovative medication. *V. amygdalina* is one of the numerous natural productswith antidiabetic properties. Several studies have shown that the extracts have antidiabetic effects in vitro and in vivo. This review examined the antidiabetic and pharmacokinetic characteristics of phytoconstituents found in *V. amygdalina*.

#### 1. Introduction

Diabetes, which is characterized by the body's difficulty in effectively regulating blood sugar and insulin levels, is one of the major silent killers worldwide.In 2021, more than 537 million people between the ages of 20 and 79 were diagnosed with diabetes mellitus globally. It is estimated that 643 million people are expected to have diabetes by 2030 and a staggering 783 million by 2045 [1]. The expenditure on diabetes is rapidly increasing worldwide, and it varies significantly from one country to another. In India, the number of diabetic cases has seen a significant upsurge (7.1% in 2009 to 8.9% in 2019) in the last few decades, which is predicted to rise in the coming years, and will hugely enhance the burden on finance as well as the healthcare sector [2]. -Type 2 diabetes mellitus (T2DM) is brought on by the interaction of a genetic predisposition and a wide range of modifiable and

nonmodifiable environmental risk factors, including obesity, a poor diet, and inactivity [3–6]. Additionally, diabetes does not attack end-organ damage alone. For instance, cardiovascular disease (CVD) progresses, and eyesight loss and kidney failure might result from the vascular and nerve damage brought on by chronic hyperglycemia. The main factor in preventable blindness is diabetes-induced retinopathy, while the main factor in kidney disease is diabetic nephropathy. Therefore, diabetes must be managed appropriately to reduce the emergence of chronic problems. Accordingly, the current therapeutic strategy includes a variety of medication classes. However, their adverse interactions and inadequate efficacy compelling researchers to develop safe alternatives. Cragg-Newman found that out of the 63 antidiabetic genre drugs approved between January 1, 1981, and September 30, 2019, biological macromolecules occupied the largest portion ( 38%), followed by synthetic

drugs inspired by natural products(25.4%), and unmodified natural products and their derivatives (14.3%). This data strongly highlights the importance of natural products [7].

Vernonia amygdalina (V. amygdalina) is a member of the Asteraceae family with dark green leaves and tiny, thistle-shaped flowers. This plant grows primarily in tropical Africa and is used as an herb and vegetable. Earlier studies suggest that this vegetable demonstrates a strong anticancer effect against breast cancer cell lines via inhibiting the DNA synthesis [8]. Today, various classes of phytochemicals (e.g., sesquiterpene lactones, steroid glycosides, and saponins) are known from this plant, making it useful for the management of human and animal diseases [9–11]. This review makes an earnest effort to put together its promise as a diabetes treatment, the gaps in existing research, and its potential future possibilities. The survey of the existing literature has been conducted using the PubMed and SCOPUS databases. The selected search period is from 2005 to 2022.

## 2. Type 2 Diabetes Mellitus (T2DM): Molecular Pathology and Medications

Although T2DM has been known to humans for ages, its complex nature and the lack of clarity in understanding the mechanistic aspects pose significant challenge. Initially thought to be a chromosomal polygene recessive condition with abnormal insulin secretion rates, the inheritance mode of type 2 diabetes (T2DM) remains uncertain. It is now understood that a complex interplay of genetic factors and external influences, such as stress and chemicals, contributes to its development. Disruptions in glucose metabolism, incretin production, and insulin sensitivity involving key enzymes like  $\alpha$ -glucosidase,  $\alpha$ -amylase, DPP-4 (dipeptidyl peptidase 4), PPAR (peroxisome proliferator-activated receptor), PTP1B (protein tyrosine phosphatase 1B), and GLUT4 (glucose transporter type 4) play a central role in T2DM pathophysiology.  $\alpha$ -amylase breaks down starch into oligosaccharides known as limit dextrins [12], which are further hydrolyzed into absorbable glucose in the small intestine by  $\alpha$ -glucosidase. Elevated blood glucose levels stimulate the synthesis of glucagon-like peptide-1 (GLP-1), which aids in insulin secretion and pancreatic cell protection. However, GLP-1 is rapidly degraded by DPP-4, disrupting glucose metabolism [13]. Insulin also influences lipid metabolism and glycogen production in the liver and muscles, necessitating regulation through the GLUT4, PTP1B, and PPAR pathways [13]. GLUT4 is crucial for postmeal glucose clearance, but altering its expression can disturb glucose homeostasis and promote insulin resistance [14]. Insulin-dependent GLUT4 translocation involves a complex cascade of biochemical signals, including tyrosine kinase activation, IRS1 substrate phosphorylation, and PI3K/Akt, CAP/Cb1/Tc10, or Ras-MAPK pathway activation. Simultaneously, GLUT4 vesicles move from intracellular pools to the cell membrane to facilitate glucose uptake [15]. Leptin enhances glucose absorption and energy metabolism by binding to its receptors and phosphorylating the JAK2/STAT pathway. PTP1B, found in the endoplasmic reticulum, skeletal muscle, and liver [16], dephosphorylates

and inactivates these signal transduction cascades, making it a potential target against T2DM [17]. Non-insulin dependent GLUT4 translocation can be induced by factors like severe exercise, increased Ca<sup>2+</sup>, and bradykinin [18], which stimulate GLUT4 translocation to the plasma membrane, aiding in efficient glucose control [19]. Peroxisome proliferator-activated receptor gamma (PPAR) plays a critical role in adipocyte development and is involved in lipid and glucose metabolism regulation [20]. Activating PPAR improves GLUT-4 and GLUT-1 mobility, facilitating glucose uptake in skeletal muscles and the liver. Inflammation is closely linked to obesity and T2DM [21, 22], with PPAR agonists showing promise in improving insulin sensitivity by reducing TNF- $\alpha$  [23] and increasing adiponectin expression [24]. Inflammatory substances like TNF- $\alpha$  impact insulin resistance through NF- $\kappa$ B signaling pathways, making this pathway a potential target for diabetes control [25]. Diabetes can increase tissue sensitivity to damage from reactive oxygen species (ROS) and free radicals, leading to oxidative liver damage and decreased antioxidant enzyme function [25]. This exacerbates insulin resistance and pancreatic cell dysfunction. Epigenetic alterations in DNA and histones are being explored through omics, including proteomics, genomics, and transcriptomics, to gain a better understanding of T2DM into pathogenesis [26, 27]. Despite these advancements, insights into the pathological mechanisms of T2DM continue to drive the discovery of novel drugs.

To treat T2DM, synthetic agents of various classes are available. This includes, but is not limited to, glimepiride (class: sulfonylurea), rosiglitazone (class: thiazolidinedione), metformin (class: biguanide), dapagliflozin (SGLT2 inhibitor) [28]. However, side effects associated with the available synthetic agents are a major concern. For example, rosiglitazone is a thiazolidinedione-based compound that enhances insulin's effectiveness by promoting hepatic function. Nevertheless, there have been reports of cardiac failure as an adverse outcome. Similarly, metformin is a biguanide that acts by decreasing hepatic glucose output [28]. In addition to synthetic agents, natural product-based antidiabetic drugs are also used globally [29].

### 3. Phytochemical Composition and Biological Effect of V. amygdalina Extract

Among numerous species of plants with medicinal applications, *V. amygdalina* is one of the most unique species. Not only are they rich in different classes of phytochemicals but they also exhibit wide ranging bioactivities as well as taste [30, 31]. Various elegant reviews have been published on the phytochemical and pharmacological applications of *V. amygdalina*. [11, 32–45] Some selected examples of phytochemicals that are found in *V. amygdalina* are collected in Table 1 and shown in Figure 1. Among others, constituents such as 1,3-, 1,4-, 3,4-, 3,5-, 1,5-,4,5- dicaffeoyl quinic acids 1,3,5-, 3,4,5 tricaffeoyl quinic acid, chlorogenic acid, vernodalin, and vernomygdin, terpene lactones, essential oils, fatty acids, metal ions, peptides, anthocyanins, and alkaloids have been reported. [42, 46]. The presence of steroidal saponins such as vernoniamyoside A-D, vernonioside D, and vernonioside B2 in leaves has also been confirmed [47]. Recently, Nowak et al. [48] carried out the identification and quantification of polyphenolics present in methanolic extract of leaves. Venditti and coworkers [49] carried out a bioassay-guided study to identify antidiabetic components found in the methanolic extract of *V. amygdalina*. They were able to identify 11 $\beta$ , 13dihydrovernolide as one of the components responsible for the antidiabetic effect. Around three decades ago, vernoniosides A1, A2, and A3 and vernonioside B1 were reported in the methanolic extract of stem bark [31]. In a recent work, Nugyen et al. [50] identified a novel compound vernonioside V (a stigmastane) present in the ethanolic extract of *V. amygdalina* leaves.

Numerous studies have indicated that the organic and aqueous extracts of different parts of V. amygdalina plant possess unique bioactivities [49]. For example, the ethanolic extract of the leaves has been found to suppress inflammatory factors like TNF, IL8, and IL6 [50]. The saponins found in V. amygdalina exhibit selective cytotoxicity against cancerous cell lines [47]. Following this work, Ejiofor et al. [51] studied the antidiabetic, anthelminthic, and antioxidant properties of the methanolic extract V. amygdalina stem bark, which contains phytochemicals responsible for the bitter taste [31]. Sesquiterpene vernodalin and vernomygdin have been reported to show anticancer activity against nasopharynx cell lines [42]. Anh et al. [8] reported the antidiabetic properties of V. amygdalina leaves. They found that the stigmastane type saponin vernoamyoside E shows remarkable antidiabetic property. Similarly, vernoniacum B was found to be mildly active against  $\alpha$  amylase. [8].

## 4. Antidiabetic Potential of V. amygdalina Extracts

In 1992, Akah and Okafor [52] reported the antidiabetic effect of V. amygdalina extract. They noted that the intraperitoneal injection of aqueous leaf extract in rabbits leads to insulin secretion- independent hypoglycemia. However, later studies found that V. *amygdalina* extracts assist in  $\beta$ -cell regrowth. It was noted that when V. amygdalina was given intragastrically to alloxanized rats for two weeks, its blood and serum glucose levels reduced significantly as compared to the control [53, 54]. Histomorphological analysis revealed the regeneration  $\beta$ -cell regrowth, which is quite an interesting finding. Another clinical study revealed that V. amygdalina produced significantly higher hypoglycaemic effects than the other vegetables at the majority of postprandial time points [55]. Saliu et al. [56] investigated the inhibitory impact of free and bound phenol extracts of it on glucosidase and amylase activities in vitro. They found a significant inhibition in amylase and glucosidase activities in a dose-dependent manner (4-16 g/ml). It was noted that glucosidase had a more significant inhibitory effect than amylase [56]. To understand how the combined extracts of V. amygdalina and Gongronema latifolium affect the pancreatic  $\beta$ -cells, Akpaso et al. [54] conducted a study on streptozotocin-induced diabetic Wistar rats. They noted

that, among other changes, there was a decrease in blood glucose by 12.49% and 14.96% during a 28-day treatment period compared to diabetes control. The synergistic effect of V. amygdalina extract on the antidiabetic activity of a synthetic drug has also been studied. Michael et al. [53] investigated the antidiabetic activity of an admix of metformin and an aqueous extract of V. amygdalina leaves. They found that a mixture in a ratio of 1:2 significantly reduced the blood sugar levels, while a 2:1 mixture was found to be better when taking drug safety into account [53]. Following this study, Atangwho et al. [57] studied the effect of a herbal combination of V. amygdalina and Azadirachta indica and concluded that the use of combined extract is a useful way to control the glucose level [57]. Ethanolic extract of V. amygdalina was found to improve glucose tolerance in animal models, and an improvement of 32.1% in fasting blood glucose was seen after a 28-day treatment. Surprisingly, V. amygdalina reduced triglyceride and total cholesterol levels by 18.2% and 41%, respectively, and shielded pancreatic  $\beta$ -cells from STZ-related harm. In addition, it was determined that polyphenols were the main candidates for mediating V. amygdalina's antihyperglycemic effect by boosting GLUT 4 translocation and inhibiting hepatic G6Pase. [58]. [57] Erasto et al. [59] examined how leaf extract of V. amygdalina affects glucose utilization in different cells (chang-liver cells, C<sub>2</sub>C<sub>12</sub> muscles, and 3T3-L1). They found that the extracts, particularly aqueous ones, greatly enhanced glucose utilization in chang-liver cells and  $C_2C_{12}$  muscles. Onvibe et al. [60] found that glutathione S transferase (GSR) activity in alloxan-induced diabetic rats was similar to that of rats given metformin [60]. The importance of hepatic gluconeogenesis in glycogen metabolism is well known [61]. Wu et al. observed reduced expression of key enzymes involved in gluconeogenesis post-V. amygdalina extract administration by activating the AMPK pathway in palmitic acid-induced HepG2 cells [62]. However, they were unable to identify the key mechanism potentiating V. amygdalina's glucose-lowering effect. Ejiofor et al. [51] isolated various active compounds from the methanolic extract of V. amygdalina stem bark and noted that only vernoniaolide-glucoside showed an antidiabetic effect, while others were inactive. Similarly, vernonioside E, a saponin, was recently identified by Uti et al. as having the antidiabetic component of V. amygdalina [63]. Recently, Djeujo et al. [64] found that polyphenolic chemicals in aqueous extracts of V. amygdalina roots and leaves made them less cytotoxic than ethanolic extracts, which primarily inhibited glucosidase activity. Although those made from leaves showed less efficacy, aqueous root extracts displayed a concentration-dependent activity. In addition, 10 g/ml V. amygdalina extracts were also found to inhibit the production of advanced glycation end products (AGEs) [64]. The fact that phytochemicals, namely polyphenols, are thermally degraded into a combination of distinct isomers and derivatives, which affects the potency of extracts, is one factor that is also implicitly implied by the differential IC<sub>50</sub> values from different extraction procedures. Therefore, it is also essential to consider how to build an effective extraction technique.

		TABLE 1: Phytoconstituents of V. amygdalina and t	heir medicinal properties.	
S. No.	Class of compound	Constituents	Medicinal properties	Ref.
1 2	Steroid glycosides Flavonoids, saponins, tannins	Vernonoisides A1, A2, A3, A4, B1, B2, B3, D, and E Luteolin 7-O- $\beta$ glucuronoside, luteolin and lutelin7-O- $\beta$ glucoside	Anthelminthic, anti-inflammatory, and gastrointestinal disorders Hepatoprotective, anticancer, antiviral, anti-inflammatory, antimicrobia and antioxidant	[36–38] [39–41]
ŝ	Sesquiterpene lactones	Vernomigdin, vernodalin, vernodalol, vernolepin, vernodalol, epivernodalolvernolepin, vernolide, vernodalin, hydroxyvernolide, vernomygdin, vernomenin, vernonioside A1, A2, A3, A4, B1, B2, B3, and B4, 4,15 dihydrovernodalin, 1,2,11,12',3' hexahydrovernodalin,	Antifungal, anti-inflammatory, antibacterial, antiprotozoal, anaesthetic and antitumor	[37, 42, 43]
4	Edotides (peptides)	1,2,4,15,11,13,2',3' octahydro vernodalın, epivernodalol Andrographidoid A, E, arabinogalactan, 3,7,8-trimethoxy-1-hydroxyxanthone, 4,8-dihydroxy-2,7-dimethoxy xanthone, 1,2-dihydroxy-6, 8-dimethoxy-xanthone, and 3,7,8-trimethoxy-1-hydroxyxanthone	Anticancer, antioxidant	[31, 44]
5	Triterpenoids	Glycine, cysteine, pyridoxine, casein hydrolysate, thiamine, ascorbic acid, eucalyptol, alphamurolol, beta pinene, myrtenal	Hepatoprotective, anticancer, antioxidant, anti-inflammatory, analgesic, antibacterial, anti-nociceptive,	[30, 45]
Reprodı	iced under the terms of the C	C-BY creative commons attribution 4.0 International license (https://creativeco	ommons.org/licenses/by-sa/4.0/) [35].	



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FIGURE 1: Antidiabetic phytoconstituents reported in V. amydalina.

## 5. Structure-activity Relationship (SAR) of Phytoconstituents Found in V. amygdalina

5.1. Luteolin. Luteolin (3',4',5,7-tetrahydroxyflavone) is a flavonoid thatis commonly present in many phytoextracts, including the V. amygdalina extracts. [8] It displays a broad spectrum of biological activities in both its aglycone and glycosidic forms, including antioxidative, antiinflammatory, antibacterial, and anticancer properties [65]. Its tendency to reduce diabetic conditions in Type I/II diabetes models and diabetes-related cognitive loss and nephropathy in rats is well established [66, 67]. Besides, it was also noted that luteolin inhibits DPP-IV [68], LPS-induced TNF $\alpha$  release, and activation of the NF- $\kappa$ B pathway [69]. One of the important factors controlling insulin secretion is the MAFA protein, whose expression is negatively impacted by uric acid [70]. Matsuoka et al. [71]. It has been reported that luteolin restores the expression of MafA and suppresses the (NF)- $\kappa$ B-iNOS-NO pathway, preventing the pancreatic  $\beta$ cells from becoming dysfunctional [71]. When used in combination with acarbose, luteolin was found to show synergistic inhibition of  $\alpha$ -glucosidase [72]. Besides, this study also found that luteolin shows respectable activity when compared to other well-known flavonoids [72, 73].

Flavonoids are known to undergo substantial first-pass phase II metabolism in the liver and epithelial lining of the small intestines to generate methyl, gluconorate, and sulphate-conjugated metabolites [74]. They are mostly absorbed by the small intestines. Yasuda et al. [75] found that oral administration of *Chrysanthemum morifolium* extract to rats resulted in relatively rapid absorption and slow elimination of luteolin. Chen et al. [76] reported low excretion of luteolin (6.6%), suggesting probable metabolism of it into simple compounds or in vivo accumulation. However, the biosynthesis of luteolin involves the conversion of apigenin into luteolin. According to a study, luteolin and luteolin glucoside show an oral bioavailability of 10.6–26.6%, and luteolin-7-O-glucoside is primarily hydrolyzed to luteolin in the GI before being absorbed into the systemic circulation [77]. Kure et al. [78] reported that the metabolites luteolin 3'O glucuronide, luteolin-7-O-glucoside, and luteolin-4'-O found in the liver, kidney, and small intestine might be the bioactive component. A recent structure-activity relationship (SAR) study on this class of compound revealed that the presence of an OH group at the 3'-4' location of ring B in luteolin type 1 flavone is an important element for greater glucosidase inhibition. Similar to hydrogenation at positions 2 and 3 of ring C, glycosylation at positions 7 and 6 of ring A, and methoxylation at positions 3' and 4' of ring B, these modifications have been found to reduce glucosidase inhibitory activity [79].

5.2. Chlorogenic Acid Derivatives. Chlorogenic acid and related compounds are abundantly found in phenolic acid in V. amygdalina [58]. It was found that the chlorogenic acid derivatives reduces the postprandial glucose level [80, 81]. During a glucose tolerance test, Bassoli et al. [82] observed a decrease in plasma glucose levels due to chlorogenic acid, pointing to its potential involvement as a glycemic indexlowering drug. Chlorogenic acid has been found to improve lipid and glucose metabolism byactivating the AMPK pathway [83]. Zheng et al. [84] reported the mixed-type inhibitory activity of chlorogenic acid on swine pancreatic amylase. Oboh et al. suggested that chlorogenic acid inhibited  $\alpha$ -glucosidase with an IC<sub>50</sub> value of 9.24 g/ml in an in vitro model [85]. In addition, reports have shown that chlorogenic acid inhibits the activity of the enzymes maltase and sucrase, with  $IC_{50}$  values of 2.99 mM and 2.18 mM, respectively [85, 86]. Besides, 4,5-dicaffeoyl quinic acid and 3,5-dicaffeoyl quinic acid are also recognized for their ability to inhibit DPPIV and glucosidase, respectively [87]. Some work also reported that 1,5 dicaffeoylquinic acid had a dosedependent protective effect against glucotoxicity in RINm5F cells [88-90]. In 2007, Ren et al. [91] administered chlorogenic acid to rats and observed rapid absorption and a relatively slow distribution followed by a slower elimination phase. In 2011, Xie et al. discovered chlorogenic acid metabolites such as O-methyl CA, hydrolyzed chlorogenic acid, and glucuronide conjugates in rats after intravenous treatment [91]. It has been shown that the bioavailability of chlorogenic acid depends on gut microflora metabolism [92].

Keeping in mind the significance of the interplay of chemical dynamics among ligands and enzymes, Hemmerle et al. reported chlorogenic acid derivatives and studied their glucose -6-phosphate translocase inhibitory activity, which is one of the key enzymes for glucose homeostasis. They established that only one phenolic -OH moiety is enough for the action and increasing lipophilicity at position 1 enhanced the activity [93]. Amylase and glucosidase are more strongly inhibited by the acylated derivatives of chlorogenic acid [94]. It was observed that their inhibitory actions on enzymes spiraled upward with a positive shift in lipophilicity (mostly) with the highest activity in the  $C_{12}$  acylated group. This could be attributed to a stronger bifurcated hydrophobic interaction within the microenvironment of amino acid residues and secondary structures [94]. The topological polar surface area for caffeic acid is 77.8 Å<sup>2</sup>, while that for quinic acid is 118 Å<sup>2</sup>; therefore, as the degree of esterification of quinic acid with caffeic acid increases, its lipophilicity increases [95]. Therefore, chlorogenic acid is inherently less polar than one of its parents, quinic acid. Recently, Song et al. reported that caffeoyl substitution downturned the  $\alpha$  amylase inhibitory activity of quinic acid mediated via reduced binding affinity postsubstitution [96]. The effectiveness of these compounds was not as strong as acarbose, but it outperformed another widely recognized glucosidase inhibitor, 1-deoxynojirimycin hydrochloride. Cardullo et al. [97, 98] synthesized 11 amide derivatives of chlorogenic acid and found that one particular derivative, which featured a tertiary amine group on an alkyl chain and a benzothiazole scaffold, exhibited significantly lower IC50 values compared to chlorogenic acid (45.5  $\mu$ M for  $\alpha$ -Glu; 105.2  $\mu$ M for  $\alpha$ -Amy). These derivatives were notably more potent as  $\alpha$ -glucosidase inhibitors than the antidiabetic medication acarbose, which had an  $IC_{50} = 268.4 \,\mu\text{M}$ . Besides, amongst its derivatives, the inhibitory effect is a function of the number and position of the caffeoyl group.

5.3. Vernoniaolide Glucoside. The compound 6,10,14-trimethylheptadecan-15-olyl-15-O-D-glucopyranosyl-1,5 olide (vernoniaolide glucoside), which was isolated from the methanolic extract of stem bark [99], demonstrated to have antidiabetic properties that are yet to be fully understood. Structure-activity relationships and pharmacokinetic profiles remains to be investigated.

5.4. Vernonioside E. Vernonioside E, a stigmastane-type steroid, is a potential antidiabetic candidate. It was found that it imparts hepatoprotective effects, lowers blood glucose levels, improves the lipid profile, and decreases cardiovascular risks. [63] This hypoglycemic effect is likely caused by the activation of glucose-6-phosphate dehydrogenase via the shunt pathway, which enhances glucose oxidation, stimulation of insulin, regeneration of pancreatic cells, upregulation of enzymes involved in glucose metabolism, reduction of gluconeogenesis, and inhibition of glucose-6-phosphate and fructose-1, 6-bisphosphatases. However, whether the oxirane moiety, donor H- atoms, or acetoxy group could have a significant impact remains unclear.

5.5. Vernoamyoside E. Anh et al. [8] isolated vernonioside  $B_1$ , vernonioside  $B_2$ , vernoniacums B and vernoamyoside E. Vernonioside  $B_1$ , vernoniacums B, and vernoamyoside E have similar skeletons with a difference at  $C_{16}$ . Vernonioside  $B_1$ , vernoniacum B, and vernoamyoside E have –OH, –OAc, and –H at  $C_{16}$  of the cyclopentane ring, respectively. Vernoamyoside E was found to outperform the other derivatives and demonstrated a potent inhibitory action against glucosidase at 100 and 500 g/mL concentrations. All of them demonstrated inhibitory action against amylase, though less effectively than acarbose.

5.6.  $11\beta$ , 13-Dihydrovernolide. Okoduwa et al. [49] observed that  $11\beta$ , 13-dihydrovernolide possessed a hypoglycemic effect, which was attributed to the oxirane ring. However, additional studies are required to determine the mechanism.

#### 6. Toxicological Studies

The assessment of the toxicological and safety profiles of a natural product is essential, especially when it is prescribed as herbal medicines. In 2016, Jamil et al. [100] reported the positive toxicity of ethanol and hexane extracts of V. anthelmintica in the brine shrimp lethality (BSL) test. Perera and coworkers [101] reported a dose-dependent toxicity of a formulation that contains V. anthelmintica as one of the components. The BSL assay revealed that the aqueous extract of the formulation shows moderate lethality (>85% at a concentration over  $900 \,\mu g/mL$ ) with  $LD_{50} = 807.6 \pm 221.0 \,\mu g/mL$ . Methanolic and dichloromethane extracts have been found to show positive micronucleus tests, suggesting their clastogenic and or aneuploidic activity [102]. However, an independent study found that the intraperitoneal LD<sub>50</sub> of the crude methanolic V. glaberrima extract was 1265 mg/kg, indicating its fairly toxic nature [103]. V. amygdalina leaf extract exhibited no observable clinical signs of toxicity or adverse toxicological properties [104, 105]. It has also been found that the aqueous extract of V. amygdalina was more potent and less cytotoxic than the alcoholic extract [64]. Similarly, Autamashih et al. [106] reported that the crude extract of V. galamensis is relatively safe for oral use [106]. Table 2 collects the biological profile and mechanism of activity of different species of Veronia.

#### 7. Future Perspective

*V. amygdalina* extracts, characterized by their remarkable dose-dependent antidiabetic effects, likely mediated by compounds such as flavonoids, stigmastane-type steroids, and saponins, have exhibited promising potential. Notably,

	Ref.	[59] [56]	[64]	[8]	[107]	[52]	[108]	[58]	[09]	[62]	[49]	[63]	[109]	[109]	[109]	[110]	[111]	[112]	[107]
TABLE 2: Antidiabetic activity of plant parts of Vernonia species.	Mechanism	Stimulation of glucose Inhihition of a analase and a alucosidase	$\alpha$ glucosidase inhibition	Inhibition of $\alpha$ amylase and $\alpha$ glucosidase	Inhibition of $\alpha$ glucosidase	Hypoglycaemic effects	Regeneration of pancreatic $\beta$ cells	Inhibition of hepatic GóPase &increase in expression & translocation of GLUT4 in skeletal muscles.	Revival of pancreatic $\beta$ cells	AMPK mediated suppression of gluconeogenesis	I	Decreased gluconeogenesis	Inhibition of aldose reductase	Inhibition of $\alpha$ glucosidase	Inhibition of $\alpha$ glucosidase	1	Induction of insulin release from the remaining beta cells within the islets of Langerhans	Sulfonyl urea like mechanism	I
	Activity	50 μg/ml 8 44 μg/ml	$5.6\mu g/ml$	$102.23 \mu g/ml$	$6.53 \pm 0.16 \mu \text{g/ml}$	80 mg/kg	400 mg/kg	400 mg/kg	$1000\mathrm{mg}$	50–150 mg/kg >200 ng/ml	10 mg/kgbw	$0.70 \text{ mg/kgbw LD}_{50} = 1.4 \text{ mg/kg bw}$	$IC_{50} = 52 \mu g/ml$	$IC_{50} = 552.3 \mu g/ml$	$IC_{50} = 328.6 \mu g/ml$	100 mg/kg bw	500 mg/kg bw	100 mg/kg	700 mg/kg LD <sub>50</sub> >5000 mg/ kgbw
	Standard	Metformin 	Acarbose	Acarbose	Acarbose	Tolbutamide	Ι	Metformin	Metformin	AMPK inhibitor	Metformin	Insulin	Quercetin	Acarbose	Acarbose	Insulin	Glibenclamide	Glibenclamide	Metformin
	Extract type/ component	Aqueous Acetone	Aqueous	Vernoamyoside E	Luteolin	Aqueous	Ethanolic	80% ethanolic	Crude	30% ethanolic	$11\beta, 13$ -dihydrovernolide	Vernonioside E	Ethyl acetate	Ethyl acetate	Ethyl acetate	Methanolic	Ethanolic	Acetone	Aqueous
	Study type	In vitro In vitro	In vitro	In vitro	In vitro	In vivo	In vivo	In vivo	In vivo	In vivo In vitro	In vivo	In vivo	In vitro	In vitro	In vitro	In vivo	In vivo	In vivo	In vivo
	Sample type	Leaves	Roots	Leaves	Flowers	Leaves	Leaves	Leaves	Leaves	Leaves	Leaves	Leaves	Leaves	Callus	Callus	Seeds	Seeds	Leaves	Leaves
	Species							V. amygdalina							V anthelmentica			V. colorata	V. galamensis

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Adiukwu et al. [105] established a minimum effective oral therapeutic dose of 250 mg/kg for the ethyl acetate fraction of methanolic leaf extract, resulting in a significant 33.5% reduction in hyperglycemia within 4 hours in STZ-induced T2DM rats. Importantly, numerous toxicity studies have underscored the safety profile of VA, with doses ranging from 500-5000 mg/kg/day for 14 consecutive days revealing no adverse toxicological effects or clinical symptoms [94]. It is to be noted that most investigations have been conducted on rats or cell lines, and the substantial physiological and genetic differences between humans and animals cannot be disregarded. Moreover, human clinical trials are needed to validate these results, especially in nondiabetic individuals. Secondly, the short-term nature of these studies raises questions about the long-term effects of V. amygdalina extracts. Safety concerns persist since the phytochemical composition can vary based on plant age, geographical location, season, and soil composition. Furthermore, potential toxic effects on vital organs and the influence of pre-existing autoimmune disorders remain unexplored. Notably, pharmacokinetic investigations for V. amygdalina extracts have not been conducted, and compounds such as luteolin, luteolin-7-O-glucoside, chlorogenic acid, and its derivatives, which exhibit poor pharmacokinetic properties, warrant further research. Nanotechnological interventions, such as solid lipid nanoparticles, have been explored to address inherent pharmacokinetic limitations and enhance drug properties. In addition, synthetic chemists are designing derivatives with improved pharmacodynamic and pharmacokinetic profiles [95]. While some compounds such as vernoniosides and vernoamyosides may not adhere to Lipinski's rule [112], recent approvals of drugs exceeding these constraints offer hope for their druggability. Challenges in drug discovery include sourcing natural products in sufficient quantities and ensuring their stability against degradation, which is being addressed through innovative extraction techniques. In summary, V. amygdalina extracts and their phytochemicals hold promise for managing T2DM and its complications. However, further research is needed to bridge the gap between their potential and practical application as therapeutic agents.

#### 8. Conclusion

Diabetes, in which a human body is unable to deal with an upregulated sugar level, is a global health crisis. Diabetes not only leads to complications like cardiovascular disease, retinopathy, and kidney failure but also poses a significant risk to overall health and well-being. The prevalence of diabetes is on the rise, with a projected increase in cases, leading to substantial economic burdens on healthcare systems worldwide. Among others, major factors that contribute to the increasing incidence of diabetes include the destruction of pancreatic  $\beta$  cells, a poor lifestyle, increased physical activity, and others. Effective diabetes management is crucial to prevent chronic complications and reduce the associated healthcare costs. However, although various therapeutic approaches involving various classes of medications are available, there is a growing interest in the

development of natural product-based alternatives. Vernonia amygdalina (V. amygdalina), a plant found primarily in tropical Africa, has garnered attention for its potential medicinal properties. VA is rich in bioactive compounds like sesquiterpene lactones, steroid glycosides, and saponins, which have shown promise for various health benefits, including antimicrobial, anti-inflammatory, and blood sugar regulation. However, despite the potential of as a diabetes treatment, its use for clinical applications has not been achieved yet. There are various factors in research that need to be addressed. For instance, studies related to mechanisms of action, safety, and efficacy are lacking. In addition, the exploration of natural products like V. amygdalina as potential therapeutic options opens up exciting possibilities for the future of diabetes management. In summary, diabetes is a growing global concern with significant economic implications. Vernonia amygdalina, with its rich bioactive compounds, presents a promising avenue for research into alternative diabetes treatments. Addressing these research gaps in research could lead to more effective and safer options for diabetes management in the future.

#### **Conflicts of Interest**

The authors declare that they have no conflicts of interest.

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