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Phytochemical, Nutritional and Pharmacological Potentialities of *Amaranthus spinosus* Linn. : A review

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Abstract

Amaranthus spinosus has long been cultivated in tropical and subtropical areas of the world, especially in South Asia. It is well accepted by the people for its nutritional, pharmacological, phytochemical, and therapeutic functions in the human body. Tender stems, leaves, shoots, grains and sometimes the whole part of *A. spinosus* are eaten by humans or fed to farm animals, which contain carbohydrates, proteins, fats, fibers, vitamins, minerals and many other phytochemicals. This review aims to represent the nutritional and pharmacological activities of *A. spinosus*. To have a better understanding, we have discussed the nutritional status of *A. spinosus*, its available phytochemicals and their functional properties. Further, we demonstrated the potentiality of *A. spinosus* in various disease condition by discussing its functional activities, which includes antioxidant, antidiabetic, immuno-modulatory, hematological, gastrointestinal, anti-inflammatory, diuretic, antimicrobial, antimalarial, anti-ulcer, antipyretic, and antigenic activity. The availability of various important phytochemicals along with their functional properties make *Amaranthus spinosus* valuable for pharmaceuticals and nutraceuticals industry.

Abbreviations

µg	Microgram
µM	Micromolar
A.	<i>Amaranthus</i>
ABTS	2,2'-azinobis-3-ethylbenzothiazoline-6-sulfonic acid
ASAE	<i>Amaranthus spinosus</i> aqueous extract
ASE	50% ethanolic extract of <i>Amaranthus spinosus</i>
BHA	Butylated hydroxyanisole
COX-2	Cyclooxygenase-2
DEX	Dexamethasone
DPPH	2,2-diphenyl-1-picrylhydrazyl
EC50	Median effective concentration required to induce a 50% effect
FST	Forced swimming test
GF1	GATA-binding protein 1
Hb	Hemoglobin
HCl	Hydrochloric acid
IC50	Half-maximal inhibitory concentration
IEF	Isoelectric focusing
kDa	Kilodalton
LD ₅₀	Median lethal dose
MEAS	Methanolic extract of <i>Amaranthus spinosus</i>
mg	Milligram
mL	Milliliter
OECD	Organisation for Economic Co-operation and Development
PGE ₂	Prostaglandin E ₂
RBC	Red blood cells
SDS-PAGE	Sodium dodecyl sulfate polyacrylamide electrophoresis
SS	Swimming stress
TST	Tail suspension test
WBC	White blood cells

1. Introduction

Plant-based medicine is potentially the oldest form of medicine, some of which are not only used as medicine but also as nutritional victuals that showed the value of in-depth scientific evaluation. *Amaranthus spinosus* Linn. (Family: Amaranthaceae) is a most frequent consumable vegetable which has many medicinal aspects and is commonly known as Katanote in Bengali, Pigweed in English, and Kanatabhajii in Hindi (Alegbejo, 2014; Kawade et al., 2013). It is widely distributed from tropical, subtropical, and some temperate regions (Tanmoy et al., 2014). The genus *Amaranthus* has approximately 60 species, but 20 are important as weeds and others are widely used as vegetables, cereals and ornamental plants (Venskutonis & Kraujalis, 2013). Its tender stems, leaves, shoots and grains are eaten by humans which contain thousands of phytochemicals and prevents several chronic and degenerative disease. Phenolic compounds named quercetin, kaempferol glycosides and hydroxycinnamates are found in *A. spinosus* with the ranges from 305 mg/100 g to 329 mg/100 g (Stintzing et al., 2004). The stem of *A. spinosus* has anti-diabetic activity, and its effectiveness is proved for diabetes mellitus (Preethi, 2013), the root is used for the treatment of malaria (Bahekar & Kale, 2013) and the whole plant contains α-spinasterols octacosanoate & saponin which are effective as anti-inflammatory substances (Sengupta et al., 2012). It is also proved that consuming *A. spinosus* extract significantly increases prolactin level and helps to produce breast milk in postpartum mothers (Kuswaningrum et al., 2017). It also contains a red

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pigment which is utilized as a coloring agent in foods or medicine (Amabye, 2016).

This plant is largely utilized as vegetables because of its excellent source of antioxidant leaf pigments such as β -cyanin, β -xanthine, betalain and as a source of other pigments such as carotenoids, anthocyanin, chlorophylls. It is also a great source of antioxidant phytochemicals such as β -carotene, vitamin C, phenolics and flavonoids (Jiménez-Aguilar & Grusak, 2017; Sarker & Oba, 2019). In Malaysia, it is used as an expectorant as well as to enhance breathing in acute bronchitis (Alegbejo, 2014). The decoction of this plant is used for toothache, the ash is used as salt, boiled leaves and roots are eaten as laxative, the whole plant is fed to livestock as forage, and it also has numerous pharmacological uses such as astringent, antidote to snake poison, menorrhagia, gonorrhoea, internal bleeding, ulcerated mouths, nosebleeds, hepatic disorder, sudorific, eczema, galactagogue, febrifuge, diaphoretic, wounds, burns and dysentery (Alegbejo, 2014; Rahman & Gulshana, 2014; Kawade et al., 2013; Sarker & Oba, 2019). Along with all these potencies, *A. spinosus* also shows antimicrobial activity. This plant is widely found in roadsides, waste places and fields (Tanmoy et al., 2014).

To provide a descriptive overview of *A. spinosus*, nutritional and phytochemicals of this vegetables, evaluation methods, internal and external factors that influence its functional properties were summarized and reviewed.

2. Proximate composition

A. spinosus is an abundant source of dietary fibers. It also contains ash, moisture, crude fat and crude protein (Sarker & Oba, 2018, 2019). After analyzing 100 g of its leaves, it was found the carbohydrate content was 1.16 g, energy 27 kcal, moisture 91 g, protein 4 g, fat 0.6 g, fiber 2.48 g, ash 2.76 g (Figure 1A). In dry weight per 100g, the mineral content was 38.4 mg iron (Fe), 968.7 mg Calcium (Ca), 912.4 mg magnesium (Mg), 816.3 mg phosphorus (P), 6.8 mg manganese (Mn), 1.2 mg copper (Cu), 6.8 mg zinc (Zn) (Figure 1B) (Kawade et al., 2013). It has high nutritive value because of its elevated concentration of antioxidant compounds, fibers, proteins and amino acids, predominantly lysine (Tanmoy et al., 2014).

3. Phytochemistry

There are abounding active phytoconstituents in *A. spinosus*, which belongs to the group of flavonoids, alkaloids, amino acids, lipids, glycosides, phenolics, steroids, terpenoids, saponins, betalains, catechuic tannins, betaines such as trigonelline and glycine betaine, and carotenoids. Amaranthosides, amaricins and a coumaroyl adenosine together with stigmaterol glycoside are also seemed to be present in this plant (Ganjare & Raut, 2019).

Amaranthines, hydroxycinnamates, isoamaranthines, quercetins and kaempferols are mostly found in stems. Quercetin, α -xylofuranosyl uracil, β -D-ribofuranosyl adenine, β -sitosterol glucoside, 7-p-coumaroyl apigenin 5-O- β -D-glucopyranoside, amaranthoside, rutin, and amaricin are found in the whole plant. α -spinasterol and saponins are found in roots of the plant. Hectriacontane, oleanolic acid, D-glucose, D-glucuronic acid, aliphatic ester- α -spinasterol octacosanoate are generally found in leaves and less frequently in stems (Tanmoy et al., 2014). The main betalains in *A. spinosus* are found as quercetin, amaranthine, isoamaranthine, hydroxycinnamates and kaempferol glycosides. Coumaroyl flavone glycoside is identified from the *n*-butanol fraction of methanol extract. In the *A. spinosus* plant, the rutins and the quercetins are present as flavonoids. Rutin is identified in the whole plant powder. Separation and identification of amaranthoside, amaricin, and stigmaterol glycoside have become possible through the phytochemicals investigating method using *n*-butanol fraction of methanol extract of whole plant of *A. spinosus*. By using petroleum ether extract, α -spinasterol and hectriacontane are isolated. Some new compounds, such as aliphatic esters and saponins, specifically saponin I and saponin II were identified in *A. spinosus* (Sarker & Oba, 2019; Tanmoy et al., 2014).

The presence of all these compounds explicitly related to some medicinal properties of *A. spinosus*. Compounds like vitamin C, flavonoids and phenolics are phytochemical compounds which show most of the antioxidant activity in different plants including *A. spinosus*. It is evident that phenolic and flavonoid compounds may prevent or lessen the effect of particular cancer and cardiovascular diseases, along with the chronic and neurodegenerative types of diseases. *A. spinosus* has reported to provide antidiabetic, anti-inflammatory, anti-malarial, anti-fertility, anti-hyperlipidemic, spermatogenic and antimicrobial effects (Figure 2) (Ganjare & Raut, 2019; Kawade et al., 2013).

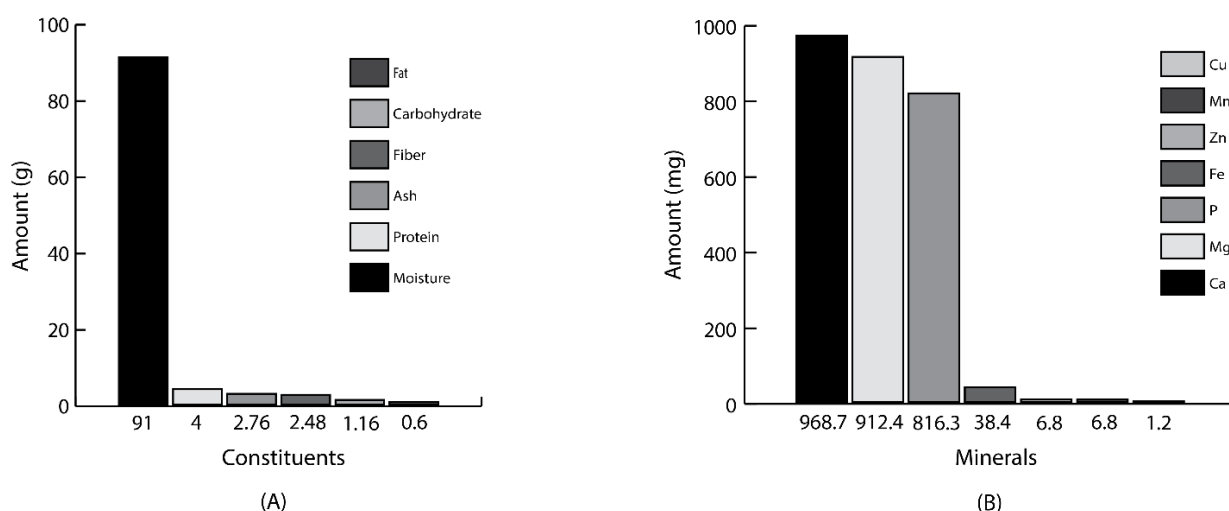


Figure 1 *A. spinosus* constituents and its mineral contents have been illustrated in the graphs, which are found in 100 gm weight. **(A)** Proximate composition or constituents of *A. spinosus* found in 100 gm of its leaves are demonstrated in this chart. This chart suggests that protein, ash, fiber and carbohydrates are quite good in *A. spinosus*, whereas fat content is the lowest in amount **(B)** Minerals found per 100 gm dry weight of *A. spinosus* has been demonstrated in the chart. It suggests that Ca, Mg and P are found in higher amount, where Fe, Zn, Mn and Cu are found in lower amount.

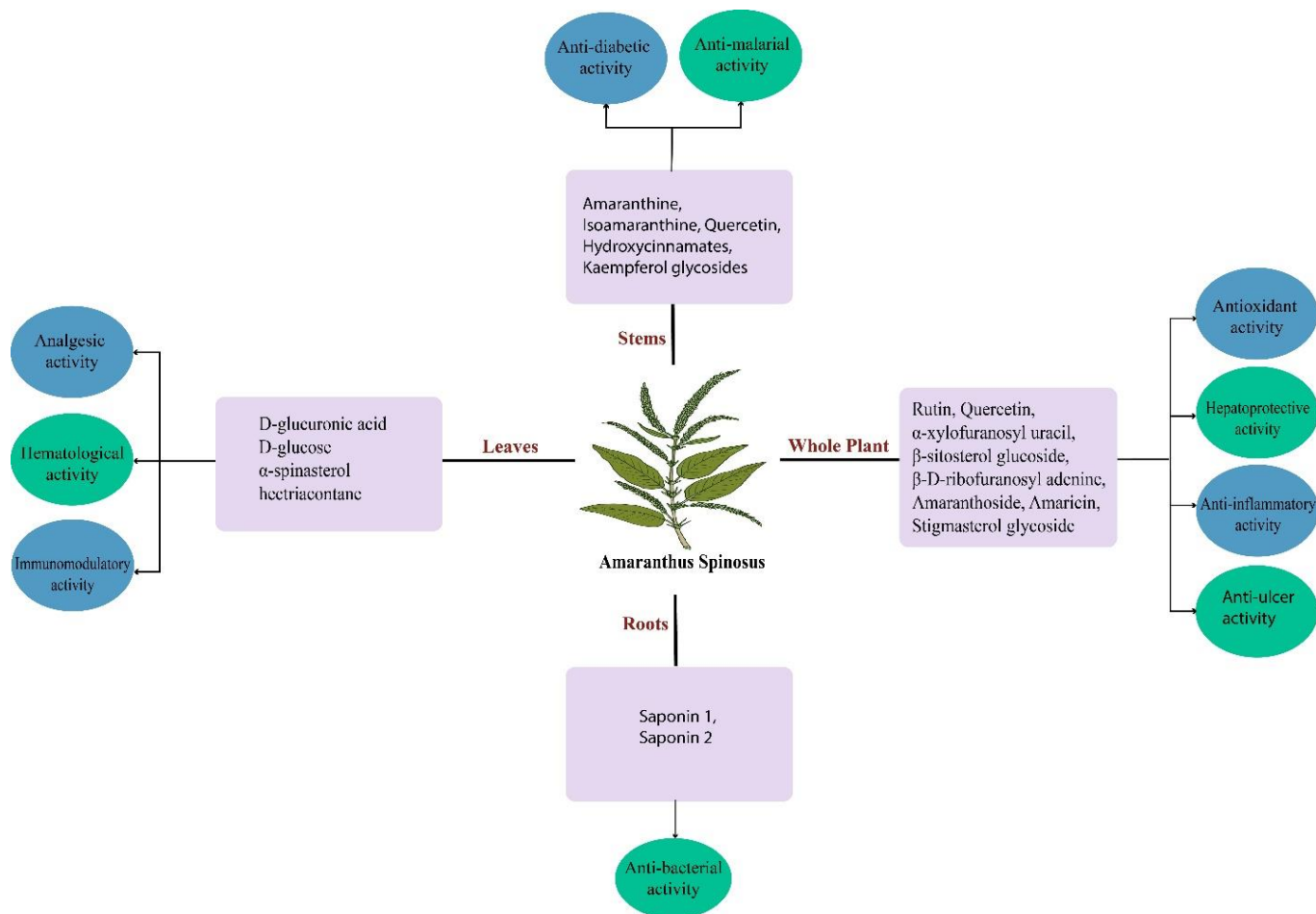


Figure 2 Phytochemicals found in whole plant, roots, leaves and stems of *Amaranthus spinosus* have been demonstrated in the rectangular boxes. Pharmacological activities of these phytochemicals have been indicated in the circles.

4. Phytochemical constituents

Most of the phytochemicals of *A. spinosus* are found in the matured leaves, because maximum metabolism in plants occurs in matured stage. To obtain the extract, leaves are air-dried at room temperature and allowed for grinding and finally extract is collected by solvent extraction. The analysis of *A. spinosus* to determine its phytochemical contents are done by standard procedures (Amabye, 2016; Maiyo et al., 2010). Numerous phytochemical and phenolic compounds are found in several

extracts of *Amaranthus spinosus*, still some other phytochemical compounds did not show their presence in the extract. The wide range of phytochemical constituents, and their presence or absence in different extracts can easily be recognized if analyzed properly (Table 1).

Amaranthus spinosus plant extract shows *in vitro* bioactivity because of the presence of innumerable phytochemical constituents (Maiyo et al., 2010). Moreover, biologically active phytochemical compounds largely contribute to the amelioration of public health (Table 2).

Table 1 Quantitative screening for phytochemical constituents of *Amaranthus spinosus*

Constituents	Presence (+) or Absence (-)					Content	Reference
	Methanol	Ethanol	Hexane	Chloroform	Aqueous		
Alkaloids	+	-	-	-	-	13.14 ± 0.86	(Amabye, 2016; Khanal et al., 2015)
Tannins	-	-	-	+	-	6.07 ± 0.93	(Amabye, 2016; Maiyo et al., 2010)
Flavonoids	+	+	+	+	+	1.70 ± 0.56	(Jiménez-Aguilar & Grusak, 2017; Khanal et al., 2015)
Saponins	+	+	-	-	-	53.0 ± 0.50	(Amabye, 2016; Khanal et al., 2015; Maiyo et al., 2010)

Table 2 Phytoconstituents and pharmacological activities of *Amaranthus spinosus*

Phytoconstituents	Plant part	Extract	Pharmacological activity	Reference
Rutin, Quercetin, 7-p-coumaroyl apigenin 4-O- β -D-glucopyranoside, α -xylofuranosyl uracil, β -sitosterol glucoside, β -D-ribofuranosyl adenine, Amaranthoside, Amaricin, Stigmasterol glycoside	Whole	Ethanol	Hepatoprotective activity	(Azhar-ul-Haq <i>et al.</i> , 2004; Suryavanshi <i>et al.</i> , 2007; Zeashan <i>et al.</i> , 2008; Zeashan <i>et al.</i> , 2009b)
	Whole	Petroleum ether, methanol, chloroform, aqueous	Antioxidant activity	(Kumar <i>et al.</i> , 2010b; Kumar <i>et al.</i> , 2010c)
	Whole	Ethanol	Anti-inflammatory activity	(Olajide <i>et al.</i> , 2004; Sengupta <i>et al.</i> , 2012)
	Whole	Aqueous	Anthelmintic activity	(Baral <i>et al.</i> , 2010; Kumar <i>et al.</i> , 2010a)
	Whole	Ethanol	Anti-diarrheal activity	(Hussain <i>et al.</i> , 2009)
D-glucuronic acid and D-glucose, α -spinasterol, hectriacontane	Whole	Ethanol	Anti-ulcer activity	(Hussain <i>et al.</i> , 2009; Mitra, 2013)
	Leaves	Ethanol	Analgesic activity	(Banerji, 1973; Taiab <i>et al.</i> , 2011)
	Leaves	Ethanol	Hematologic activity	(Akinloye & Oloredo, 2000; Olufemi <i>et al.</i> , 2003)
	Leaves	Aqueous	Immunomodulatory activity	(Alegbejo, 2014; B.-F. Lin <i>et al.</i> , 2005)
Amaranthine, Isoamaranthine, Quercetin, Hydroxycinnamates, Kaempferol glycosides	Leaves	Aqueous	Gastrointestinal activity	(Kumar <i>et al.</i> , 2008)
	Stems	Methanol	Anti-diabetic activity	(Sangameswaran & Jayakar, 2008; Stintzing <i>et al.</i> , 2004)
	Stems	Aqueous	Anti-malarial activity	(Hilou <i>et al.</i> , 2006)
Saponins	Roots	Hexane	Anti-bacterial activity	(Ahmad & Basha, 2006; Banerji, 1973, 1979, 1980; Vardhana, 2011)
	Roots	Ethyl acetate	Anti-bacterial activity	

antioxidant activity is also examined by determining the oxidation of linoleic acid (Amabye, 2016). Some of the methods include DPPH or 2,2-diphenyl-1-picrylhydrazyl scavenging; superoxide anion radical scavenging; nitric oxide scavenging;

hydroxyl free radical scavenging; and ABTS or 2,2'-azinobis-3-ethyl-benzothiazoline-6-sulfonic acid radical scavenging assays (Kumar *et al.*, 2010c).

Table 3 The antioxidant activity of *Amaranthus spinosus* illustrated by several methods

Evaluation method	Standard antioxidant	Extracts used	Conc. used	Preventive effect	Reference
Non-enzymatic haemoglycosylation assay	α -tocopherol (vitamin E)	Petroleum ether	0.5 mg/mL	13.1 %	(Kumar <i>et al.</i> , 2010b)
		Chloroform	1 mg/mL	16.4 %	
			0.5 mg/mL	5.7 %	
		Methanol	1 mg/mL	12 %	
			0.5 mg/mL	36.91 %	
DPPH radical scavenging assay	Ascorbic acid (vitamin C)	Methanol	1 mg/mL	56.07 %	(Bulbul <i>et al.</i> , 2011)
			0.5 mg/mL	22.2 %	
			1 mg/mL	31.01 %	
ABTS radical scavenging assay	Ascorbic acid (vitamin C)	Methanol	25-250 μ g/mL	147.50 \pm 2.61 μ g/mL IC ₅₀ value	(Kumar <i>et al.</i> , 2010c)
Hydroxyl radical scavenging method	Butylated hydroxyanisole (BHA)	Ethanol (50%)	400 μ g/mL	140-145 μ g/mL IC ₅₀ value	(Zeashan <i>et al.</i> , 2009b)

The antidiabetic activity

A. spinosus has alpha amylase enzyme, which is a potential compound associated with carbohydrate digestion and glycemic balance. Antidiabetic activity was studied by introducing methanolic extract of *A. spinosus* stem in diabetic rats (Jhade *et al.*, 2009). The antidiabetic potential of methanolic extract of *A. spinosus* was diagnosed through *in vitro* alpha amylase inhibition

by a compound named CNPG-3 or 2-chloro-4-nitrophenyl- α -D-maltotrioside, in alloxan-induced diabetic rats. The study manifested significant restraining capacity of glucose level on a 15-day study. *A. spinosus* leaf extract also reduces hyperglycemia by abating pancreatic cells damage (Figure 4) and oxidative stress in streptozotocin-nicotinamide induced diabetes albino rats (Mishra *et al.*, 2012; Tanmoy *et al.*, 2014).

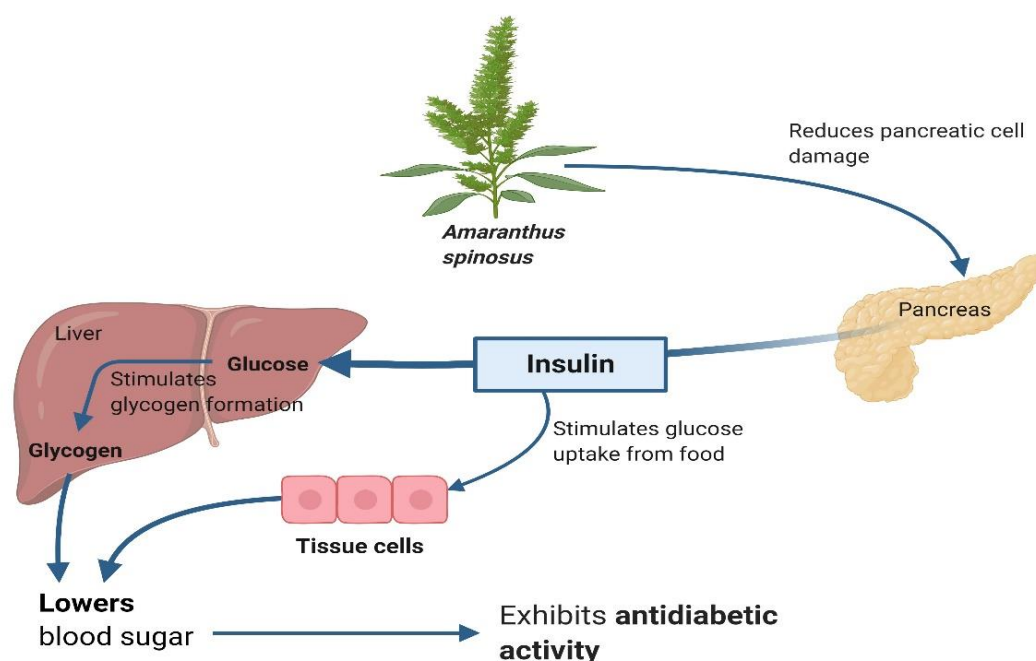


Figure 4 Active phytochemicals of *Amaranthus spinosus* can reduce pancreatic cell damages. Insulin released from pancreatic cells stimulate both glucose uptake and glycogen formation, therefore, manifests antidiabetic activity by lowering the blood sugar level.

The immuno-modulatory activities

A. spinosus has been demonstrated to possess immunological effects. It was determined by investigating the stimulatory effects of aqueous *A. spinosus* extract on spleen cells of female albino rats. The result showed that the extracts affiliate in stimulation of more B lymphocytes without increasing T lymphocytes level, and the immuno-stimulating effects of aqueous extract help B lymphocyte activation and proliferation of T-cell *in vitro*. Several studies substantiated that the extract of this plant can significantly escalate the splenocyte growth. The results also proved immuno-modulatory activity via direct *in vitro* B lymphocyte activation. A new immuno-stimulatory protein (GF1) having molecular mass of 313 kDa, which is thought to be a glycoprotein and heat labile, have 309 times higher immunostimulatory activity than that of water extract. This compound is highly potential for immune pharmacological use (Ganjare & Raut, 2019; Gotyal *et al.*, 2016).

Immuno-modulatory effects were also discovered using dexamethasone (DEX)-induced apoptosis in murine primary splenocytes and wild *A. spinosus* water extract. The result determined that *A. spinosus* water extract is capable of inhibiting the spontaneous, as well as the DEX induced apoptosis of splenocyte cells (Lin *et al.*, 2008). Cell-mediated immune response by delayed type of hypersensitivity reaction to sheep RBC, and humoral immune response measured by hemagglutination antibody titre also corroborated the immuno-modulatory activity. The result showed both immuno-modulatory and immunosuppressant activity due to presence of various glycosides, steroids and other phytochemicals. Aqueous and alcoholic extracts showed more immuno-modulatory effect, and petroleum ether extract showed immunosuppressant effect (Tatiya *et al.*, 2007).

The hematological activity

There has been a study on the effect of aqueous extract of leaves of *A. spinosus* on hematological parameters along with blood coagulation time in rat model, which showed little changes in the hematological activity and several enzymes level such as glutamate pyruvate transaminase, alkaline phosphatase and serum glutamate oxaloacetate transaminase (Akinloye & Olorede, 2000).

In another study, *A. spinosus* leaf extract was fed to growing pigs and administered to determine its effects on packed cell volume, white blood cell, red blood cell, and hemoglobin concentration. The result showed that there has been temporary decrease in levels of packed cell volumes, white blood cell, red blood cell and hemoglobin (Olufemi *et al.*, 2003).

Change in hematocellular constituents of albino rats using *A. spinosus* methanolic extract has been also been diagnosed, which showed that WBC, RBC and Hb level was highly restored after using *A. spinosus* methanolic extract (Gul *et al.*, 2011).

The hepatoprotective activity

A. spinosus extract (whole) in 50% ethanol can be assessed against carbon tetrachloride induced hepatic damaged rats. The *A. spinosus* whole plant in 50% ethanol extract was fed to the carbon tetrachloride induced rats for 14 days and introduced oral doses of 100, 200 and 400 mg/kg in an experiment. The result showed substantive escalation of different enzymatic levels, such as serum glutamate pyruvate transaminase, glutamate oxaloacetate transaminase, and serum alkaline phosphatase; also, the bilirubin level came back to normal significantly. Conclusively, hepatoprotective activity is significant at higher dose of *A. spinosus* ethanol (50%) extract against CCl₄ inducing hepato-toxicity, because 400 mg/kg dose

showed greater hepatoprotective activity than other doses (Zeashan *et al.*, 2008).

In another study, 50% ethanolic extract of *A. spinosus* whole plant is evaluated for hepatoprotective and *in vitro* antioxidant activity. Standard gallic acid curve (0-1.0 mg/mL) and an antioxidant naming butylated hydroxy anisole showed both the total phenolic compounds level and the hepatotoxicity reduction capacity of ASE respectively. The result had shown the hepatotoxicity reducing capability of ASE is 2.26 times of BHA and total polyphenolics expressed as gallic acid equivalent was 336 ± 14.3 mg/g, which proved that ASE possesses significant hepatoprotective activity (Zeashan *et al.*, 2009b).

The gastrointestinal activity

To determine the effect of *A. spinosus* in gastrointestinal tract, aqueous extract of *A. spinosus* was evaluated in mice using charcoal meal method. In the experiment, first group served as the basis of control, second group was made the standard, and the remaining 3-5 groups were fed *A. spinosus* aqueous extract at a dose of 50, 100 and 200 mg/kg weight of the mice respectively. Result of the experiment showed significant gastrointestinal motility at 100 mg/kg dose (Kumar *et al.*, 2008).

Another *in-vivo* experiment in mice using crude extract of *A. spinosus* showed the laxative activity, which is partially intervened through the cholinergic action (Chaudhary *et al.*, 2012).

The anti-inflammatory activity

The anti-inflammatory activity of *A. spinosus* has been evaluated by an experiment in which the methanolic extract of *A. spinosus* was evaluated in various animal models. The extract inhibited the carrageenan inducing mice paw edema at a dose of 25-100 mg/kg and helped in inhibition of acetic acid inducing increased vascular permeability. Though the 50 and 100 mg/kg extract dose produced gastric erosion in rats along with indomethacin, 25 mg/kg extract along with tween-80 (polysorbate-80) did not show this effect in macroscopic evaluation. However, several experiments corroborated the anti-inflammatory properties of *A. spinosus* (Baral *et al.*, 2010; Olajide *et al.*, 2004).

The diuretic activity

To examine the diuretic activity of *A. spinosus* aqueous extract (ASAE) in rats, various doses of ASAE (200, 500, 1000, 1500 mg/kg), thiazide (10 mg/kg) and vehicle were fed to rats orally and their urine was collected and analyzed after 24 hours. The result showed that ASAE increased the concentrations of Na⁺, K⁺, Cl⁻ and caused alkalization of urine. The report of this test proved that the *A. spinosus* may act as a thiazide-like diuretic, occupying the carbonic anhydrase inhibiting property (Amuthan *et al.*, 2012).

The antidepressant activity

By using Forced swimming test and Tail suspension test patterns, the anti-depressant activity of the methanolic extract of *A. spinosus* (MEAS) was examined, where reference standards were Escitalopram and Imipramine. The result of this experiment showed that the MEAS cause significant diminution of immobility in FST and TST comparing with Escitalopram and Imipramine (Kumar *et al.*, 2014).

The Antimicrobial Activity

The *A. spinosus* has various pharmacologically active compounds, which shows antimicrobial activities in the disc

diffusion assay. Even though the trend for anti-fungal activity is same as of the anti-bacterial activity, the potency towards fungal strain is not as effective as for bacterial strains (Amabye, 2016). Terpenoids show antimicrobial activity against bacterium, fungus, virus and protozoa, and it is used to control *Listeria monocytogenes*. These terpenoid compounds demonstrate antimicrobial activity by lipophilic membrane disruption mechanism. If the hydrophilic behavior of ent-kaurene diterpenoid compounds can be increased by addition of methyl group, this will reduce the antimicrobial activity of these compounds. Flavonoids generally are known to show response against microbial infections. Flavonoids form complexes with

extra-cellular and soluble proteins, as well as with bacterial cells *in vivo*. This is why flavonoids are also effective as antimicrobial agents (Maiyo *et al.*, 2010).

Usage of dichloromethane extract of *A. spinosus* moderately shows antiprotozoal activity, especially on *Blastocystis hominis* when 2 mg/mL dose is introduced (Kawade *et al.*, 2013). Ethanol extracts of *A. spinosus* leaves show anthelmintic activity if fed to growing pigs. The result shows that mean egg count of helminths, predominantly *Ascaris suum*, is remarkably decreased due to ethanol extract of *A. spinosus* leaves (Assiak *et al.*, 2002).

Table 4 Antimicrobial activities of *A. spinosus* methanol leaf extract

Organism types	Test Organisms	Extract	Zone of growth inhibition (mm)	Reference
Bacteria	<i>Staphylococcus aureus</i>	Methanol 100%	24.0	(Amabye, 2016)
Bacteria	<i>Escherichia coli</i>	Methanol	11.6 ± 0.2	(Barku <i>et al.</i> , 2013)
Bacteria	<i>Bacillus subtilis</i>	Methanol	8.0 ± 0.26	(Cherian & Sheela, 2016)
Bacteria	<i>Salmonella typhi</i>	Hexane 100%	13.0 ± 1.2	(Maiyo <i>et al.</i> , 2010)
Bacteria	<i>Klebsiella sp.</i>	Distilled water	7.0	(Sheeba <i>et al.</i> , 2012)
Fungi	<i>Fussarium solani</i>	Methanol 100%	17.0	(Amabye, 2016)
Fungi	<i>Aspergillus flavus</i>	Methanol	6 ± 0.76	(Cherian & Sheela, 2016)

The antimalarial activity

Screening of *A. spinosus* shows impressive antimalarial activity in mice when evaluated by suppressive antimalarial assay for 4 days. A study on *Plasmodium berghei berghei* parasite strain, which was inoculated in mice, had been inhibited greatly by *A. spinosus* at elevated doses of the extract produced from the plant. With 100 mg/kg dose introduction, the inhibition percentage of parasitemia is low, but increased doses (300 mg/kg to 900 mg/kg) had given better inhibition percentage. Though the plant has lower antimalarial activity compared to malarial medicine chloroquine, it's parasitemia inhibition percentage can also be increased by introducing relatively higher doses (1000 mg/kg or more) (Hilou *et al.*, 2006).

The anti-ulcer activity

In an interesting study on antiulcerogenic activity of *A. spinosus*, it was found out that *A. spinosus* possesses anti peptic ulcer activity when powdered leaves of *A. spinosus* is fed to gastric and duodenal ulcerated albino rats. The result shows that *A. spinosus* leaves can immensely protect ethanol inducing peptic or gastric ulcers, and cysteamine inducing gastric ulcers (duodenal ulcers). Though the anti-ulcer activity of *A. spinosus* showed less effect than a peptic ulcer drug, omeprazole, still it's role is crucial because of no known side effects or prolonged effects (Debiprasad *et al.*, 2013).

In another study of anti-ulcer activity, 50% ethanolic extract of *A. spinosus* (ASE) whole plant was evaluated in rats. Induction of acute gastric mucosal lesions were monitored using three different assay models in the investigation. The result proved that ASE can significantly protect both ethanol induced and aspirin induced ulcer (Hussain *et al.*, 2009). One of the important factors in ulcerogenesis is lipid peroxidation, and studies have substantiated that ASE can control this type of lipid peroxidation (Hussain *et al.*, 2009; Sairam *et al.*, 2002).

Search for new drugs of gastric ulcer led to another experiment on *A. spinosus* for its antiulcer activity. Result of the study showed that leaves of *A. spinosus* can prevent the loss of gastric protein, and the lipid peroxidation due to aspirin induced ulceration. Simultaneously, the leaves of this plant had increased gastric mucin, and showed cytoprotective effect against gastritis (Mitra *et al.*, 2014).

More studies showed that roots and stems along with leaves of *A. spinosus* exhibits antiulcer activity in albino rats. The effect is prevalent against ethanol, HCl, Swimming stress (SS), pyloric ligation, and indomethacin inducing peptic or gastric ulcer. However, the result showed that utilization of powdered roots of *A. spinosus* with one of the ulcerogenic medicine can bring about more ulcerative protection in ethanol, HCl, SS, pyloric ligation, and indomethacin induced gastric ulcer than utilization of stems and leaves of *A. spinosus* (Mitra, 2013).

The antipyretic and antinociceptive activity

Antipyretic drugs can suppress the expression of COX-2 so that the higher temperature of the body is reduced through inhibition of PGE₂ biosynthesis, whereas natural COX-2 inhibitors have fewer side effects (Luo *et al.*, 2005). Methanol extract of *A. spinosus* shows antipyretic activity without inhibition of COX-2 expression when introduced to rats with yeast-induced elevated level of body temperature (Lakshman & Jayaveera, 2011). The antipyretic effect of *A. spinosus* is quite significant, and is similar to the paracetamol group when the introduced dose is 400 mg/kg (Bagepalli *et al.*, 2011; Kumar *et al.*, 2010c). In a study, 50% ethanolic extract of *A. spinosus* (ASE) had shown antinociceptive activity when introduced to male swiss albino rats by some methods, including acetic acid test, formalin test, tail suspension test and hot plate test. Result from the study had shown that ASE possesses the central as well as the peripheral antinociceptive activity, and significantly blocks pain in the first phase at higher dose of the extract (400 mg/kg). In the second

phase, ASE had blocked pain from inflammation with introduction of all the doses (Zeashan *et al.*, 2009a).

The antigenic and allergenic activity

Pollen antigen standardization can assist in immunotherapy and diagnosis of allergenicity. *A. spinosus* is an essential aero-allergen and it has significance in type 1 hypersensitivity disorders. SDS-PAGE and IEF Analysis of five *A. spinosus* pollen samples showed that seven protein fractions have IgE binding capacities and nine proteins have allergenic properties (Singh & Dahiya, 2002).

6. Toxicities

The toxicity of *A. spinosus* was detected by scientists using OECD guidelines. By following this guideline, lethal dose 50% (LD50) was determined in a lab experiment using albino rats. The dose was 5 mg/kg, 50 mg/kg, 200 mg/kg, 300 mg/kg (later the dose was 250 mg/kg and 300 mg/kg for further study) with sufficient amount of water orally in a single dose. The result showed *A. spinosus* extract did not cause any toxicity in the animals up to 2000 mg/kg (Jhade *et al.*, 2011; Mishra *et al.*, 2012).

The genotoxicity of *A. spinosus* leaf extract was determined using meristematic root cells of *Allium cepa* and the antigenotoxic effects was determined by evaluating against H₂O₂-induced genetic damage in *Allium cepa*. The report of this experiment manifested that higher dose of *A. spinosus* extract cause mitodepressive and clastrogenic effects, and lower dose of the *A. spinosus* extract reverts the clastrogenicity caused by H₂O₂. This clastrogenicity, which is induced by the *A. spinosus* extract demonstrates its genotoxicity. These results determined that *A. spinosus* extract has genotoxic as well as the antigenotoxic property (Prajitha & Thoppil, 2016).

7. Conclusion

A. spinosus is abundant in phytochemicals. This review of *A. spinosus* shows that it holds nutritional potential along with various pharmacological properties. Leaf extract of this plant shows antimicrobial activities, especially against food-borne pathogens, and some fungus. Though some detailed studies have been done on this plant, it has not been developed as a drug yet. More exhaustive data on *A. spinosus* is not available till now even though the study on this potential plant was started in 1970s through analyzing its grain composition. Data on extraction of super-critical fluid and fractionation, high pressure and extrusion processing techniques are unavailable. Yet the screening or analysis within last 3 decades revealed so many information of this plant, such as its application for high quality baked goods production, edible films, functional ingredients etc. More extensive research and analysis is required for the cataloging, documentation and commercialization of this plant, considering its potentialities.

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Declaration of interest

All authors declare that there is no conflict of interest.

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