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ISOLATION AND DIAGNOSIS OF PHENOLIC COMPOUNDS OF *GLOEOCAPSA* SP PCC7428 AND THEIR ANTIMICROBIAL ACTIVITY

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Abstract

This study aims to isolate and diagnose phenols in the raw ethanol extract and the phenolic extract of *Gloeocapsa* sp. PCC 7428 alga by means of thin layer chromatography method (TLC) and high performance liquid chromatography (HPLC). The results of (TLC) showed the existence of Gallic acid, Phenol and Tannic acid. The R_f of the phenolic extract was consistent at (0.93) for Gallic acid, (0.98) for phenol and (0.91) for tannic acid. The results of (HPLC) showed the existence of the phenolic compounds; Apigenin, Catechine, Keampferol, Rutin, Qucetine, Gallic acid and Coumarin. The highest concentration and percentage among these compounds was of the phenolic compound Gallic acid in the raw ethanol extract (63.2 ppm and 2.7%) and in the phenolic extract (58.6 ppm and 5.1%). The efficacies of the extracts were studied against bacteria *Staphylococcus aureus, Esherichia coli, Klebsiella pneumoniae, Proteus mirabilis and Pseudomonas aeruginosa* and against the pathogenic fungi *Fusarium solani, Alternaria alternata* and *Candida albicans;* the ethanol extract showed no inhibition against all kinds of bacteria and fungi, whereas the phenolic extract inhibited all kind of bacteria and fungi with different diameters; the highest inhibition against bacteria *Klebsiella pneumonia* with a diameter of 25 mm, and the least inhibition was against bacteria *Staphylococcus aureus* with a diameter of 19 mm. The highest inhibition against fungi was seen against *Alternaria alternata* with a diameter of 42 mm, whereas the least was against *Candida albicans* with a diameter of 18 mm.

Key words: Phenolic compounds, *Gloeocapsa* sp PCC 7428, raw ethanol extract, phenolic extract, antimicrobial activity.

Introduction

Algae are autotrophic living beings *i.e.* they are characterized by their ability to form Carbon (C) through photosynthesis using Sun light, CO_2 and water H_2O (Bajpai, 2019; Frost *et al.,* 2012). Among them are the blue green algae (cyanobacteria) which are of the oldest and largest groups of negative Gram living beings on Earth (Sergeev, 2018; Shestakov and Karbysheva, 2017). Since cyanobacteria have a large group of secondary metabolic compounds, they were used as substances of anti- effect against viruses, fungi, bacteria and cancer. Cyanobacteria are also used in producing biofuel (Kumar *et al.,* 2019; Vijayakumar and Menakha, 2015). Among the genera of cyanobacteria is the alga *Gloeocapsa* sp. which is characterized by being of single spherical cell after this cell divides, other cells are formed either half- spherical, oval or ellipsoidal surrounded by distinguished gelatine sheath which is also surrounded by older gelatine sheath; these cells do not have clear gas vesicles (Athbi, 2014). The cell is constituted of two areas the outer is blue green that contains Chlorophyll and Phycocyanin, and a central area that contains Chromatin granules (Dutta, 2018). *Gloeocapsa* alga has the potency to produce carbohydrate and fat and is used to produce food additive, animal fodder, fertilizers and biofuel (De Farias Silra *et al.,* 2019).

Cyanobacteria are characterized by producing phenolic compounds, and there are different types of phenolic compounds such as Apigenin which is one of the flavonoids compounds found in fruit and vegetables as well as red alga *Acanthophora spicifera* (Mohamed *et al.,* 2020; Shoubaky *et al.,* 2016) which is effective against many cancerous diseases like leukaemia, Colorectal cancer, pancreatic cancer, breast cancer, stomach cancer, liver cancer, skin cancer, cervical cancer, prostate cancer and lung cancer and against viruses and bacteria, and it also reduces blood pressure (Imran *et al.,* 2020; Yan *et al.,* 2017). Catechine is another flavonoid compound that exists in some types of fruit such as apple, pear, cocoa, as well as in wine and green tea (Pervin *et al.,* 2019; Mangels *et al.,* 2017); it is an anti- oxidant,

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anti- bacterial and anti- inflammation, and is used in the treatment of heart and cancer diseases (Bernatoniene and Kopustinskiene, 2018; Wang *et al*., 2014). Kaempferol is another flavonoid that exists in many edible plants like tea, broccoli, cabbage, turnip, beans, chicory, leek, tomato, strawberry, grape and in plants or plant products that are commonly used in traditional medicine like Ginkgo Biloba. Some epidemical studies found a positive correlation between the consumption of food that contains Kaempferol and the reduction of risk of developing many disorders like cancer and cardiovascular diseases. Many studies showed that Kaempferol has a wide group of medicinal activities including anti- oxidation, anti-inflammation, antibiotic and anti- cancer. It is also useful for the heart and the nervous system and is anti-diabetics, anti- Osteoporosis, anti- germs and anti- anxiety (M Calderon- Montano *et al.,* 2011). Rutin is also a phenolic compound that exists in fruit and vegetables and it is anti- bacteria, fungi, inflammation, oxidation and cancer; it is also a nervous sedative and a vasodilator and is used in the treatment of blood sugar illness (Al-Dhabi *et al.*, 2015; Gullon *et al.*, 2017). Qurcetine is one of the flavonoids and it exists in fruit and vegetables like red onion, green tea and red wine and is used as a treatment; it is used in the manufacture of medicines and is considered an anti- oxidant, anti- cancer, anti- virus and anti- inflammation; it also is used in treating cardiovascular diseases, asthma, diabetes and schizophrenia (Zaplatic *et al.,* 2019). Gallic acid is one of the phenolic acid which exists in fruit and vegetables as well as in seeds, fruit juices, edible fungi and different tea leaves (Badea *et al.,* 2019; Tang and Cheung, 2019); it is used to treat cardiovascular diseases and is an antibacterial, anti- viral, anti- cancer and anti-inflammatory (Abdul Gafor e*et al.*, 2020; Yan *et al.*, 2019). Finally, Coumarin which has a strong odour and is anti- cancer and anti- oxidant; it fights human immunodeficiency virus (HIV) and is also an anti- asthma, anti- inflammation, a sedative and a vasodilator (Mishra *et al.*, 2020; Miao *et al.*, 2016).

Materials and methods

Growing and preserving isolates

After they were obtained from the laboratories of the research unit in Mosul University/College of Pure Sciences - Department of Biology, Pure isolate Gloeocapsa sp. PCC7428 (AL-Katib *et al*., 2017) is cultured on the agricultural adjusted liquid medium Chu 10 and incubated in an incubator under conditions of growing, at a temperature of 25±2ºC and lighting of 2500 Lux (Falch *et al.,* 1995). The liquid cultures (simple photobioreactors) were used by preparing the liquid Chu 10 medium. And put it in 4-5 liters glass bottles, with made two holes in the cap of the bottle to insert sterile rubber tubes through them. The first hole passes through a rubber tube to the inside of the bottle and is connected with a bubble stone so that the rubber tube does not float inside the glass bottle. As for the second side of this tube that goes outside the bottle, it is connected with a millipore filter (0.45 mm), then a second sterile rubber tube connected to a small developer which used in the fish tank to enter the air into the glass bottles is connected, while the second hole for the glass bottle is provided with a rubber tube that is used as a ventilation hole to extract the excess air, and these simple reactors are placed in a incubating room controlled by conditions of 2±25°C and lighting 2500 Lux has a 16 hour light sequence: 8 hours dark as in Fig. 1, (Falch *et al.,* 1995).

Preparation of Raw Ethanolic Extracts

Le Grand *et al.,* (1988) method was used in preparing the ethanolic extract of *Gloeocapsa* sp PCC7428, after collecting a fresh biomass from it and making a centrifuge and drying it in the oven at a temperature of 45°C for a period of approximately one day. Then the dry biomass was collected and kept in a closed box Until the ethanol extract was prepared, as 5 g of dried algae were weighed and thoroughly ground with ceramic mortar and placed in a glass beaker and added to it 50 ml of ethanol alcohol at a concentration of 95% and then placed inside a snow bath and using the electric motor device for 10 hours after which the mixture was left in the refrigerator for 24 hours An hour for soaking, then filtered with Buechner funnel containing on (Whatman No. 1) filter paper. The filtrate was taken and the ethanol was evaporated using the Rotary Vacuum Evaporator, as the apparatus operates under vacuum pressure and a temperature of no more

Fig. 1: Shows a simple hand-made bio-reactor.

than 40°C. The layer consisting of the raw extract after evaporation was taken and kept in sealed glass bottles until use.

Acid hydrolysis

Since the phenols are not found in a free form, but are linked with sugar by glycoside bond, then they form cyclosides inside the plant. Thus, to purify and diagnosis of phenols, acid decomposition process is performed to break glycoside bonds and release the phenols from sugar . Depending on (Harborne, 1973), the process done by using HCL (2 M) as a solvent for 4 mL of ethanolic algae extract and put the mixture in a glass jar and heated in a rocking water bath at a temperature of 90-100°C, For a period of half an hour, then it is allowed to cool down at the laboratory temperature. Then, after applying the separation suppression, add 100 ml of ethyl acetate to it. On the bottom layer, good shake is required in the separating funnel, and two layers are formed as well and concentrated using a rotary evaporator device to obtain the ethyl acetate extract and then stored in the refrigerator until use.

Diagnosis of Phenolic Compounds using Thin Layer Chromatography (TLC) technique

The researcher method (De *et al.,* 2017) was used to diagnose phenols by using Silica gel G f 254, thickness 0.25 mm, Merck, Germany) and put the phenolic extract on the plate and used a solvent mixture system (separation solution) butanol: glacial acetic acid: distilled water in proportions (70:25:5) respectively, as a mobile phase, was placed in the consist jar and left for three hours, then the spots were diagnosed using FeCl3 ferric chloride at a concentration of 1% and the rate of flow (Rf) of a particular material is the ratio of the distance the spot moved above the origin to the distance the solvent front moved above the origin.

Diagnosis of Phenolic Compounds using High Performance Liquid Chromatography (HPLC) technique

The High Performance Liquid Chromatography (HPLC) separator uses a capillary and polar property to separate phenolic compounds separated from plants, as most of these compounds are characterized by being weak acid that varies with base conditions and dissolves in solvent easily (Liang *et al.*, 1997; Dolan, 2000; Majors, 2001). The diagnostic process for these compounds was carried out in the laboratories of the Ministry of Science and Technology/Department of Environment and Water by means of a high-performance liquid chromatography HPLC type (SYKAM) German origin, where the carrier phase used methanol: distilled water: formic acid (5: 25: 70) and the separation column (C18 - ODS) was at a dimension of 25 cm * 4.6 mm (25) to separate the phenols and use the UV detector: UV 280 nm, where The velocity of flow of the carrier phase was: 1.3 ml/min (Mradu *et al.,* 2012).

Antibacterial activity of cyanobacterial extract against a number of pathogenic bacteria

The antibacterial activity was done by using Well Diffusion Assay Method (Perez *et al.,* 1990) as young colonies of pathogenic bacteria were transferred to the broth medium and incubated at a temperature of 37°C for 24 hours and the bacterial suspension was diluted by using the normal saline solution. The effect of extracts on bacteria showed after putting it (50 µl) in a well diameter of 6 mm and incubated in incubator at 37°C for 24 hours, after which inhibition zones were measured (Vandepitte, 1991).

Antifungal activity of cyanobacterial extract against a number of pathogenic fungi

Well Diffusion Method (Magaldi *et al.,* 2004) followed. The medium for the development of fungi Potato Dexteros Agar (PDA) was prepared under sterile conditions poured into Petri dishes and then inoculum with pathogenic fungi (as method mentioned previously in (6)) and incubated the dishes in the incubator at a temperature of 27°C for a period of 4 days, after which the inhibition zones inhibition were measured in mm around each well.

Table 1: Rf for phenolic extract and standard phenolic compounds for *Gloeocapsa* sp.

Standard phenolic compounds	Rf values	Rf values for phenolic extract
Galic acid	0.93	0.93
Phenol	0.98	0.98
Tanic acid	0.91	0.981

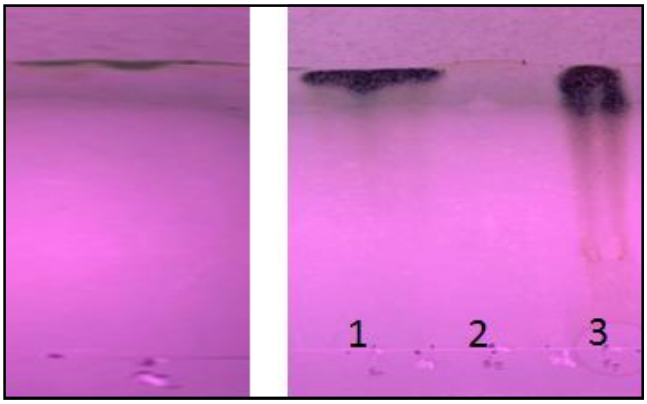


Fig. 4: Shows the separation of the phenolic compounds of ***Gloeocapsa*** sp and (1) Gallic acid, (2) phenol and (3) Tannic acid (TLC) compared to standard compounds.

Results and discussion

The results of TLC separating technique table 1 and Fig. 4 showed identical values of phenols (phenol, gallic acid and tannic acid) Rf with phenols in the phenolic cyanobacterium *Gloeocapsa* extract, that are respectively (0.93, 0.98, 0.91) as shown In table 1 and Fig. 4.

Table 2:

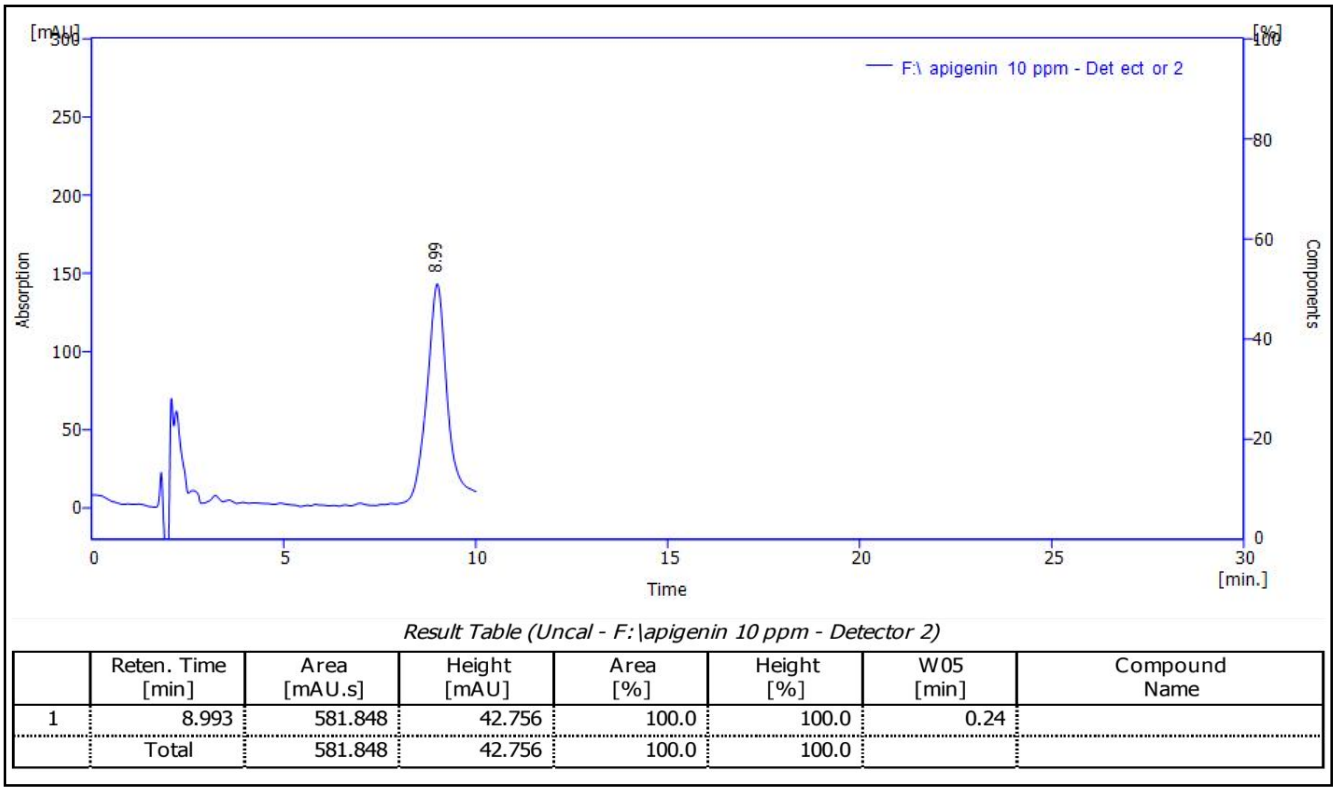
No.	Standard phenolic compounds	Standard Retension Time (min)	% Area
1	Apigenin	8.993	581.848
2	Catechine	7.480	502.578
3	Keampferol	3.793	7368.436
4	Rutin	7.083	691.278
5	Qucetine	6.137	3296.538

Table 3: Shows the results of separation of phenolic compounds in Gloeocapsa sp algae extract using HPLC technology.

Gloeocapsa sp.	Holding time (min)	Area% of ethanol ore extract	Area% for ethyl acetate extract	Concentration for ethanol raw extract ppm	Concentration of ethyl acetate extract ppm
Apigenin	8.860	2.7	7.0	13.6	10.6
Catechine	7.583	11.3	10.2	15.8	10.3
Keampferol	3.767	3.6	3.0	22.8	20.0
Rutin	7.203	8.1	7.1	16.0	13.5
Qucetine	6.150	15.9	14.2	20.7	11.5
Gallic acid	2.400	2.7	5.1	63.2	58.6
Coumarin	12.830	7.3	7.0	24.1	20.5

As well as, the HPLC technique for separating and purifying the raw ethanol extract and phenolic algae extract, that principle of it, device's work is by two phases, one is fixed and the other is moving, which leads to the appearance of curves and the area inside the curve is calculated for the sample to be separated and compared to the area inside Curved for standard sample. A number of phenolic compounds were separated and diagnosed from the ethanol ore extract as well as the phenolic extract, because the phenolic compounds are in the form of cyclosides within the algae cells, as shown in table 2 standard phenolic compounds and the standard retention time (minutes) and the area under the curve of the standard compounds and shows Fig. 5-11 standard curves for different phenolic compounds.

The Apigenin compound appeared in the crude extract and phenolic extract at 2.7 (and 7.0%), respectively, at a concentration of 13.6 (ppm and 10.6), respectively and the Catechine compound appeared at (11.3 and 10.2%), respectively, at a concentration of 15.8 ppm and 10.3, respectively and the Keampferol

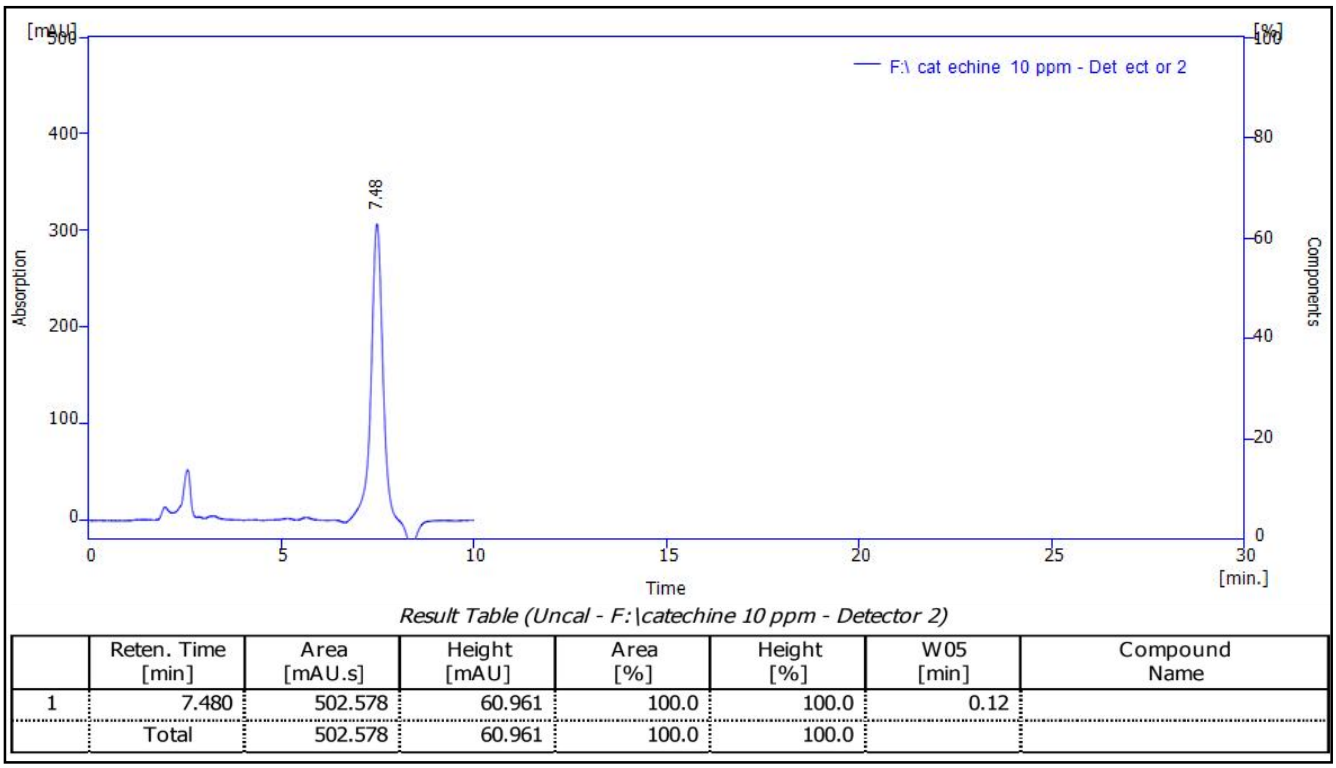


Result Table (Uncal - F:\apigenin 10 ppm - Detector 2)

	Reten. Time [min]	Area [mAU.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	8.993	581.848	42.756	100.0	100.0	0.24	
	Total	581.848	42.756	100.0	100.0		

Fig. 5: Standard Curve for HPLC-Designated Apigenin Compound.

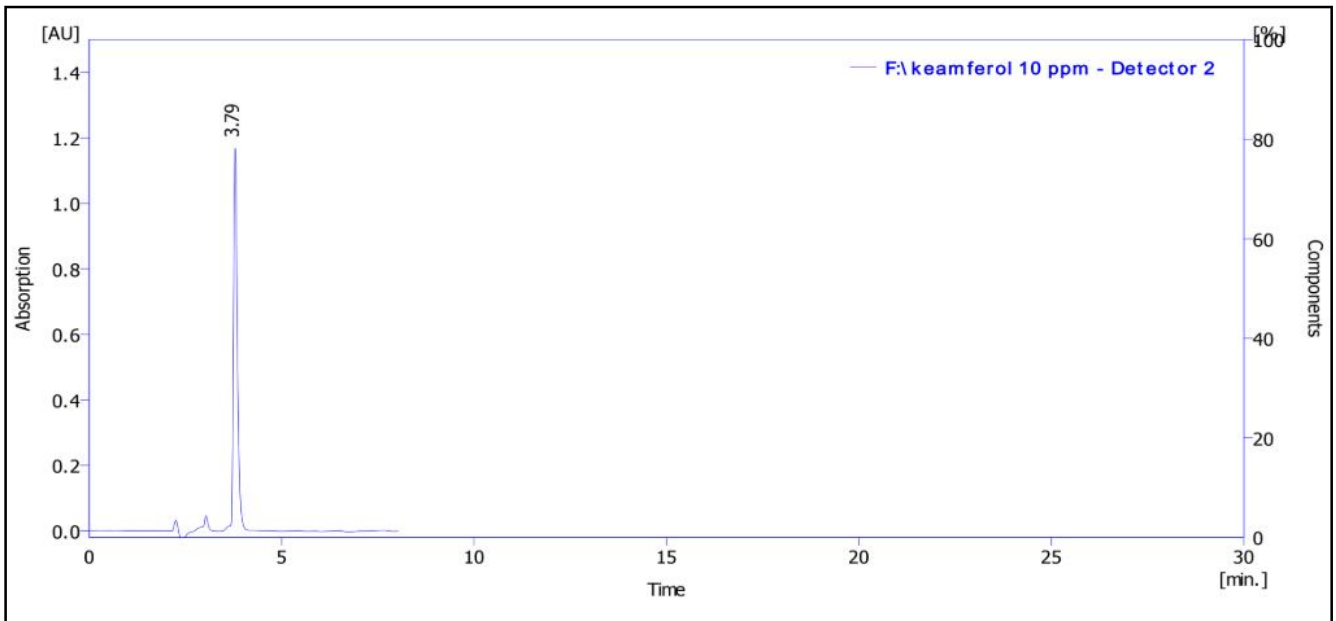
compound appeared in the ethanol crude extract and phenolic extract at a rate of (3.6 and 3.0%), respectively, with a concentration of (22.8 ppm and 20.0) respectively, as well as the Rutin compound appeared in the ethanol raw extract and phenolic extract at a ratio of (8.1 And 7.1%), respectively, with a concentration of (16.0 ppm and 13.5) respectively, and the Qurcetine compound appeared in the ethanol ore extract and phenolic extract



Result Table (Uncal - F:\catechine 10 ppm - Detector 2)

	Reten. Time [min]	Area [mAU.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	7.480	502.578	60.961	100.0	100.0	0.12	
	Total	502.578	60.961	100.0	100.0		

Fig. 6: Standard Catechine HPLC Curve.

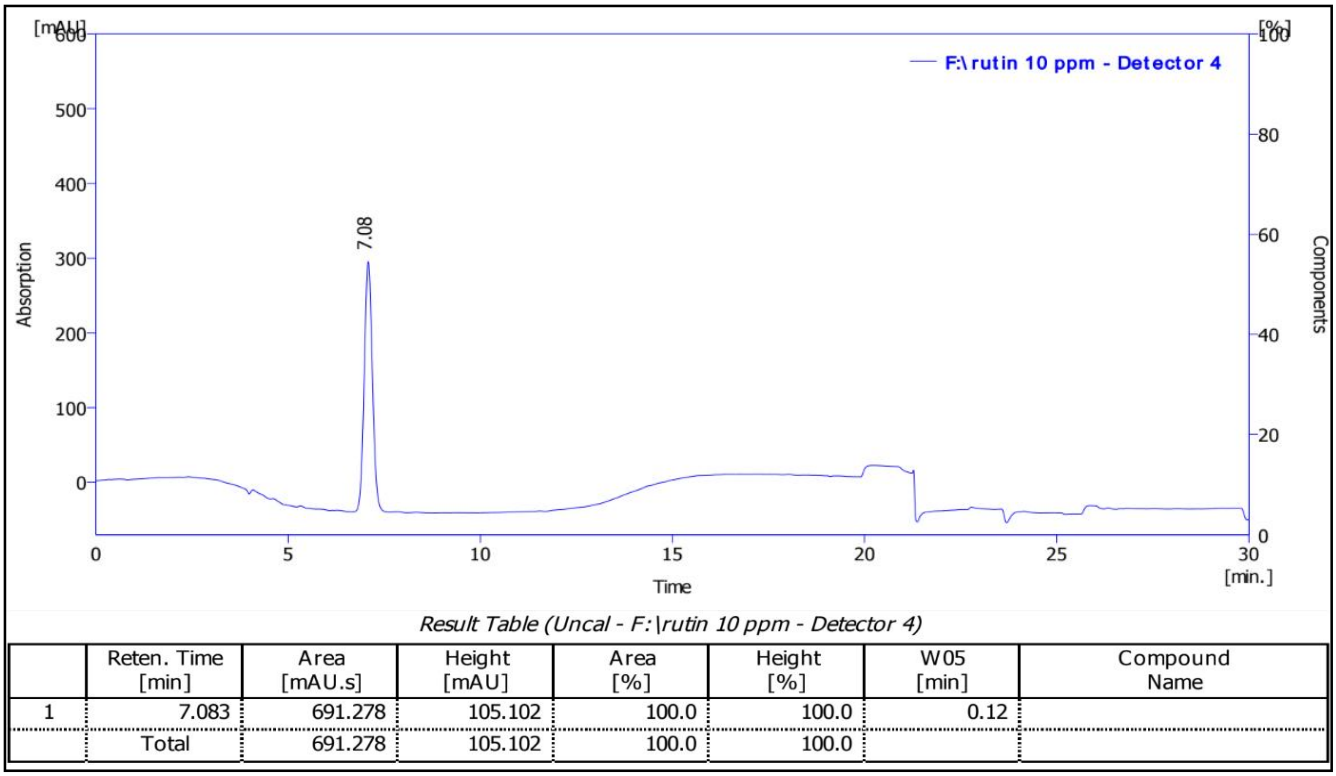


Result Table (Uncal - F:\keamferol 10 ppm - Detector 2)

	Reten. Time [min]	Area [mAU.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	3.793	7368.436	1121.103	100.0	100.0	0.11	
	Total	7368.436	1121.103	100.0	100.0		

Fig. 7: Standard Curve for HPLC Diagnosed Keampferol.

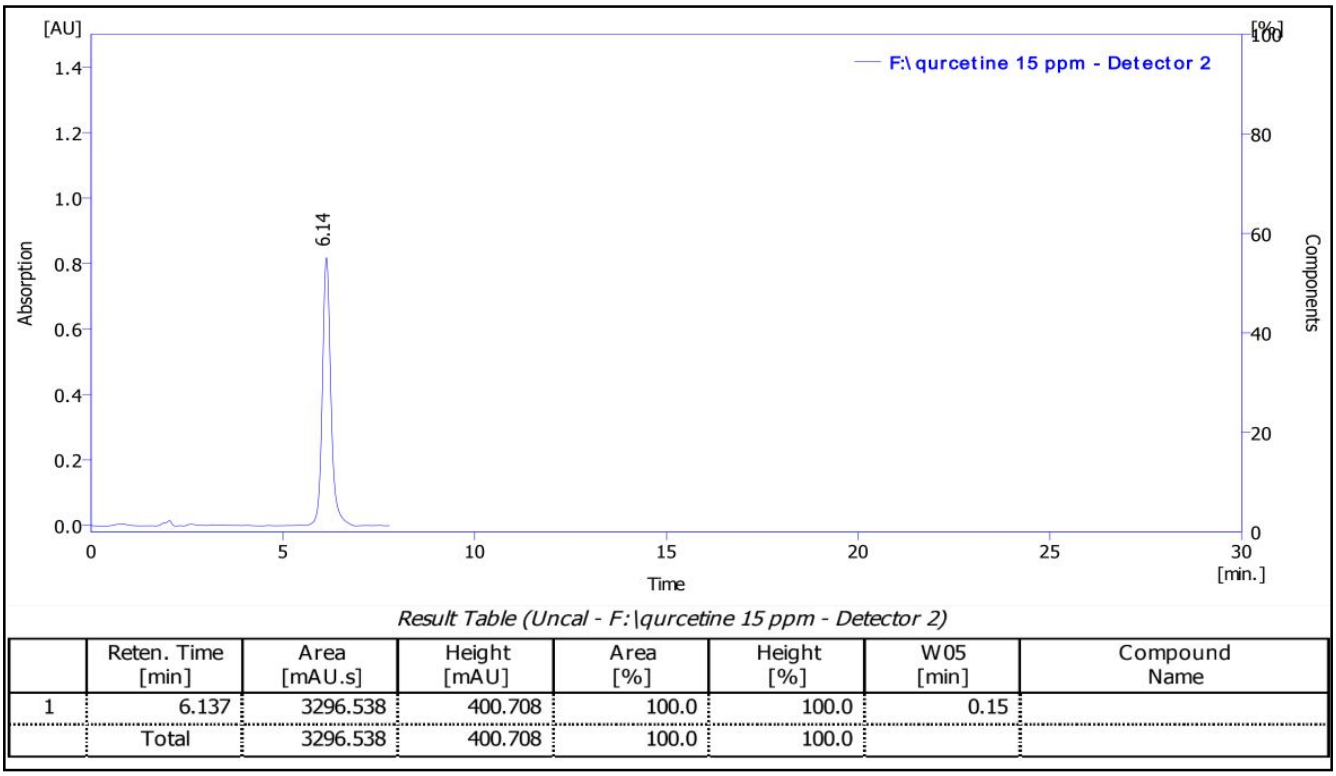
at a rate of (15.9 and 14.2%), respectively and at a concentration of (20.7 ppm and 11.5), respectively.Gallic acid also appeared in the ethanol ore extract and phenolic extract at a rate of (2.7 and 5.1%), respectively, with a concentration of (63.2 ppm and 58.6) respectively, and the coumarin compound appeared in the ethanol ore extract and phenolic extract at a rate of 7.3) and 7.0%), respectively, at a concentration of (24.1ppm and 20.5),



Result Table (Uncal - F:\rutin 10 ppm - Detector 4)

	Reten. Time [min]	Area [mAU.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	7.083	691.278	105.102	100.0	100.0	0.12	
	Total	691.278	105.102	100.0	100.0		

Fig. 8: Standard Curve for HPLC Diagnosed Rutin.



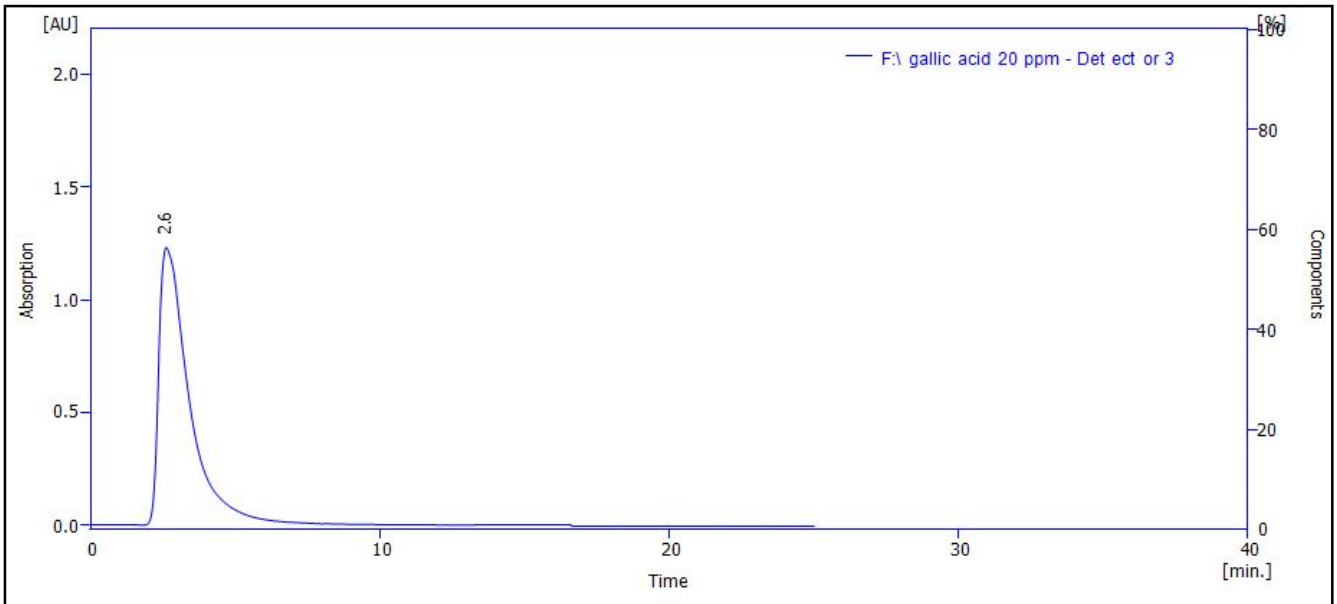
Result Table (Uncal - F:\qurcetine 15 ppm - Detector 2)

	Reten. Time [min]	Area [mAU.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	6.137	3296.538	400.708	100.0	100.0	0.15	
	Total	3296.538	400.708	100.0	100.0		

Fig. 9: Standard curve for HPLC diagnosed Qucetine.

respectively and Table 3 shows the retention time (min), the area% for the ethanol ore extract, the area% for the phenol extract, the concentration for the ethanol ore extract, ppm, and the concentration for the ethyl acetate ppm extract Diagnosed with HPLC for *Gloeocapsa* sp.

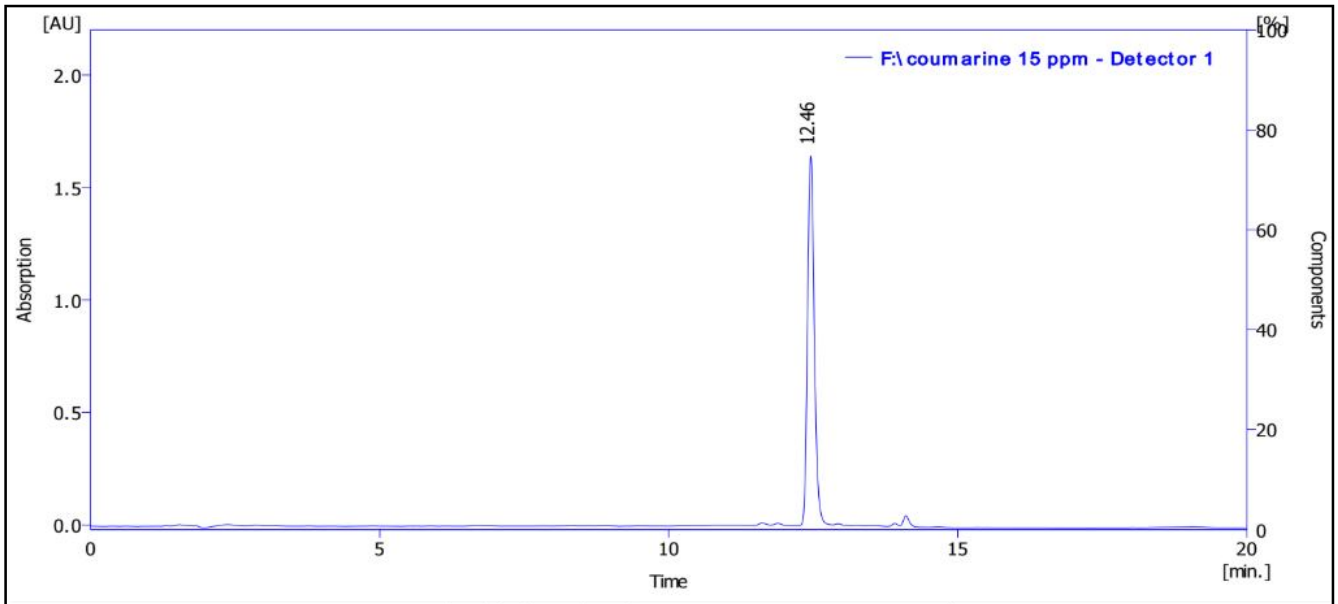
The phenolic content in algae products depends on the different extraction conditions used in terms of the type of solvent used for the extraction if it is water, methanol, chloroform, ethanol, hexane and ethyl acetate (Machu *et al.*, 2015), that in agreement with what was



Result Table (Uncal - F:\gallic acid 20 ppm - Detector 3)

	Reten. Time [min]	Area [mAU.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	2.627	1693.816	122.565	100.0	100.0	0.28	
	Total	1693.816	122.565	100.0	100.0		

Fig. 10: Standard Curve for Gallic Acid Designed with HPLC Technology.

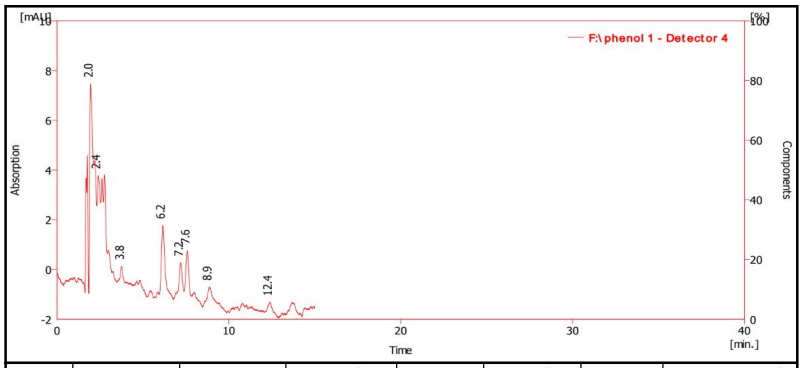


Result Table (Uncal - F:\coumarine 15 ppm - Detector 1)

	Reten. Time [min]	Area [mAU.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	12.457	9680.573	1387.952	100.0	100.0	0.11	
	Total	9680.573	1387.952	100.0	100.0		

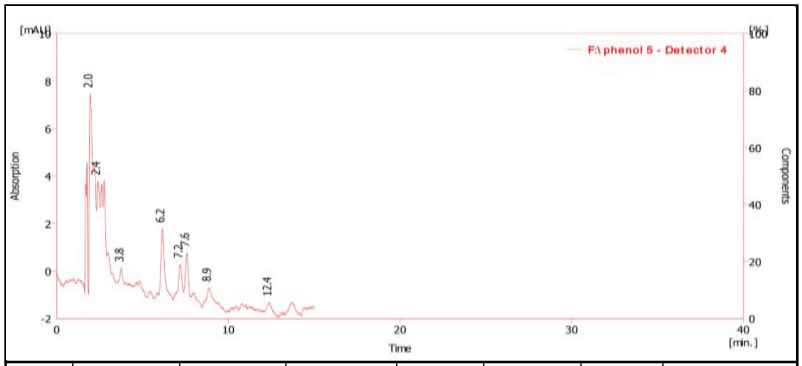
Fig. 11: Standard Curve for HPLC Diagnosed Coumarin.

reached (Pradhan *et al.*, 2014) ,that cyanobacteria and microalgae contain phenolic compounds and other active compounds, as these results agreed with the study (Al-Harbaoui, 2019) which refer to cyanobacteria contain different types of phenolic compounds, as well as agreed with (Singh *et al.*, 2017) which showed the raw extract of twenty species of cyanobacterial species have Keampferol, Qurcetine, Rutin, Gallic acid, thus the cyanobacterium can be considered as a new source rich in phenolic compounds (Xue and He 2015). In the antibacterial study, the results showed the efficacy of the ethanol extract and ethyl acetate extract on both the pathogenic Gram positive bacteria *Staphylococcus aureus* and Gram negative bacteria represented by *Esherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis* and *Pseudomonas aeruginosa*, the first (1) extract does not show any activity against all pathogenic that mentioned previously, While ethyl acetate extract (2) inhibited all bacteria in different diameters. Also, the solvents (as negative controls) 95% ethanol and ethyl acetate (3, 4) respectively, had no effect on all bacteria, and the highest inhibition occur on *Klebsiella pneumoniae* with an inhibition zone 25 mm and the least inhibition with bacteria *Staphylococcus aureus* (19 mm), as shown in table 10 and Fig. 14.



	Reten time [min]	Area [mA U.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	1.950	52.827	6.349	48.4	50.1	0.21	
2	2.400	2.987	0.705	2.7	5.6	0.09	Gallic acid
3	3.767	3.907	0.520	3.6	4.1	0.13	Keampferol
4	6.150	17.395	1.938	15.9	15.3	0.16	Qucetine
5	7.203	8.816	0.980	8.1	7.7	0.16	Rutin
6	7.583	12.385	1.395	11.3	11.0	0.16	Catechine
7	8.860	2.918	0.336	2.7	2.7	0.15	Apigenin
8	12.380	7.942	0.445	7.3	3.5	0.26	Coumarin
	Total	109.179	12.668	100.0	100.0		

Fig. 12: Shows the separated and diagnosed phenolic compounds from the crude ethanol extract *Gloeocapsa* sp.



	Reten time [min]	Area [mA U.s]	Height [mAU]	Area [%]	Height [%]	W05 [min]	Compound Name
1	1.950	52.827	6.349	48.4	50.1	0.21	
2	2.400	2.987	0.705	2.7	5.6	0.09	Gallic acid
3	3.767	3.907	0.520	3.6	4.1	0.13	Keampferol
4	6.150	17.395	1.938	15.9	15.3	0.16	Qucetine
5	7.203	8.816	0.980	8.1	7.7	0.16	Rutin
6	7.583	12.385	1.395	11.3	11.0	0.16	Catechine
7	8.860	2.918	0.336	2.7	2.7	0.15	Apigenin
8	12.380	7.942	0.445	7.3	3.5	0.26	Coumarin
	Total	109.179	109.179	12.668	100.0	100.0	

Fig. 13: Shows the separated and diagnosed compounds from the phenolic extract of *Gloeocapsa* sp.

This result agreed with (Al-Harbaoui, 2019; Shaieb *et al.*, 2014), that refer to the ethanol crude extract of cyanobacterium is not inhibiting the pathogenic bacteria used in the study. This result also came in line with the study of (Khairy and El-Kassas, 2010; Farghl *et al.*, 2019) that the ethyl acetate extract for cyanobacterial species has inhibited pathogenic bacteria and the sensitivity of different types of bacteria to algae extracts varies depending on the nature of the cellular contents and the composition of each type of bacteria in addition to the nature of the extract of different types of algae in terms of physiological and environmental factors, for example, the time in which the algae are collected, the method of their development and preservation in addition to the type of extract and the method of extraction (Qassim *et al.*, 2003; Tüney *et al.*, 2006) The effectiveness of the extracts on *Fusarium solani*, *Alternaria alternata* and *Candida albicans* was also studied, As the

raw extract (1) did not show any inhibition on all types of pathogenic fungi while the ethyl acetate extract (2) inhibited all types of pathogenic fungi with different diameters and the 95% ethanol and ethyl acetate (3, 4) respectively did not inhibit all fungi types and it was higher Inhibition of the fungus *Alternaria alternata* with a diameter of 42 mm and less inhibition of the fungus *Candida albicans* (18 mm) as shown in table 11 and Fig. 15.

This results agreed with a study (Shaieb *et al.,* 2014), whereby the raw ethanolic extract of some types of cyanobacteria inhibited certain types of fungi. Also, by genera of cyanobacteria inhibited for *Candida albicans* and other types of fungi was occured (Khairy and El-Kassas), 2010; Seddek *et al.,* 2019). The cause of fungi inhibition may be due to the location and number of hydroxyl groups on the phenol group are related to their relative toxicity to microorganisms and their effect on the fungal cell membrane that It changes its validity (Arif *et al.,* 2009).

Table 10: Shows the effect of raw ethanol extract and phenolic extract and control of algae *Gloeocapsa* sp. on pathogenic bacteria.

Pathogenic bacteria	raw Ethanol extract	Ethyl acetate extract	Ethanol 95% control	Ethyl acetate control
Staph. aureus	-	19	-	-
E.coli	-	21	-	-
K.pneumoniae	-	25	-	-
Pro. mirabilis	-	21	-	-
Pse. aeruginosa	-	21	-	-

Each value is a rate of three repeaters, (-) no inhibition.

Table 11: Shows the effect of raw ethanol extract and phenolic extract and control of algae *Gloeocapsa* sp. on pathogenic fungi.

Pathogenic bacteria	raw Ethanol extract	Ethyl acetate extract	Ethanol 95% control	Ethyl acetate control
Fusarium solani	-	25	-	-
Alternaria alternaria	-	42	-	-
Candida albicans	-	18	-	-

Each value is a rate of three repeaters, (-) no inhibition.

Conclusions

Microalgae (cyanobacterium *Gloeocapsa* sp. PCC 7428) produce phenolic compounds Apigenin, Catechine, Keampferol, Rutin, Qucetine, Gallic acid and Coumarin that diagnosed by TLC and HPLC. The efficacy of extracts against *Staphylococcus aureus*, *Esherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis* and *Pseudomonas aeruginosa* has also been studied, as well as against the pathogenic fungi *Fusarium solani*, *Alternaria alternata* and *Candida albicans*. As the raw ethanol extract did not show any inhibition against all types of bacteria and fungi, while the phenolic extract inhibited all types of bacteria and fungi.

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Fig. 15: Shows the effect of raw ethanol extract of *Gloeocapsa* sp. (1) and ethyl acetate extract (2) 95% ethanol (3) and ethyl acetate (4) on various pathogenic fungi.

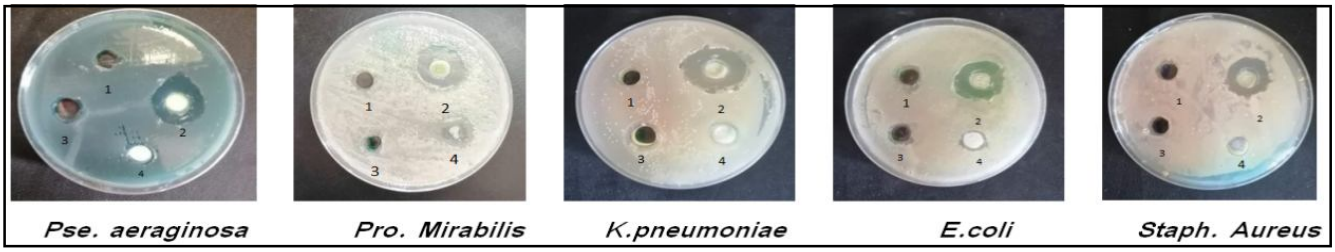

Fig. 14: Shows the effect of raw ethanol extract of *Gloeocapsa* sp. (1) and phenolic extract (2) 95% ethanol (3) and ethyl acetate (4) on different pathogenic bacteria.

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