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A Phytopharmacological Review on Vigna Species

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ABSTRACT: Plants are the almost exclusive source of drugs for a majority of the world's population. Therefore, itremains a challenge for scientists to provide efficient, safe and cheap medications, especially for rural areas,with the available resources at the nearest. In the present review an attempt has been made to gather theinformation regarding taxanomical characters, various species, climatic conditions, various folklore usages,and some of phytochemical, pharmacological, antimicrobial studies conducted so far on these vignagenusplants which belongs to leguminosae family. This review reveals that legumes can be used as antioxidant intreating variousailmentslike liver diseases,cancer, diabetes,kidney disorders andfor curingvariousmicrobial infections. These legumes acts as good nutritive as well as cures various ailments hence theselegumes comes under nutraceuticals. With this review we conclude that there is a need of conducting furtherstudies for isolation of individual constituents, to find our theirpharmacognostic characters, and going fordifferent formulations and screening of various activities on these genus plants.The flavonoids wereisolated by preparative layer chromatography. The HPTLC revealed thepresence of vitexin andisovitexinindifferent fractions ethylacetate, n-butanol.

KEYWORDS: vignaspecies, chemical constituents, TLC, HPTLC, pharmacologicalactivitie

I. INTRODUCTION

The name of the Vigna genus is derived from an Italian botanist of the 17th century Dominico Vigna. It comprises around 150 species. It is closely related to Phaseolus. Most commonly cultivated crops of the Vigna genus are cowpea (Vigna unguiculata), greengram (Vigna radiata), moth bean (Vigna aconitifolia) and zombie pea (Vigna vexillata) etc. Vigna which is widely cultivated and used as neutraceuticals. They grow in varied climatic zones, in high temperatures, low rainfall and poor soils with low input in form of fertiliser and irrigation that make them valuable crop plants. As Vigna is an important genus that fulfils the food demand, useful in cosmetics and medicines, there is scope to enhance its productivity resource conservation, optimum use of rainwater, bridging the yield gaps and innovations in technology transfer and up scaling. Which is cultivated and used as a neutraceutical in all over world. In the traditional system of medicine this genus is mainly used in the treatment of liver disorders, ulcers, to decrease the weight, and also used in hormonal balance. Some wild species of Vigna are well known for various uses such as soil enrichment material, human food, medicinal plants, soil erosion-preventing materials and animal feed. The seeds of nearly all species of Vigna have antioxidant properties and are used to treat different diseases like rheumatism, liver diseases, diabetes, coughs, cancer, fevers, microbial infections, kidney disorders, paralysis, hormonal disorders and for weight reduction. Therefore further research is required to find out pharmacognostic importance of individual components of these species.

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Different species of vigna are

Vigna radiata -: Mung Bean, Green Gram, Golden Gram, Mash Bean, Green Soy

Vigna unguiculata -: Cowpea, Crowder Pea, Southern Pea Vigna

aconitifolia -: Moth Bean, Mat Bean, Turkish Gram

Vigna vexillata

(L.) A.Rich. -: Zombi Pea

Vigna mungo -: Urad Bean, Black Matpe Bean, Black Gram, White Lentil, "black lentil"

Table1. Different Vigna species and their medicinal importance

Sr. No.	*Vigna species*	Medicinal value
1.	*V. radiata*	Light diet, fever, dysentery, cooling and astringent, vertigo, beri-beri, polyneuritis granuloma, poultice (treatment of scabies, psoriasis and other skin ailments), heat stroke, antidiahrroeal, antioxidant, antimicrobial, anti-inflammatory, antitumor, antidiabetic, antihypertensive, gastritis, uraemia, hypercholesterolemia, coronary heart disease, lipid metabolism accommodation, hepatitis, toxicosis, cholera, corneal opacity and macula.
2.	*V. unguiculata*	Neuritis, insomnia, weakness of memory, indigestion, dyspepsia, sensation of pins and needles in limbs, periodic palpitation, congestive cardiac failure, stomatitis, corneal ulcers, coleic diseases, kwasiorkar, marasmus, hyperacidity, nausea and vomiting, malnutrition and micronutrient deficiencies.
3.	*V. aconitifolia*	Antibacterial, antifungal, antiviral, anticancer, antioxidant and anti-inflammatory
4.	*V. vexillata*	Joint disorders, arthritis, swelling in joints, antihypertensive, hypoglycemic, Cholesterol reduction, Antioxidants, Antibacterial, anti cancer bioactivities
5.	*V. mungo*	Liver disorders, rheumatism, infection of nervous system, aching bones, dropsy, cephalgia. anti-hypertensive and antidiabetic, hypolidimic action.

Table 2. Phytochemicals reported from Vigna species

Sr. No.	*Vinga species*	Phytochemical Reported
1.	*V. radiata*	Steroids, triterpenoids, glycosides, flavonoids, alkaloids, polyphenols, tannins, saponins,daidzin, daizein, ononin, formononetin, isoformononetin, 6,7,4′-trihydroxyisoflavone,6,7,4′-trihydroxyisoflavone, genistin, sissotrin, genistein, prunetin, vitexin,isovitexin, rutin, quercetin-3-glucoside, quercetin, kaempferol, myricetin, rhamnetin,kaempferitrin, kaempferol-3- rutinoside, naringenin-7-glucoside, naringin, neohesperidin, hesperetineriodictyol-7-glucoside, eriodictyol, naringenin, rhododendrin,scopoletin, pomiferin, delphinidin, phloretin, coumestrol, osajin,p-hydroxybenzoic, protocatechuic, syringic, gallic acid, vanillic acid, gentisic acid, shikimicacid, p-coumaric, cinnamic acid, caffeic acid, ferulic, chlorogenic acid.
2.	*V. unguiculata*	Alkaloids, saponin, flavonoids, tannins, glycosides, sterols, carbohydrates, polyphenols, reducing sugar, fats, oil, proteins.
3.	*V. aconitifolia*	Amino acids, carbohydrates, glycosides, flavonoids, tannins, phenolic compounds, saponins, ascorbic acid.
4.	*V. vexillata*	Sterol, isoflavone.
5.	*V. mungo*	Glycosides, tannins, alkaloids, flavonoids, saponins, terpenoids, quinone, sterols,ethylbenzene, pentane, 1,1-diethoxy-, hexanoic acid, propane, octanoicacid, decanoic acid, dodecanoic acid, 3-hydroxy-, (1,1'- bicyclopropyl)-2-octanoic acid, 2'-hexyl-, methyl ester, desulphosinigrin, 3-O-methyl-d-glucose, phthalic acid, butyl isohexylester, ethanol, 2-(9- octadecenyloxy)-,(Z)-, n-hexadecanoic acid, hexadecanoic acid, 9,12- octadecadienoyl chloride, (Z,Z)-, oleic Acid, octadecanoic acid, ethyl ester, genistein, 2'-hydroxy-genistein, 2'-hydroxydaidzein, kievitone, cyclokievitone

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II. THIN LAYER CHROMATOGRAPHY

Vigna radiata

Thin layer chromatography examination of extracts obtained from seeds and sprouts of Vigna radiata plant

In this qualitative identification using a readymade aluminum plates of silica gel GF254 and using 3 different developing solvent systems for detection the plant flavonoid in fractions of ethyl acetate and n-butanol for seeds and sprouts. Comparing with flavonoids standards and detection under UV 254, 366 nm, and they are listed in the Tables (3 and 4):

No.	Composition
SK4	Ethyl acetate: chloroform: formicacid: water (8:1:1:1)
SK5	Ethyl acetate: acetic acid: formic acid: water (84:4:4:10)
Sk6	Ethyl acetate: formic acid: acetic acid: water (100:11:11:27)

Table 3. Developing solvent systems were used in the identification of expected Flavonoids in above fraction.

Table 4. Thin layer chromatography for separated spots:

Mobile phase	Standard name	Rf value of standard	Rf value of matched flavonoid	Compound name
Sk4	Isovitexin	0.33	0.32	R1
	Vitexin	0.51	0.5	R2
Sk5	Isovitexin	0.3	0.29	R1
	Vitexin	O.41	0.41	R2
Sk6	Isovitexin	0.62	0.62	R1
	Vitexin	0.73	0.74	R2

III. HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY

Identification of vitexin and isovitexin by HPTLC

Ethyl acetate and n-butanol before and after hydrolysis fractions for the seeds, and ethyl acetate and n-butanol fractions of sprouts were analyzed also for its flavonoids, coumarin and phenolic acid contents utilizing HPTLC (Eike Reich/CAMAG Laboratory, Switzerland), using silica gel GF254 plates developed in a mobile phase composed of ethyl acetate: formic acid:acetic acid: water (84:4:4:10 V/V) examined at 254 and 366 nm wavelength.

International Journal of Pharmaceutical Research and Applications
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Isolation and purification of vitexin and isovitexin by preparative thin layer chromatography (PTLC)

Vitexin and isovitexin were isolated fraction from n-butanol fraction of seeds by preparative layer chromatography (PLC) using (1 mm) thickness plate (20 x 20 cm) and 100 ml of mobile phase (ethyl acetate: acetic acid: Formic acid: water) in the volume ratio of (84: 4: 4: 10) V/ V.

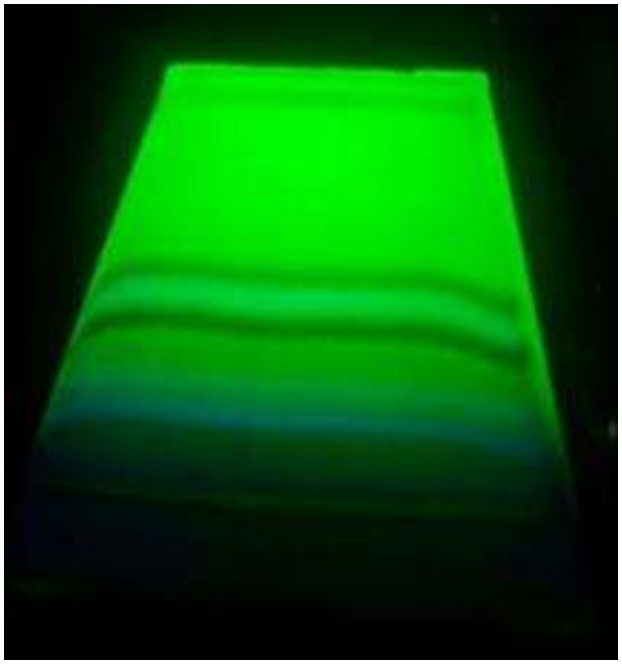

Figure 1. Preparative layer chromatography chromatogram of flavonoids isolation observed at254 wavelength

The purity of each isolated compound was examined using the analytical thin layer chromatography until obtaining single spot on TLC plate and detection was recorded under UV light 254 and 366 nm. All these steps were done at College of Pharmacy / University of Baghdad. The isolated compound was recrystallized using hot methanol and compared with vitexin and isovitexin standards by different identification methods, including:

1. Thin layer chromatography (TLC) using the best mobile phase system. Ethyl acetate: Formic acid: acetic acid: water (84: 4: 4: 10)
2. Fourier transforms infrared Spectroscopy (FTIR) in KBr disk.
3. High performance thin layer chromatography HPTLC using the previously mobile phase.

IV. FOURIER TRANSFORMS INFRARED (FT–IR) SPECTRUM

FT – IR spectroscopy is most commonly used in phytochemical studies as finger printing device. The FT – IR spectra of separated compounds was detected in the College of Sciences Al-Mustansirya University using SHIMADZU device. IR spectra of isolated compounds and the characteristic IR absorption bands of isolated compounds are below.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 932-941 www.ijprajournal.com ISSN: 2249-7781

Table5. Characteristic FI–IR absorption bands in(cm^{-1}) of isolated compounds:

Functional group	Group frequency wave number in cm – 1		Assignment
	Measured for isolated compound vitexin	Measured for isolated compound isovitexin	
O – H	3381 -3231	3433-3000	O-H stretching of phenol
C = C – H	3210	3285	C – H Stretching of aromatic ring
C – H	2910	2928 ,2843	Asymmetric and symmetric stretching of CH2
C = O	1651	1641	C = O stretching conjugation and H – bonding
C = C	1614,1570,1506	1608, 1568,1516	C = C stretching aromatic ring
C – H	1419	1444	C – H bending of CH2.
O-H	1384	1363	O-H bending of phenol
C-O-C	1251	1238	C-O-C stretching of ether
CH	1091 – 1066	1084 – 1072	C – H bending of aromatic (in plane)
CH	987 , 879, 758	916 , 887 , 775 ,698	C-H bending of aromatic (out of plane)

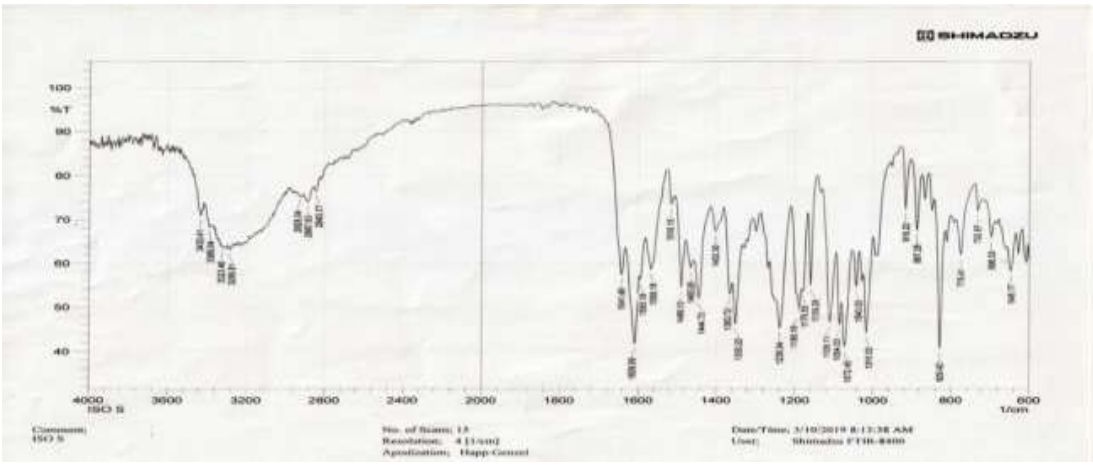

Figure2.Spetrum of isolated compound R1

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 932-941 www.ijprajournal.com ISSN: 2249-7781

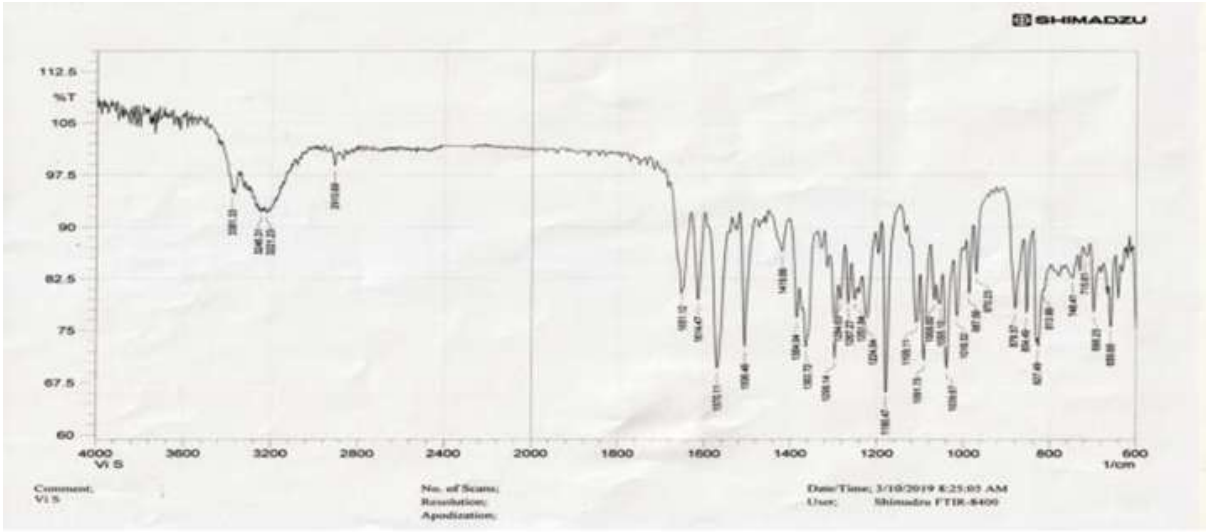

Figure3.IR spectrum of isolated compound R2

V. IDENTIFICATION OF PHENOLIC COMPOUNDS BY HPLC-DAD/MS-MS

V. unguiculata

Table 6. Phenolic compounds identified in Crude Methanol Extracts (CME) of cowpea CC and CT based on massspectrometry (MS) and fragments (MS^2).

Amount	Substance	$[M-H]^-$ *m/z*	MS^2 *m/z*
Cowpea CT	Gallic acid	[169]	[169] 125
	Protocatechuic acid	[153]	[153] 109
Cowpea CC	Quercetin	[301]	[301] 273, 193, 179, 151
Fraction 14	*p*-Coumaric acid	[163]	[163] 119
Fraction 17	Quercetin	[301]	[301] 273 257 179 151
	Protocatechuic acid	[153]	[153] 109
	Kaempferol	[285]	[285] 257 217 199 175 151

The chromatogram (Figure 2a) showed many overlapping peaks, however, it was possible to identify the flavonol quercetin that was identified based on the peak with retention time of 16.51 min, whereas the mass spectrum corresponding to this peak exhibited the mass of its deprotonated molecular ion at m/z 301 [M - H] - , in addition to the fragments at m/z 273, m/z 193, m/z 179 and m/z 151 , and then it could be confirmed the presence of flavonol quercetin.

The chromatogram of the cowpea CT (CME) (Figure 2a) was quite similar to that shown in Figure 2b, indicating that both extracts had similarity in chemical compounds, in which gallic acid and PCA were identified. The identification (Figure 3) of gallic acid was performed based on the chromatogram peak with retention time of 4.26 min, whereas the mass spectrum corresponding to

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 932-941 www.ijprajournal.com ISSN: 2249-7781

this peak resulted in a corresponding deprotonated molecular ion at m/z 169 [M - H] - mass and at m/z 125 fragment. For PCA, it could be observed in the chromatogram a peak with retention time of 4.33 min, which exhibited a deprotonated molecular ion peak at m/z 153 [M - H] - and the fragment at m/z 109. These data compared to the patterns could confirm the occurrence of these phenolic acids.

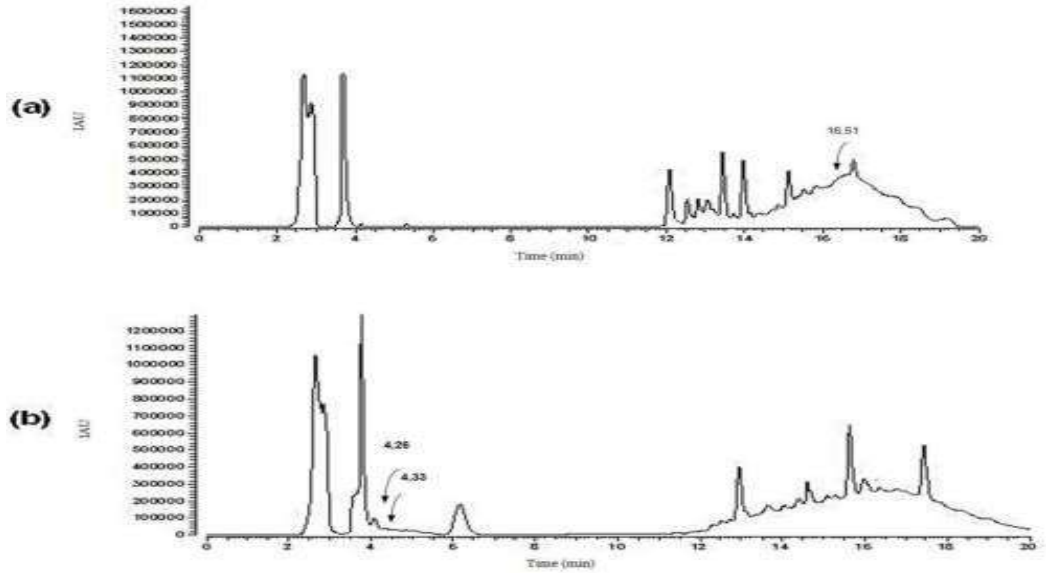

Figure4. Chromatograms of cowpea CC**(a)** and CT**(b)**.Crude Methanol extracts (CME).

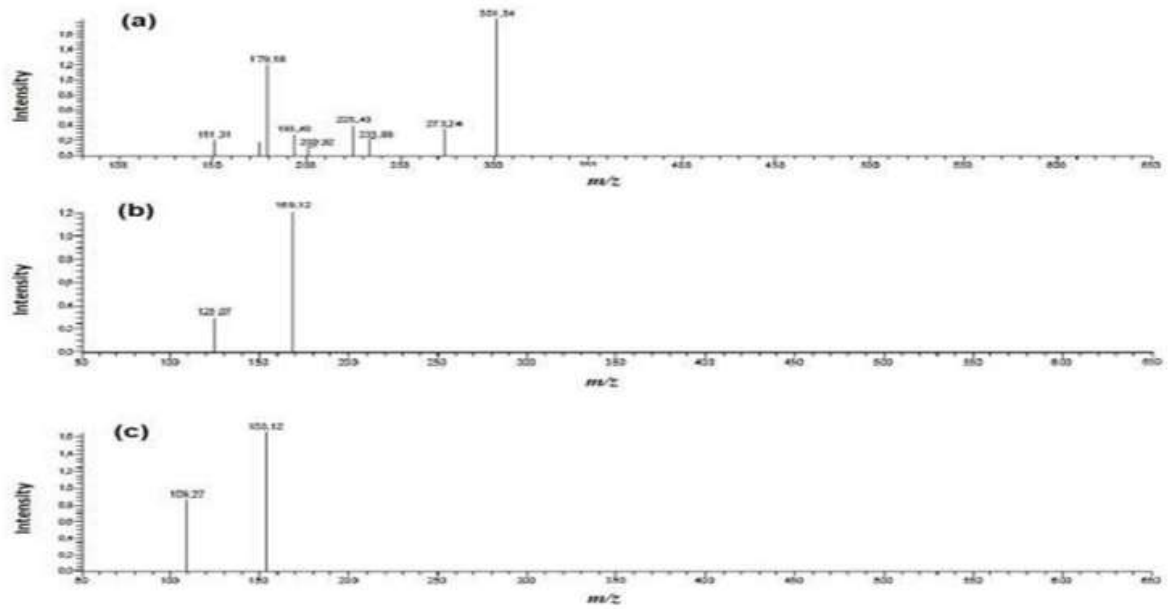

Figure 5. Mass spectra with molecular ion at m/z [M - H] - and fragments of phenolic compounds identified icowpea CC and CT. (a) quercetin (b) gallic acid and (c) protocatechuic acid (PCA).

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 932-941 www.ijprajournal.com ISSN: 2249-7781

Aliquots 14 and 17 chromatographic profiles

From the chromatogram analysis of the fraction 14 (Figure 4a), it was possible to identify the p- coumaric acid. The peak with retention time of 13.71 min exhibited a peak corresponding to the mass of its deprotonated molecular ion at m/z 163 [M - H] - and at m/z 119 fragment (Figure 5a), which compared with the standard it allowed the identification of p-coumaric acid. Regarding the chromatogram analysis of the fraction 17 (Figure 4b), it was possible to identify the flavonol quercetin (5b) and PCA (Figure 5c), which could be identified above, as well as the kaempferol, identified by comparison with standard, where the peak with a retention time of 18.12 min showed themolecular ion peak at m/z 285 [M - H] - and fragments at m/z 257, 217, 199, 175 and 151

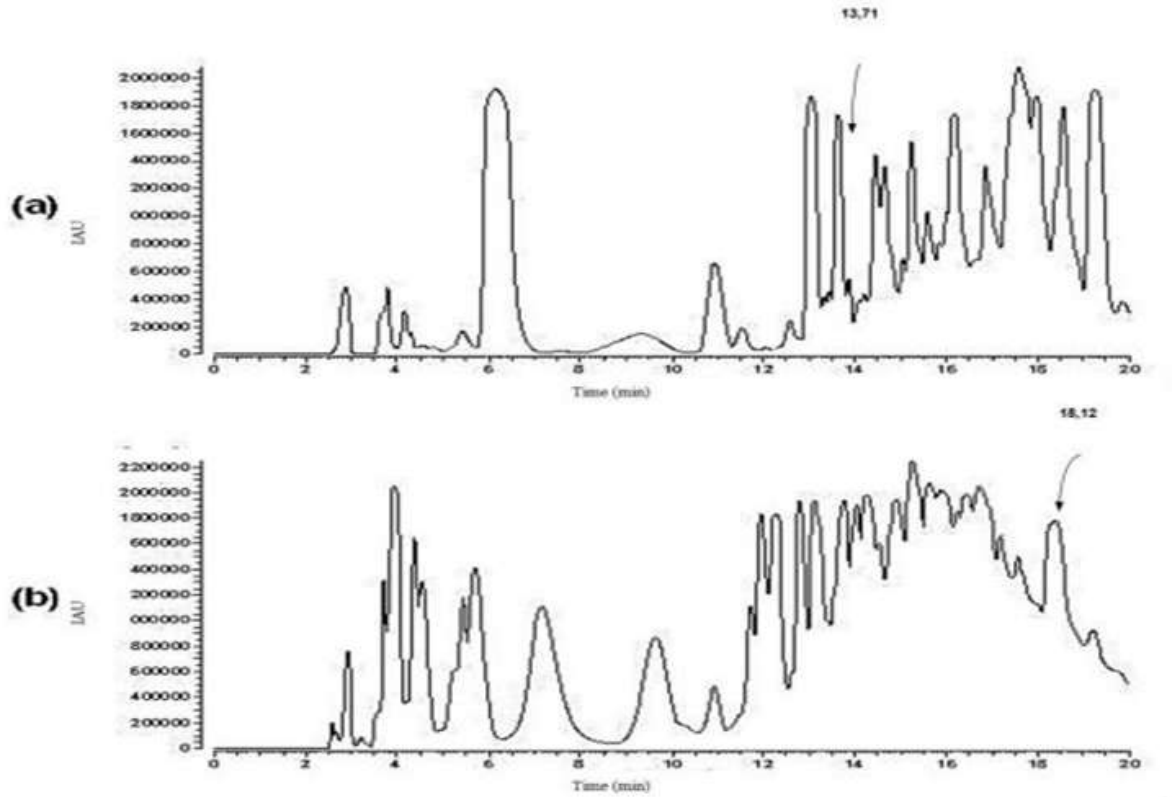

Figure 6. Chromatograms of the cowpea CC from fractions 14 (a) and 17 (b).

VI. PHARMACOLOGICAL WORK:

Stimulation of phenolics, antioxidant and antimicrobial activities in dark germinated mung bean sprouts
in response to peptide and phytochemical elicitors

Reena Randhir, Yuan has worked on The phenyl propanoid pathway (PPP) was stimulated in mung bean sprouts through the pentose phosphate and shikimate pathways, by natural elicitors such as fish protein hydrolysates (FPH), lactoferrin (LF) and oregano extract (OE). The higher antimicrobial activity was also observed with the higher stimulation of G6PDH and GPX activity during early stages of germination. This leads to the hypothesis that enhanced mobilization of carbohydrates (as indicated by G6PDH activity on days 2 and 4), enhanced polymerization of simple phenols (as indicated by GPX activity on day 3) contributed to high antioxidant activity producing intermediary metabolites.

The antioxidant and free radical scavenging activities of processed cowpea (Vigna unguiculata (L.) Walp.) seed extracts

Perumal Siddhuraju has worked on the antioxidative properties and total phenolic contents of two varieties of cowpea (Vigna unguiculata) were extracted with 70% acetone and the extracts were freeze-dried were examined. The unprocessed light brown seeds (LB) contained significantly higher level of total phenolics and tannins than the

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 932-941 www.ijprajournal.com ISSN: 2249-7781

dark brown seeds (DB). The extracts were screened for their potential antioxidant activities using tests such as , OH , ±,±- diphenyl-²- picrylhydrazyl (DPPH), ABTS+ , FRAP, linoleic acid emulsion and ²-carotene– linoleic acid in vitro model systems. At 800 ¼g of extract in the reaction mixture, the superoxide anion radical scavenging activity was found to be significantly higher in the raw and dry heated seed extracts than the hydrothermally processed seed samples of the respective varieties. The DPPH radical and ABTS cation radical scavenging activities were well proved and correlated with the ferric reducing antioxidant capacity of the extracts. Interestingly, among the various extracts, dry heated samples of LB and DB showed the highest hydroxyl radical scavenging activity of 83.6% and 68.2%, respectively. All extracts exhibited good antioxidant activity (74.3– 84.6%) against the linoleic acid emulsion system.

The antioxidant activity and free radical-scavenging capacity of phenolics of raw and dryheatedmoth bean (Vignaaconitifolia) (Jacq.) Marechalseed extracts

The antioxidative properties and total phenolic contents of Vigna aconitifolia were examined. The raw and dry heated samples were extracted with 70% acetone and the extracts were freeze- dried. The raw seeds contained higher levels of total phenolics (6.54%) and tannins (1.91%) than the dry heated seeds. The extracts were screened for their potential antioxidant activities using, OH , ±,±-diphenyl-²-picrylhydrazyl (DPPH), 2,22 - azinobis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS+), Ferric reducing/antioxidant power (FRAP), linoleic acid emulsion and Fe2+ chelating systems. At 1 mg of extract in the reaction mixture, the superoxide anion radicalscavenging activity. was found to be similar in raw and dry heated seed extracts. The DPPH radical and ABTS cation radical scavenging activities were well proved and correlated with the ferric reducing antioxidant capacity of the extracts. Interestingly, both raw and dry heated seed extracts showed the highest hydroxyl radical scavenging activity of 67.3% and 68.5%, respectively, at concentration of 1 mg/g extract. In addition, both extracts exhibited good peroxidation inhibiting activity (54.2% and 58.2%, respectively) against the linoleic acid emulsion system and the values were lower than BHA and Trolox. Fe2+ chelating activity was also detected in both raw and dry heated seed with EDTA equivalent of 0.61 mg and 0.45 mg/g extracts, respectively. Mung beans processed by solid-state bioconversion improves phenolic content and functionality relevant for diabetes and ulcer management.

Immunostimolatory activities of Vignamungo L. extract in male Sprague–Dawley rats

YogendrasinhB. solankidy has made a work to evaluate any immunostimulatory activities of the extract of V. mungo seeds in an animal model. The induction of any immunostimulatory effects were evaluated using measures of sheep red blood cells (SRBC)-induced humoral antibody titer, SRBC-induced delayed-type hypersensitivity (DTH), neutrophil adhesion, and in vivo phagocytosis (via the carbon clearance method) after host treatment with the extract. The results here indicated that primary and secondary antibody titers in the rats were significantly increased by treatment with the V. mungo extract as compared with those noted among rats in a control group. Increases in DTH response, the percentage (%) neutrophil adhesion, and in situ phagocytosis were also observed after treatment with the extract. The findings in there study suggest that V. mungo seed extract possesses profound immunostimulatory activities. These present study provides evidence that could help explain how the traditional use of V. mungo has been successful in the treatment of various disorders in humans.

VII. CONCLUSION

This review's conclusion is that several Vigna species have antioxidant activity and can treat a variety of ailments caused by the production of free radicals. It is possible thanks to the collaborative efforts of botanists, phytochemists, and pharmacists to understand the taxonomical characteristics of various medicinal plants, climatic conditions, various folklore usages, phytochemical, pharmacological, and antimicrobial values.Even though vigna species has demonstrated antioxidant properties and the ability to treat human illnesses linked to the production of free radicals, more phytochemical, pharmacognostical, and pharmacological research on this plant is still required.

CONFLICT OF INTEREST STATEMENT

All authors declare no conflictof interest.

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