



Received: 28-02-2023
Accepted: 08-04-2023

International Journal of Advanced Multidisciplinary Research and Studies

ISSN: 2583-049X

Study of the phenolic compounds of some species of malvaceae

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Abstract

This study was conducted to investigate the chemical and molecular relationships and compare them to the leaves of eight species, varieties and species belonging to the Malvaceae family in Iraq for the period from 10/8/2022 to 1/2/2023, as the following species were studied: *Abelmoschus esculentus* var. BATHRAH, *Abelmoschus esculentus* var. LAHLOUBA, *Abelmoschus esculentus* var. HUSSAINAWI, *Hibiscus sabdariffa* var. RED, *Hibiscus sabdariffa* var. STRIPED, *Malva parviflora*, *Althaea officinalis* var. RED, *Althaea officinalis* var. WHITE. Obtained from field trips and tours. The results of the chemical study determined the concentrations of six secondary (phenolic) compounds using High Performance Liquid Chromatography (HPLC) technology belonging to *Abelmoschus esculentus* L. They are (protocatechuic acid, catechin, Quercetin-3-o-gentobioside, Rutin, Isoquercetin, Quercetin). The highest concentration of the compound Quercetin-3-o-gentobioside, which reached (1.2 mg / g), and the lowest concentration of the compound Rutin, which reached ((0.35 mg / g). Also, five secondary (phenolic) compounds belonging to the type L *Malva parviflora* were identified (P-coumaric acid, Apigenin (Naringenin, Luteolin, Cinnamic acid), as the highest concentration of the

compound Naringenin reached ((1.4 mg / g in When the lowest concentration of the Luteolin compound was ((0.19 mg / g) and nine secondary (phenolic) compounds belonging to the species L *Althaea officinalis* were identified, which are (Gallic acid, Hydroxy benzoic acid, ferulic acid, Caffeic acid, Chrysin, P-coumarins, Vanillic acid, Ellagic acid, Syringic acid), where the highest concentration of the compound P-coumarins was (2.1) mg / g, and the compound with the lowest concentration was the compound Vanillic acid, which reached ((0.15 mg / g). Ten secondary (phenolic) compounds belonging to the *Hibiscus sabdariffa* species were identified. L are (Gossypetin, Sabdaretin, Gossypitin, Chlorogenic acid, (Hibiscetin, Rutin, protocatechuic acid, Eugenol, Quercetin, Kaempferol), as the highest concentration of the Quercetin compound reached ((1.4 mg / g) and the lowest concentration of the Eugenol compound reached (0.45) mg / On the basis of these compounds, it became clear that all varieties differed by the concentration of the separated compounds, and the chemical study proved its usefulness in distinguishing, separating, and finding evolutionary links between the studied varieties.

Keywords: Malvaceae, Phenolic Compounds, Sonicator Degaser

Introduction

Previously, the chemical study of plants included knowledge of chemicals with medicinal efficacy and how to extract them. Recent studies have become dependent on chemical studies in understanding the phylogenetic and phenotypic relationships between taxa, especially the study of chemical compounds formed as secondary metabolites such as alkaloids, flavonoids, phenolics, terpenes, saponins, and others. It can be recognized and diagnosed with high efficiency by means of modern scientific techniques and devices, and the method of building and distributing these compounds within the plant body has good taxonomic importance for distinguishing between taxonomic ranks, especially at the level of species belonging to one genus and difficult to separate and distinguish ^[1] The difference in chemical compounds in plants is almost similar to the difference shown by other characteristics such as morphological characters and anatomical characters. Including in several aspects, including pharmacological and industrial ^[2]. The secondary compounds are produced in varying amounts in plants, as they are thought to be produced for defensive purposes. Plants can use it to fight insects. It also plays a role in plant survival and adaptation to the environment in which it lives, and some plants need it to attract insects and produce volatile oils from flowers for pollination ^[3]. Compound and Volatile oils ^[4]. Among the chemicals known and used in the field of chemical classification, are the phenolic constituents that are commonly found in leaves, flowers, seeds, and all groups of green plants. It tends to dissolve in water, so it is often found in association with sugar, forming what is known as glycosides, which are usually found inside cell vacuoles ^[5]. Malvaceae plants are characterized by the presence of many active ingredients as ^[6] indicated. *Malva*

parviflora contains many flavonoids, gum, vitamin A, terpenes and polysaccharides. It also showed the presence of many flavonoids, phenols, alkaloids, saponins and tannins in the methanolic extract of leaves and stems of *Malva parviflora* and showed high antioxidants [7]. While a variety of phytochemicals and antioxidants were isolated from different cultivars of *Abelmoschus esculentus*, a high level of polyphenols and flavonoids responsible for antioxidant and other cumulative activities were obtained, so they can be used as food and medicine mainly to improve insulin sensitivity [8]. indicated that *Ab.esculentus* leaves contain some amino acids, vitamins, flavonoids, phenolic acids and many other compounds [9]. indicated that the *H.sabdariffa* plant is extremely rich in minerals and vital nutrients such as iron, copper, calcium, magnesium and manganese, and the preliminary phytochemical analysis showed the presence of some phytochemicals in the methanolic extract such as alkaloids, tannins, saponins, glycosides, phenols and flavonoids, and the result of their quantitative presence was as follows; Tannins (17.0%), saponins (0.96%), phenols (1.1%), glycosides (0.13%), alkaloids (2.14%), flavonoids 20.08%) [10].

Preparation of samples to estimate the concentration of phenolic compounds in them

I followed the method used by [11] to prepare the crude alcoholic extract for the purpose of injecting it into the (HPLC) device. 1.0 g of crushed leaves powder was taken and dissolved in 20 ml of pure hexane, to dissolve and precipitate the fat. After an organic layer is formed, it is dissolved. In a mixture of methanolic alcohol and deionized water, at a ratio of 20:80 ml, for the purpose of extracting its active components by means of an ultrasonic device (Branson sonicator) made in the USA for 25 minutes at a temperature of 25c After obtaining a solution, it is centrifuged at 7500 revolutions / min for 15 minutes. The pure filtrate is passed on a glass column filled with garcol (fine charcoal) for the purpose of limiting the dyes, then the resulting solution is concentrated by passing a stream of nitrogen to evaporate it, and the resulting solution is added to 1 ml. By adding methanolic alcohol, then it is passed through a fine filter whose pores do not exceed 2.5 mm to remove the residual fibers in it and to obtain a very high purity. The concentration of phenolic compounds was measured by injecting 20 microliters to estimate the concentration of their compounds. The quantitative calculation of each compound in the mixture was done by comparing the area of the measurement band of known concentration with the measurement of the required sample band and finding the concentration of each compound.

Quantitative determination of phenolic compounds in leaves of the species under study by high-performance liquid phase separation method

The process of estimating the content of flavonoid compounds in the leaves of the studied species was carried out using the (HPLC) device of the Green Hills Company in Baghdad, the type of device (Shimadzu, Japan), which is an advanced technique for measuring low concentrations of the compounds to be estimated. The concentration of the compounds was measured by injecting (20) A microliter of sample in a separation column by means of an injector (Injection Rheody – 7211) and an ODS (C18) separation column was used with dimensions of 3 um particle size (50

x 2.0 mm I.D), where 3 um = the size of the particles of the dissolved substance and 50 = the length of the separation column and 2.0 mm width of the separation column from the inside and the mobile phase solvent system was use 0.1% acetic acid in Diionized Water Solvent, acetonitrile Solvent B Using Linear Gradients from 0-100% B in 10 minute And in the descent of the samples through the separation column after it was filtered and the gases were expelled from it using the ultrasonic device, the Sonicator degaser, with a flow rate equal to 1.5 ml / min, and UV detection appears at a wave length of 280 nanometers, and an automatic control device was used. system controller and two Shimadzu model LC-10A Pumps, and the separation conditions are shown in Table (1-2). Standard laboratory solutions were used for the separated compounds, the concentration of which is (50) µg / ml, the retention time and the period of time indicated in the following tables. A curve was drawn for each sample by itself by the electronic computer. Measurement readings were taken that included the area under the curve and the time required for retention (disability). From the time of retention, the vehicles could be diagnosed by comparing them with the retention time of the standard sample and by applying the following equation: Unknown concentration = sample band area / standard band area x standard concentration x number of dilution times [12].

Results

The studied species and cultivars, which were collected from different regions, showed a wide and significant variation in terms of their containment and concentration of phenolic compounds, Flavonoid compounds. Chemical compounds, while other species recorded the loss of these compounds and vice versa.

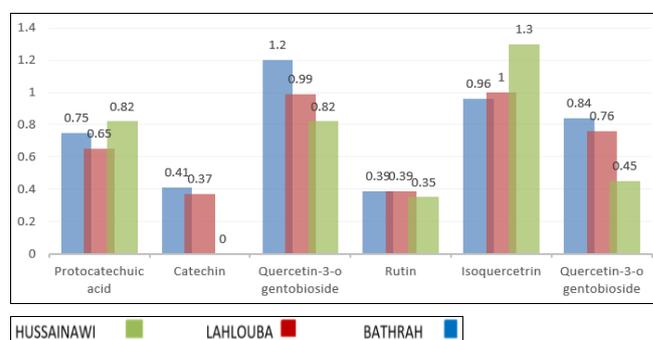


Fig 1: Showing the overlap between the concentration of phenolic compounds for the varieties of the species *Ab.esculentus*

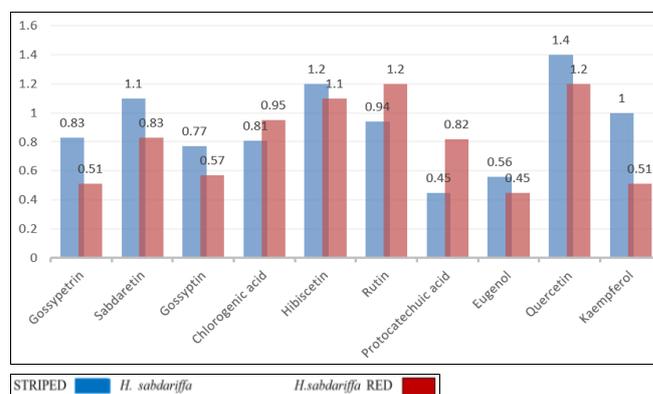


Fig 2: Shows the overlap between the concentration of phenolic compounds for *H. sabdariffa* cultivars

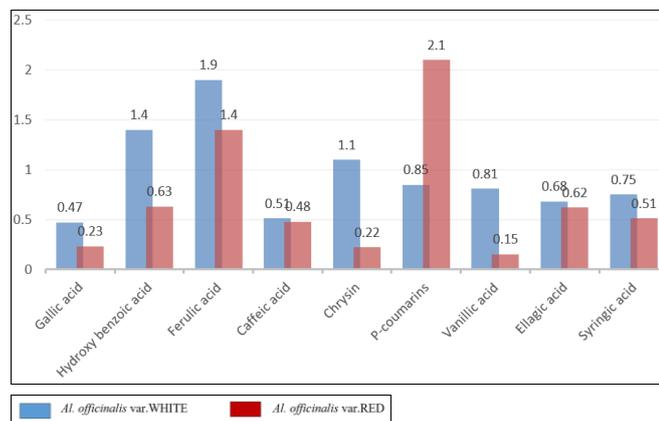


Fig 3: Shows the overlap between the concentration of phenolic compounds for Al-type cultivars. Officinalis

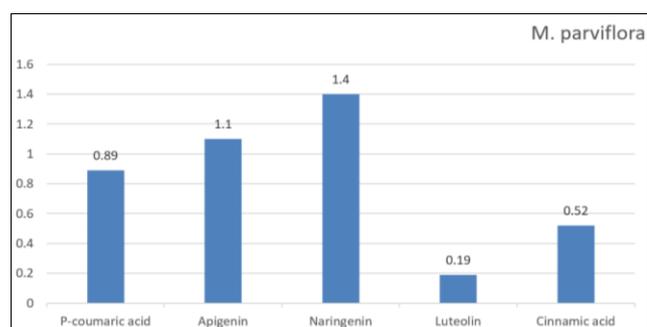


Fig 4: Shows the concentration of phenolic compounds in *M. parviflora*

Discuss the chemical results

The studied varieties of the Malvaceae family plants showed wide variations in terms of containing secondary metabolite compounds, which increased interest in them due to their medical and economic importance, in addition to the great taxonomic role they showed in separating the varieties (varieties) of the mallow family^[13].

The results of the chemical study, which resulted in the separation and identification of 30 phenolic compounds in all studied cultivars belonging to the family, using the high-performance liquid phase separation (HPLC) technique. Isoquercetin, quercetin and five compounds in baker are P-coumaric acid, Apigenin, Naringenin, Luteolin, Cinnamic acid and ten compounds in gujarat cultivars are Gossypetrin, Sabdaretin, Gossypitin, Chlorogenic acid, Hibiscetin, Rutin, Protocatechuic acid, Eugenol, Quercetin, Kaempferol, and nine compounds in seal rose cultivars are Gallic acid, Hydroxybenzoic acid, Ferulic acid, Caffeic acid, Chrysin, P-coumarins, Vanillic acid, Ellagic acid, Syringic acid The variation in the concentration and types of these compounds is one of the taxonomic evidence that occupies an important space in resolving the ambiguities and overlaps between the varieties of the same species that are difficult to distinguish between them except by using or seeking help from the results and many other studies, including chemical evidence, in agreement with the findings of^[14]. When using the anatomical, phenotypical, and chemical evidence in the separation and distinction between some plants of the mallow family in the city of Diwanayah, leading to a separation of 34 phenolic compounds, 31 terpene compounds, 29 alkani compounds, in addition to 13 alkaloid compounds, 9 esteric compounds, and 4 steroidal compounds.

The association of taxa with compounds supports their return to one species, while the variation shown by the taxa confirms the strength of their isolation as taxa belonging to a particular species, in agreement with the findings of^[15] on the great taxonomic importance in separating chemical compounds using modern scientific equipment, including GC technology -Mass in distinguishing between the types of the family, explaining the degree of kinship and divergence between the species, the biological activities of the compounds, and the chemical activity of each compound.

The same applies to *Hibiscus sabdariffa*, as the two cultivars shared the presence of the compounds Gossypetrin, Sabdaretin, Gossypitin, Chlorogenic acid, Hibiscetin, Rutin, Protocatechuic acid, Eugenol, Quercetin, Kaempferol, as the H. The lowest concentration ratio in the cultivar *H. sabdariffa* RED of Eugenol This is what the results of our study did not agree with what was presented by^[16] when they studied the separation and diagnosis of lignan compounds, where the phenolic compounds were separated, which are cinnamic acid, benzoic acid, vanillic acid, coumarin and thymol, and treated them as antioxidants on some types of bacteria. The study agreed with the findings.^[17] found that the aerial parts of *H. sabdariffa* contained a high concentration of secondary metabolites, including phenols The study agreed with all that was presented by^[18] in the presence of nine phenolic compounds in the leaves of this species, as well as the study agreed with^[19] revealed the presence of phenols, alkaloids and sterols in the leaves of this plant. This study also agreed with what was concluded by^[20] in the presence of a variation in the concentration of phenolic compounds between species of the genus, which can be considered as taxonomic evidence that supports phenotypic, cellular and environmental studies. *M. parviflora* leaves contained five phenolic compounds, p-coumaric acid, Apigenin, Naringenin, Luteolin, and Cinnamic acid, consistent with what was found by^[21] in the presence of the same phenolic compounds that were diagnosed by HPLC In the current study, three common compounds were found between *H. sabdariffa* and *Ab. esculentus* species, protocatechuic acid, quercetin, and rutin, which indicates a related relationship between them. Indicated^[22] that the increase in secondary metabolite products is related to the extent of the spread of the root system and the absorption of substances and elements that lead to the efficiency of the photosynthesis process. The increase in chemical compounds and the availability of good conditions for growth lead to an increase in photosynthesis and then an increase in the compounds stored in the plant. The proximity of plants From each other leads to an increase in competition between them and then an increase in chemical compounds^[23].

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