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RESEARCH ARTICLE

PRELIMINARY PHYTOCHEMICAL SCREENING, GC-MS PROFILING AND IN VITRO EVALUATION OF BIOLOGICAL ACTIVITIES OF GARCINIA ATROVIRIDIS ROOT EXTRACTS.

Nur Salsabila Ahmad Roslan, Seema Zareen, Normaiza Zamri and Muhammad Nadeem Akhtar
Faculty of Industrial Sciences & Technology, Universiti Malaysia Pahang, 26300 Gambang, Malaysia.

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Abstract

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Therapeutic properties of the medicinal plant are due to the presence of phytochemical constituents. *Garcinia atroviridis* is locally known as 'asam gelugur' belongs to the Guttiferae family. Bioassay-guided solvent-solvent extraction method and yielded, hexane, dichloromethane, ethyl acetate, butanol, and methanolic extracts. These extracts were used to investigate the presence of phytochemicals. The preliminary phytochemical screening showed the existence of fixed oils and fats, carbohydrate, saponins, phenolic, flavonoids and anthraquinone glycosides in *G. atroviridis* roots. The chemical compositions were investigated by using Gas Chromatography-Mass Spectroscopy (GC-MS). Major compound identified in hexane, DCM, EA, BuOH and methanolic extracts was (*Z*)-13-docosenoic acid methyl ester (24.32%), ethyl-9-hexadecenoate (6.36%), *bis*(1,3-diisopropylcyclopentadienyl) (12.09%), 2-methyl-2-phenyl- 1,3-dioxolane (2.34%) and furfural (33.55%) respectively. The antibacterial and antioxidant activities of the extracts were investigated. The methanolic crude extract exhibited resistance towards both bacteria tested; *Bacillus subtilis* and *Escherichia coli*, thus suggesting its antibacterial activity.

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Introduction:-

Garcinia atroviridis is commonly known as 'asam gelugur' in Malaysia, India, Myanmar, and Indo-China. This plant species specifically belongs to the Guttiferae. This plant is endemic species in Peninsular Malaysia. It grows wildly in lowland and hill forest up to 600-meter altitude. It was also planted by the locals for its economic and medicinal purpose (MacKeen et al., 2000). The dried *G. atroviridis* fruits known as 'asam keping' are sold commercially as seasoning. It is sour and is used to season curry, dressing fish and others. *G. atroviridis* have medicinal values as traditionally, it has been used to treat a cough, the decoction of its leaf with roots can be used to treat an earache, and the juice of the leaf is given to female after delivery (Tisdale et al., 2003; Burkill, 1966). In the previous study (MacKeen et al., 2000), it was reported that the crude methanolic extract of the different parts of the *G. atroviridis* such as fruit, leaf, stem, and trunk barks showed it possessed antibacterial, antifungal, antioxidant, and antitumor-promoting properties. This study analyses the chemical compositions of the different extracts of *G. atroviridis* roots via preliminary phytochemical screening and GC-MS. The antibacterial activities of all extracts were determined by the disc diffusion method while antioxidant activities were evaluated based on the DPPH radical scavenging activities.

Corresponding Author:-Nur Salsabila Ahmad Roslan.
Address:-Faculty of Industrial Sciences & Technology, Universiti Malaysia Pahang, 26300

Materials and method:-

Collection of plant materials

3 kg of *Garcinia atroviridis* roots were collected from Maran, Pahang in June 2017. The roots were cleaned and air-dried for two weeks. The dried roots were then chopped into small pieces and ground into powdered form. The ground roots (1.76 kg) were subjected to the extraction process using the solvent-solvent extraction method as described in Figure 1.

Solvent-solvent extraction

The dried roots were ground into a coarse powder using a pulverizer. Fine ground pulverized material was dissolved in different solvents as described in Figure 1. Powdered plant material was soaked in methanol for three days at room temperature and the solvent was filtered by using a sieve. This was repeated 3-4 times until the extract gave no coloration. The extract was distilled and concentrated under reduced pressure in the Buchi rotavapor yielding a gum-like residue (45.45 g). The same was repeated with organic solvents and distilled water of increasing polarity (starting with lipophilic solvent *n*-Hexane, ending with the more hydrophilic *n*-Butanol). The solvent from each extract was filtered and concentrated under reduced pressure in Buchi rotavapour. Finally, the extracts of *n*-hexane, dichloromethane, ethyl acetate, and *n*-butanol were collected, weighed (8.72 g, 4.45 g, 9.79 g and 1.08 g, respectively) and stored in the refrigerator at 4 ^{o}C for further phytochemical analysis.

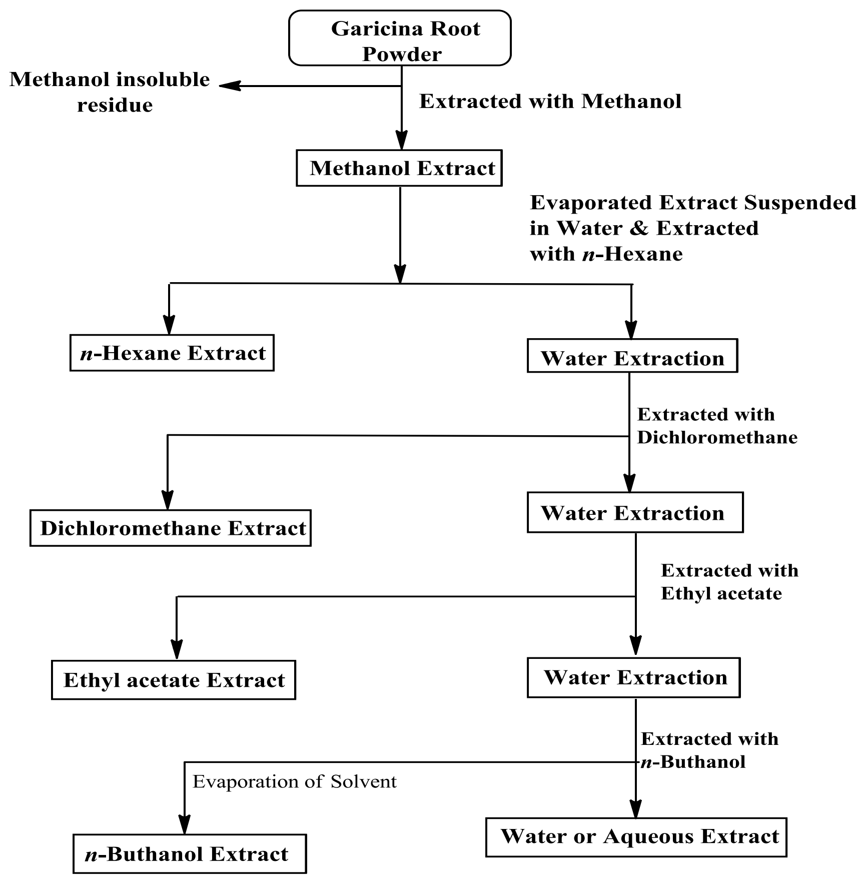

Figure 1:-Preparation of different extracts from the root of *G. atroviridis* using the different solvents.

Phytochemical Screening

The extract was subjected to preliminary phytochemical tests to determine the group of secondary metabolites present in the sample. The screening of the phytochemicals such as alkaloids, carbohydrates, anthraquinones, glycosides, saponin, protein and amino acids, phytosteroids, oils and fats, and phenols and flavonoids was carried out by following the standard procedure as mentioned by Sofowora (1993), Trease and Evans (1989) and Harborne (1984).

Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

GC-MS analysis was carried out by Agilent 7980A series GC instrument. The DB-1MS column with the dimensions of 30 m X 0.25 mm capillary column was used for the analysis. The initial temperature was kept at 50 °C for 3 minutes and the maximum temperature was up to 250 °C. Helium was used as a carrier gas at a flow rate of 1.0 mL/min and the total run time was 60 minutes. Interpretation of mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology MS library (NIST-MS library).

Antibacterial Test

Test microorganisms

The procedure was according to MacKeen et al., 2000. Four bacterial strains, i.e. *Bacillus cereus* (ATCC 11778), *Staphylococcus aureus* (ATCC 6538), *Escherichia coli* (ATCC 10536) and *Proteus vulgaris* (ATCC 33420) was used in this study. The nutrient broth was prepared, and the microorganism was cultured and incubated in the incubator shaker at 30°C for 24 hours. The concentration of the cultures was adjusted turbidometrically at a wavelength of 600 nm to 10^8 colony forming units (CFU) per mL.

Disc Diffusion Method

The bacterial cultures (10^8 CFU/mL) were inoculated on the prepared nutrient agar. 15 µl of samples with a concentration of 20 mg/mL was loaded onto blank filter paper discs. The loaded disc was placed on the previously inoculated agar. The plates were inverted and incubated in the incubator for 24 hours at 30 °C. The presence of clear inhibition zones around the discs showed the antibacterial activity of the extracts (MacKeen et al., 2000). The disc diffusion tests were presented in triplicate and the antibacterial activity was expressed as the mean of inhibition diameters (mm) produced by the extracts. Commercially available antibiotics discs were used as a positive control whilst the solvent used to dissolve the extracts acted as a negative control.

Antioxidant Assay

α, α-diphenyl-β-picrylhydrazyl (DPPH) radical scavenging method

The scavenging effects of samples for DPPH radical was determined according to the method by Hui et al. (2017), with slight modification. Briefly, the initial concentration of the sample was prepared at 1 mg/mL as well as concentration for ascorbic acid which acted as standard in this assay. 0.2mM DPPH was prepared freshly and must be protected from light throughout the experiment. 100 µL of DPPH was added to 100 µL of sample with different concentrations. After incubated for 30 minutes in the dark at room temperature, the absorbance of the solution was read at 517 nm. The solution without extract; only methanol and DPPH were used as a control. All test was performed in triplicates. The data were expressed as mean ± standard deviation (SD). The calculation of the percentage of radical scavenging activity was as followed:

$$\frac{Abs\ (C - S)}{Abs\ C}\ X\ 100$$

Abs C, Absorbance of control at t= 30 minutes
Abs S, Absorbance of sample t= 30 minutes

Results and Discussion:-

Preliminary Phytochemical Screening

The results of the presence of important metabolites in hexane (HEX), dichloromethane (DCM), ethyl acetate (EA), butanol (BuOH) and methanol (MeOH) extracts of *Garcinia atroviridis* roots are tabulated in Table 1. Anthraquinone glycosides are known to possess laxative property hence it is recommended to investigate the content of the compound present in the sample to determine the effectiveness of the laxative effects in the sample (Sakulpanich and Gritsanapan, 2009). Abdillah et al., 2015, reported the presence of flavonoid, tannin, steroid, triterpenoid, and cumarine in the *G. atroviridis* ethanolic extract and the extract showed significant anti-plasmodium activity both *in vitro* and *in vivo*. Hydroxycitric acid (HCA) and flavonoids presence in *G. atroviridis* fruit extract contributed to its hypolipidemic properties (Amran et al., 2009). HCA is usually promoted as a weight loss supplement either alone or in combination with other supplements (Roongpisuthipong et al., 2007; Downs et al., 2005). In this study, flavonoids were found in ethyl acetate, butanol and methanolic extracts of *G. atroviridis* roots.

G. atroviridis leaves and fruits have potential to be used as a source of natural antioxidants and nutrients for therapeutic purposes against free radical-mediated health conditions as it contained high phenolic compound (Nursakinah et al., 2012). The present study showed the presence of the phenolic compound in ethyl acetate, butanol, and methanolic extracts. Saponins present in ethyl acetate, butanol, and methanolic extracts are therapeutically essential as they are revealed to have hypolipidemic and anticancer activities (Doughari et al., 2012).

Table 1:-Phytochemical screening of different extracts from *Garcinia atroviridis* roots

Phytochemical components	HEX	DCM	EA	BuOH	MeOH
Alkaloids	-	-	-	-	-
Anthraquinones glycosides	-	-	-	+	+
Carbohydrates	-	-	+	+	++
Flavonoids	-	-	++	+++	+++
Glycosides	-	-	-	-	-
Oils and fats	+++	+	+	+	+
Phenolic compounds	-	-	+++	+++	+++
Phytosteroids	-	-	-	-	-
Protein and amino acids	-	-	-	-	-
Saponin	-	-	+++	+++	+++

+++ Prominently Present, ++ Moderately Present, + Slightly Present, - Absent

GC-MS Analysis

The GC-Mass Spectrometry analysis of all extracts was given in Table 2 to 6. Major compounds in hexane (HEX), dichloromethane (DCM), ethyl acetate (EA), butanol (BuOH) and methanol (MeOH) extracts are (*Z*)-13-docosenoic acid methyl ester (24.32%), ethyl-9-hexadecenoate (6.36%), *bis*(1,3-diisopropylcyclopentadienyl) (12.09%), 2-methyl-2-phenyl- 1,3-dioxolane (2.34%) and furfural (33.55%), respectively. Some compounds were already identified in the previous study as important phytochemicals. Fatty acids such as methyl stearate, linoleic acid, oleic acid, and 9-octadecenoic acid were reported present in *G. schomburgkiana, G. xanthochymous* and G. *mangostana* (Meechai et al., 2015; Meechai et al., 2016; Manohar et al., 2014; Ajayi et al., 2007). Oleic acid helps in lowering low-density lipoprotein (LDL) cholesterol level (Grundy, 1989). The oil obtained from *G. xanthochymus* seeds showed significant antimicrobial antioxidant activity (Manohar et al., 2014). Furfural in *G. schomburgkiana* and *G. indica* contributed to its antimicrobial activity (Sutar et al., 2012; Meechai et al., 2016).

Hexadecanoic acid, octadecadienoic acid, 9-octadecene, (*Z, Z*)-9,12-octadecadienoic acid, (Z)-9-octadecenoic acid and *bis*(2-ethylhexyl) phthalate was detected in *G. cambogia*, *G. atroviridis, G. mangostana* and *G. indica* (Mahadkar et al., 2013; Tan et al., 2013; Irulandi et al., 2016; Rahmayanti et al., 2017). Phthalate is a renowned synthetic plasticizer (Saeidnia and Abdollahi, 2013). Even though the toxicity of phthalate is different depending on its structure, the existence of phthalate on medicinal plants often displayed antimicrobial activities (Rahmayanti et al., 2017). Hexadecanoic acid plays an essential role in the anti-inflammation process which by helping to design a specific inhibitor of phospholipase A (Wanxi et al., 2014).

Table 2:-The chemical composition of *G. atroviridis* hexane root extract

No.	Compound Name	RT	Area%	Mol. formula
1	Ethylbenzene	5.14	6.94	C_8H_{10}
2	1,3-Dimethyl benzene	5.36	2.33	C_8H_{10}
3	1-Ethyl-2-methyl benzene	7.95	0.41	C_9H_{12}
4	1,2,4-Trimethyl benzene	8.21	0.24	C_9H_{12}
5	1,2,3-Trimethyl benzene	8.96	0.91	C_9H_{12}
6	Hexadecanoic acid methyl ester	37.79	2.08	$C_{17}H_{34}O_2$
7	(*Z, Z*)-9,12-Octadecadienoic acid methyl ester	44.31	1.50	$C_{19}H_{34}O_2$
8	(*Z*)-9-Octadecenoic acid methyl ester	44.53	2.53	$C_{19}H_{36}O_2$
9	Methyl stearate	45.25	0.41	$C_{19}H_{38}O_2$
10	*Cis*-Vaccenic acid	45.73	0.14	$C_{18}H_{34}O_2$
11	*Cis*-11-Eicosenoic acid methyl ester	48.22	4.23	$C_{21}H_{40}O_2$
12	Methyl-13-eicosenoate	48.31	0.25	$C_{21}H_{40}O_2$
13	Methyl-9-eicosenoate	48.45	0.40	$C_{21}H_{40}O_2$
14	Eicosanoic acid methyl ester	48.58	0.22	$C_{21}H_{42}O_2$
15	*Cis*-11-Eicosenoic acid	49.05	0.27	$C_{20}H_{38}O_2$
16	(*Z*)-13-Docosenoic acid methyl ester	50.84	24.32	$C_{23}H_{44}O_2$
17	Methyl-11-docosenoate	51.08	1.67	$C_{23}H_{44}O_2$

18	Erucic acid	51.68	2.14	$C_{22}H_{42}O_2$
19	(*Z*)-9-Hexadecenoic acid methyl ester	52.40	1.00	$C_{17}H_{32}O_2$
20	(*Z*)-15-Tetracosenoic acid methyl ester	54.44	14.24	$C_{25}H_{48}O_2$
21	Tetracosanoic acid, methyl ester	54.98	0.35	$C_{25}H_{50}O_2$
22	5- (1,1-dimethylethyl)-1,3-Benzenedicarboxylic acid	55.65	0.16	$C_{12}H_{14}O_4$
23	1,1,3-trimethylcyclopentane	55.76	0.16	C_8H_{16}
24	1-Bromo-11-iodoundecane	60.49	0.60	$C_{11}H_{22}BrI$
25	Hexacosanoic acid methyl ester	61.12	0.31	$C_{27}H_{54}O_2$
26	4,4-dimethyl-cholesta-6,22,24-triene	63.66	0.38	$C_{29}H_{46}$

Table 3:-The chemical composition of *G. atroviridis* dichloromethane root extract

No.	Compound Name	RT	Area%	Mol. formula
1	1-Methyl-1-hydroxymethyl adamantane	4.96	4.07	$C_{12}H_{21}O$
2	*Trans*-4-ethyl-5-octyl-2,2-*bis*(trifluoromethyl)-1,3-dioxolane	46.27	0.42	$C_{15}H_{24}F_6O_2$
3	2-Bromo-4,5-dimethoxycinnamic acid	47.62	0.94	$C_{11}H_{11}BrO_4$
4	11-Eicosenoic acid methyl ester	48.22	2.40	$C_{21}H_{40}O_2$
5	(*E*)-2-Bromobutyloxychalcone	48.59	0.75	$C_{19}H_{19}BrO_2$
6	5,14,23-Octadecatrien-14,15-diol	48.66	1.90	$C_{28}H_{52}O_2$
7	6-Methoxy-9-methyl-tricyclo [7.2.1.0(3,8) dodeca-3(8)],4,6-triene-2,12-dione	48.86	2.94	$C_{14}H_{14}O_3$
8	3,5-*Bis*(1,1-dimethylethyl)-1,2-benzenediol	49.28	0.43	$C_{14}H_{22}O_2$
9	2-(Acetoxymethyl)-3-(methoxycarbonyl)biphenylene	49.92	0.81	$C_{17}H_{14}O_4$
10	(5*α*,14*β*)-androstan-17-one	51.60	2.77	$C_{19}H_{30}O$
11	3-[(4-Methyl-5-oxo-3-phenylthio)tetrahydrofuran-2-yloxymethylene]-cyclopenteno[4.3-b] tetrahydrofuran	51.65	0.61	$C_{19}H_{18}O_4S$
12	Ethyl-9-hexadecenoate	51.81	6.36	$C_{18}H_{24}O_2$

Table 4:-The chemical composition of *G. atroviridis* ethyl acetate root extract

No.	Compound Name	RT	Area%	Mol. formula
1	*Tert*-butyl 3-Methylbenzyl alcohol	4.95	1.93	$C_{12}H_{18}O$
2	1-(3,4-Methylenedioxyphenyl)-1-methoxypropan-2-one	6.90	0.66	$C_{11}H_{12}O_4$
3	Mesitylene	8.98	0.44	C_9H_{12}
4	1,3,5-Benzenetriol	28.57	1.31	$C_6H_6O_3$
5	Methyl 2,4,6-trihydroxybenzoate	32.12	0.82	$C_8H_8O_5$
6	Hexadecanoic acid ethyl ester	40.60	1.21	$C_{18}H_{36}O_2$
7	2,5-*Bis*(1,1-dimethylethyl)-1,4- benzenediol	42.33	1.46	$C_{14}H_{22}O_2$
8	Di(2-isopropoxyphenyl) ester glutaric acid	44.40	1.24	$C_{23}H_{28}O_6$
9	(*E*)-9-Octadecene	44.53	0.47	$C_{18}H_{36}$
10	Linoleic acid ethyl ester	45.94	0.85	$C_{20}H_{36}O_2$
11	n-Propyl-9-octadecenoate	46.08	1.28	$C_{21}H_{40}O_2$
12	Octadecanoic acid ethyl ester	46.62	0.22	$C_{20}H_{40}O_2$
13	(*E*)- 1-(2,6-Dihydroxy-4-methoxyphenyl)-3-phenyl-2-propen-1-one	49.47	0.85	$C_{16}H_{14}O_4$
14	1,1,1-Trifluoro-2-(trifluoromethyl)-4-penten-2-ol	50.07	0.38	$C_6H_6F_6O$
15	Cholest-5,9(11)-dien-3*β*-ol acetate	50.39	2.10	$C_{29}H_{46}O_2$
16	1,6,10,11-Tetrahydroxy-8-(α-methylbenzyl)-5,12-naphthacenedione	50.45	2.32	$C_{26}H_{18}O_6$
17	3,5,3',5', Tetrakis-trifluoromethylbiphenyl	50.48	1.09	$C_{16}H_6F_{12}$
18	3,3',4,4',5,5',6,6'-Octamethoxy-2,2'-Bisphenol	50.54	2.51	$C_{20}H_{26}O_{10}$
19	*Bis*(1,3-diisopropylcyclopentadienyl), cobalt	50.80	12.09	$C_{22}H_{34}Co$
20	Ethyl-13-docosenoate (ethyl erucate)	51.81	6.88	$C_{24}H_{46}O_2$
21	14-Bromo pentadecanoic acid	52.11	0.97	$C_{15}H_{29}BrO_2$
22	*β*-Carotene	52.24	0.19	$C_{40}H_{56}$
23	(*E*)-9-Tetradecenoic acid	55.93	1.04	$C_{14}H_{26}O_2$
24	(*E*)-11-Hexadecenoic acid ethyl ester	55.95	1.52	$C_{18}H_{34}O_2$

Table 5:-The chemical composition of *G. atroviridis* butanol root extract

No.	Compound Name	RT	Area%	Mol.formula
1	2-Methyl-2-phenyl- 1,3-dioxolane	4.95	2.34	$C_{10}H_{12}O_2$
2	2-Chloroethanol	9.07	0.25	C_2H_5ClO
3	1-(1-Hydroxybutyl)-2,5-dimethoxybenzene	13.67	0.17	$C_{12}H_{18}O_3$
4	2,6-*Bis*(1,1-dimethylethyl) phenol	23.84	0.14	$C_{14}H_{22}O$
5	2,5-*Bis*(1,1-dimethylethyl)- 1,4-benzenediol	49.94	0.76	$C_{14}H_{22}O_2$
6	6,7-Dihydro-10-hydroxy-1,2,3-trimethoxy-benzo(a)heptalen-9(5*H*)-one	50.39	1.60	$C_{19}H_{20}O_5$

Table 6:-The chemical composition of *G. atroviridis* methanolic root extract

No.	Compound Name	RT	Area%	Mol.formula
1	Furfural	4.57	33.55	$C_5H_4O_2$
2	4-Hydroxy-4-methyl-2-pentanone	4.94	0.65	$C_6H_{12}O_2$
3	4-Ethyl-3-hexanol	4.96	0.58	$C_8H_{18}O$
4	Furyl hydroxymethyl ketone	11.97	5.29	$C_6H_6O_3$
5	Levoglucosenone	12.83	13.38	$C_6H_6O_3$
6	2-Furanmethanol	12.94	0.46	$C_5H_6O_2$
7	Dimethyl 2-hydroxysuccinate	13.31	1.97	$C_6H_{10}O_5$
8	2,3-Dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one	14.00	4.76	$C_6H_8O_4$
9	1,4:3,6-Dianhydro-α-D-glucopyranose	16.19	1.79	$C_6H_8O_4$
10	6-Bromo hexanoic acid	24.65	1.57	$C_6H_{11}BrO_2$
11	Propyl propanedioic acid	24.67	0.13	$C_6H_{10}O_4$
12	3,4-Altrosan	24.80	0.21	$C_6H_{10}O_5$
13	1,6-Anhydro-β-D-glucopyranose	24.95	0.81	$C_6H_{10}O_5$
14	3,4,5-trihydroxy-5-(3-methyl-2-butenyl)-2-(3-methyl-1-oxobutyl)-2-cyclopenten-1-one	44.54	0.45	$C_{15}H_{22}O_5$
15	*Bis*(2-ethylhexyl) phthalate	51.21	1.09	$C_{24}H_{38}O_4$

Table 7:-Comparative inhibition zones of different extracts of *G. atroviridis* roots

Extract	Inhibition zone (mm)			
	B. *cereus*	S. *aureus*	E. *coli*	P. *vulgaris*
HEX	9.0±0	9.3±0.58	0.00	0.00
DCM	10.33±1.15	9.67±0.58	0.00	0.00
EA	7.0±0	7.0±0	0.00	0.00
BuOH	10.33±2.31	10.67±1.53	0.00	0.00
MeOH	0.00	0.00	0.00	0.00

The concentration of the root extracts is 20mg/mL. Tests were carried out in triplicate. Data was expressed as mean ± standard deviation (SD).

Table 8:-Inhibition zone of positive and negative control

Control	Inhibition zone (mm)			
	B. *cereus*	S. *aureus*	E. *coli*	P. *vulgaris*
Chloramphenicol (+ve)	25±1	ND	20±0	ND
Gentamicin (+ve)	ND	20±1	ND	25±0
Acetone (-ve)	0.00	0.00	0.00	0.00
Hexane (-ve)	0.00	0.00	0.00	0.00

Tests were carried out in triplicate. Data was expressed as mean ± standard deviation (SD)
ND: not determine; the particular antibiotic disc was not tested on the microorganisms
+ve: positive control

-ve: negative control

Antioxidant test DPPH assay

DPPH radical scavenging method was used to evaluate the antioxidant activity of the root extracts of *G. atroviridis*. DPPH assay is a fast and efficient method to determine the free radical scavenging activity. The color changed from purple to yellow signifies the decline in absorbance of the DPPH radical (Jadid et al., 2017). In this study, all extracts showed a similar increasing trend in antioxidant activity with an increase in their concentrations (Table 9).

IC means inhibition concentration and IC_{50} is the concentration of the sample or antioxidant that is required to inhibit 50% of DPPH radicals (Jadid et al., 2017). Hence, the lower the IC_{50}, the better the antioxidant activity. Phongpaichit et al., 2007, considered the IC_{50} range from 10 to 50 mg/mL to display strong antioxidant activity, while 50 to 100 mg/mL is considered intermediate and weak antioxidant activity for IC_{50} value that more than 100 mg/mL. According to the IC_{50} of the extracts (Table 10), butanol (50.59 µg/mL) showed better antioxidant activity followed by ethyl acetate (51.7 µg/mL) and dichloromethane (53.17 µg/mL). Meanwhile, methanol (124.7 µg/mL) and hexane (131.8 µg/mL) extracts displayed weak activity. However, none of the extracts were comparable to the standard, ascorbic acid (AA) with IC_{50} of 13.21 µg/mL. Previous study (MacKeen et al., 2000), methanolic roots extracts of *G. atroviridis* showed significant antioxidant activity.

Table 9:-Effect of *Garcinia atroviridis* root extracts on DPPH radicals

Conc. (µg/mL)	Percentage of inhibition (%)					
	AA	HEX	DCM	EA	BuOH	MeOH
7.8125	49.2±0.041	1.1±0.006	10.3±0.011	9.2±0.031	6.4±0.033	1.2±0.009
15.625	85.4±0.007	5.5±0.017	15.6±0.017	14.6±0.010	16.6±0.022	6.8±0.010
31.25	86±0.005	10.9±0.011	27.6±0.005	28.2±0.018	27.3±0.011	13.3±0.032
62.5	86.1±0.006	20.2±0.014	49.5±0.020	47.8±0.024	48.3±0.024	22.8±0.031
125	86.7±0.007	36±0.011	79.0±0.012	77.8±0.010	76.7±0.015	38.4±0.014
250	86.1±0.003	59.7±0.013	82.1±0.008	79.9±0.015	81.8±0.008	58.8±0.027
500	86.6±0.005	81.6±0.005	82.1±0.003	79.9±0.003	82.0±0.002	80.8±0.004
1000	87± 0.004	81.5±0.005	80.4±0.004	72.9±0.026	80.9±0.004	82.2±0.005

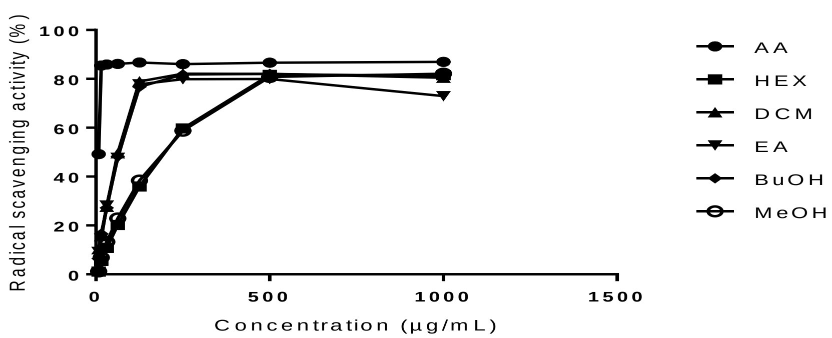

Figure 2:-Graph chart of the percentage activity of the samples. The values represent the percent of the DPPH radical scavenging activity. Measurements were carried out in triplicate. Means and standard deviation are indicated.

Table 10:-IC_{50} (µg/mL) of *G. atroviridis* root extracts

Sample	IC_{50} (µg/mL)
AA (control)	13.21±2-14
HEX	131.8±3.11
DCM	53.17±8.76
EA	51.7±2.18
BuOH	50.59±13.21

MeOH	124.7± 11.11

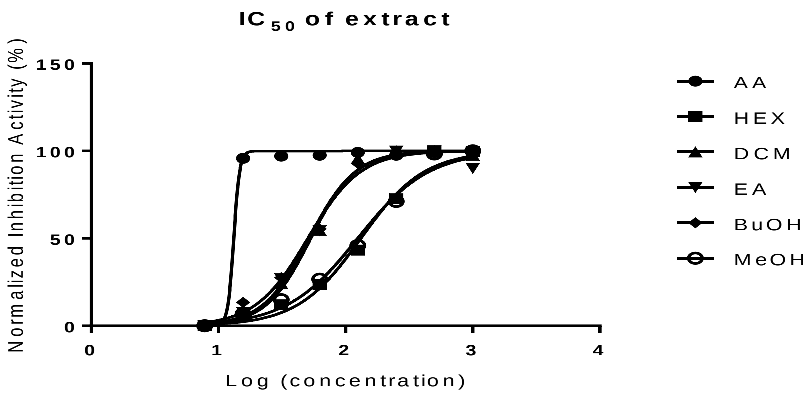

Figure 3:-IC_{50} (μg/mL) graph of the *G. atroviridis* root extracts

Conclusion:-

This paper emphasizes the phytochemical profiling of *G. atroviridis* roots extracts using preliminary phytochemical screening and GC-MS analysis. In this study, it was shown that the *G. atroviridis* roots extract contained many biologically active constituents, hence correlates with previous studies. The extracts of the sample were then tested for its antibacterial and antioxidant activity where promising results can be observed, though, further study such as bioprospecting is important to support its biological properties.

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