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A chemotaxonomical Study of Four Species Belonging the Genus *Arabis* L. Cruciferae (Brassicacae) in Iraq

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Abstract

This study has been focused on the detection of phenolic compound in aerial parts (stem and leaves)of the species belonging the genus ***Arabis*** L. *A.Caucasica* Willd, *A.Sagittata* (Bertol)DC, *A.Aucheri* Boiss, *A.nova* Vill of family cruciferou (Brassicaceae) which were collected during field trips of the districted of Iraq. Phenolic compounds have been identified by using High Performance Liquid Chromatography (HPLC). Nine standard compounds used for comparison five of them flavonoids (Rutin, Quercetin, kaempferol, Luteolin and Apigenin) and the other phenolic acid (Chlorogenic acid, Caffeic acid, Ferulic acid and Rosmarinic acid) , results showed that species vary in containing phenolic compound which can be counted as a taxonomic evidence supporting the taxonomic studied, this researchRosmarinicacid، Kaempferol، Quercetin، Rutin، Apeginin، Chloregenic acid: This compound exist in all species (stem and leaves part) is regarded as a first chemical study of genus *Arabis* L. in Iraq.

Keywords: *Arabis*.,flavonoids, phenolic acids and HPLC.

التصنيف الكيميائى لاربعة انواع من الجنس .Arabis L من العائلة الصلبية Brassicacae في العراق

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الخلاصة

الدراسة الحالية سلطت الضوء على المركبات الفينولية في الاجزاء الهوائية (الساق والاوراق)لاربعة أنواع من الجنس *A.Caucasica* Willd, *A.Sagittata* (Bertol)DC, *A.Aucheri* Boiss, *A.nova* Vill *Arabis* L العائد للعائلة الصلبية Cruciferae والتي جمعت خلال الجولات الحقلية لمقاطعات العراق . شخصت المركبات الفينولية باستخدام جهاز الكروماتوغرافي السائل عالي الاداء HPLC باعتماد تسعة من المركبات القياسية خمسة منها مركبات فلافونية وهي (Rutin, Quercetin, kaempferol, Luteolin and Apigenin) والباقية أحماض فينولية وهي

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(Chlorogenic acid, Caffeic acid, Ferulic acid and Rosmarinic acid) . وقد أظهرت النتائج
ان الانواع تغايرت في أحتوائها من المركبات الفينولية كما ان بعض المركبات مثل Rosmarinic Acid
كانت غير موجودة في Chloregenic acid، Apeginin، Rutin، Quercetin، Kaempferol،
الانواع المدروسة . والتي يمكن عدها كدلائل تصنيفية تدعم الدراسات التصنيفية الاخرى هذا وتعد الدراسة
الحالية هي الاولى من نوعها في دراسة هذا الجنس من الناحية الكيميائية.

Introduction

The genus Arabia L. was a largest with about 160 is a largest genera (Megar genera) about 160 species [1]. Arabis (Rosk cress) was grown as ornamentals [2]. Is about 120 species in temperate urope and asaia ، north America and mountens ،in Iraq tropical Africa .، four species [3] chemotaxonomy was used in classification of plants depending their chemical contain. During metabolism processes plant produce secondary metabolites, the type of these compounds is often specific and useful in classification [4].

Brassicaceae (cruciferae) was rich in glucosinilate (mustard-oil glucoside) and often cyanogenic but without iridoid substance, mostly not tanniferouse lacking both ellagic acids and pranthyocyanins sometimes present in seed coat and only seldom with alkaloids, cardenoides, present in all organs [5]. The common oil yielding species are ; mustard sarson (***Brassica compester***var.sarson), white muastard, safe sarson(***Brassica alba***), Indian mustard, Rail (***Brassica juncea***), Rape seed, toria (***Brassica napus***), Taramir (***Erucasative***) [6,1 & 2]. Most USA cruciferae plants ,contain sulpher compound, the seeds of many plant yield vegetable oil of multipurpose use [7].

Many types of natural product, found in plants , these products different , these compounds differ from one taxonomic group to another. Chemosystematics often divided the vast array of natural plant products in to tow major group according to molecule size micro molecules and macromolecules [8]. Phenolic compounds are constituted in one of the biggest and widely distributed groups of secondary metabolites in plant.[9].In Iraq there are many studied in chemotaxonomy like [10- 16].

Materials and methods

Plant extract preparation

Of each species used more than 25 specimens, 2 gm taken from shoot apical portion and leaves of all species.

The weight was put inside a conical flask , then add 50ml ethanol %70 for 48 hours.

After that the separation of samples were done by using filter paper medium size pores (110mm).

Stayed in the room temperature under dry air current to concentrate the sample by evaporation of Ethanol to extremely half of volume.

Add petroleum ether which a boiling point (80-100^{o}C).

The mixture put inside the separating funnel and left until separated in to two distinguished layers.

The lower layer is taken that contain the extract of plant.

The samples were leaved under dry current until it reach of half volume [17].

HPLC analysis

All chemical reagents used for analysis HPLC were analytical or HPLC Grade (99.99%). HPLC-grade methanol and formic acid were purchased from Scharlauchemie, S.A., Europian Union. Ultra-pure water was used for sample preparation and preparation of mobile phases for HPLC analysis. Caffeic acid, chlorogenic acid, ferulic acid, rosmarinic acid, kaempferol, quercetin, rutin, luteolin and apigenin as a standard material was purchased from Chroma Dex, USA.

Preparation of Standard Solutions

Caffeic acid, chlorogenic acid, ferulic acid, rosmarinic acid, kaempferol, quercetin, rutin, luteolin andapigenin (1 mg), were accurately weighed into a 5 ml volumetric flask, dissolved in methanol: water (1:1).

Chromatographic condition

The qualitative analysis of caffeic acid, chlorogenic acid, ferulic acid , rosmarinic acid, kaempferol,quercetin, rutin, luteolin and apigenin were performed on Knauer HPLC instrument equipped with ChromGate HPLC software provided by Knauer was use with Eurospher 100, C18 column (4.6 mm i.d. x 250 mm, 5 mm) and UV/Visible detector. The flow rate of the

mobile phase were kept at 1 ml/min. Mobile phase A was methanol and B water containing (1% formic acid) isocratic conditions were as 25 % A and 75 % B. The temperature of column was controlled at 30 °C. Injection volume was 20 µl, and the detection wavelength was set to 350 nm.

Results and Discussion

This study is regarded as a first chemical study of the genus ***Arabis*** in Iraq by. Using nine diagnostics compounds belonging to phenolic compounds, four of them flavonoids and the other phenolic acids, as standards, the following results were gotten:

- **Phenolic acid**

A. Caffeic acid: This compound found in the stem of A.Caucasio an A.auchar leaves exist in all species (stem and leaves part)except in stem of ***A.caucasia*** and ***A.auchari*** leaves.
B. Chloregenic acid: This compound exist in all species (stem and leaves part) except in ***A. aureculata*** and ***A. nova*** (stem and leaves).
C. Ferulic acid: presented only in ***A.caucasia***leaf and ***A. aureculata***stem.
D. Rosmarinic acid: not found in any species.

- **Flavenoids**

E. Kaempferol: not found in any species.
F. Quercetin: not found in any species.
G. Rutin: not found in any species.
H. Luteolin: found only in ***A.auchari*** (stem and leaves).
I. Apeginin:not found in any species.

Table 1- Retention times for Phenolic acids and Flavonoids that used in this study by HPLC method.

Compound number	Name of compound	Retention time
1	Caffeic acid	2.1
2	Chlorogenic acid	1.9
3	Ferulic acid	2.6
4	Rosmarinic acid	2.4
5	Kaempferol	8.4
6	Quercetin	4.3
7	Rutin	2.7
8	Apigenin	7.3
9	Luteolin	6.3

Table 2- distribution of phenolic acid and flavonoids in studied species.

Ethanolic extract	Standard phenolic acid				Standard flavonoids				
	Caffeic acid	Chlorogenic acid	Ferulic acid	Rosmarinic acid	Kaempferol	Quercetin	Rutin	Luteolin	Apigenin
A. caucasia leave	+ve	+ve	+ve	-ve	-ve	-ve	-ve	-ve	-ve
A. caucasia stem	-ve	+ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
A. aureculata leave	+ve	-ve	+ve	-ve	-ve	-ve	-ve	-ve	-ve
A. aureculata stem	+ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve

A. aucheri leave	**-ve**	**+ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**+ve**	**-ve**
A. aucheri stem	**+ve**	**+ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**+ve**	**-ve**
A. nova leave	**+ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**
A. nova stem	**+ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**	**-ve**

HPLC analysis were carried out to evaluate the chemical content of phenolic acid and flavenoids. the species showed a good content of phenolic acid especially (Caffeic acid and Chloregenic acid),this explain that the presence of the phylogenic associated relationship among the studied ***Arabis*** species in their chemical content, Ferulic acid found only in leaf of ***A.caucasia***and ***A.aureculata*** stem, this means it was less distributed between studied species, finally there have no any relation between the studied ***Arabis*** species in Rosmarinic acid because did not found in any plant parts of species, Table- 2.

According to the content of the Flavonoids compounds there have no any relation between ***Arabis*** species and flavonoids except Luteoline that found in ***A.auchari*** stem and leaves part Table- 2.

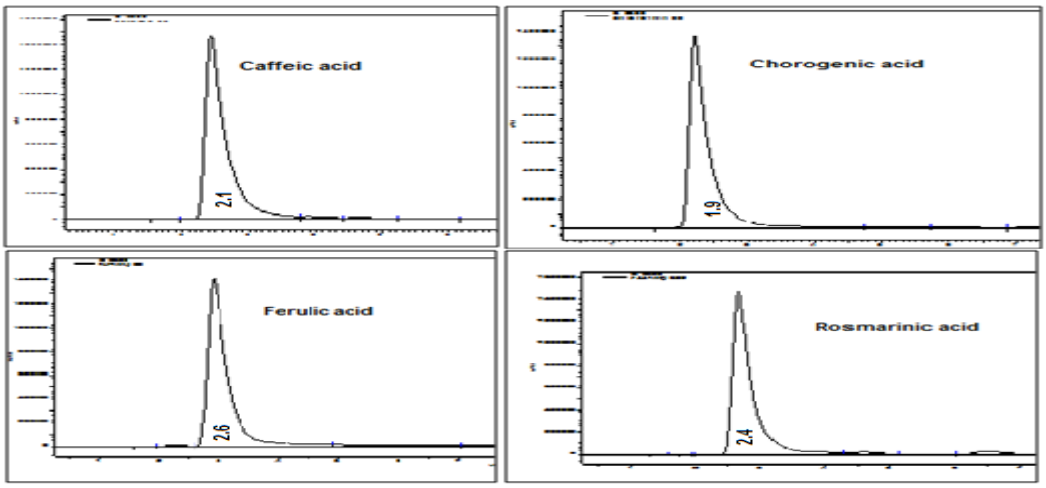


Figure 1- Chromatogram of standard phenolic acids.

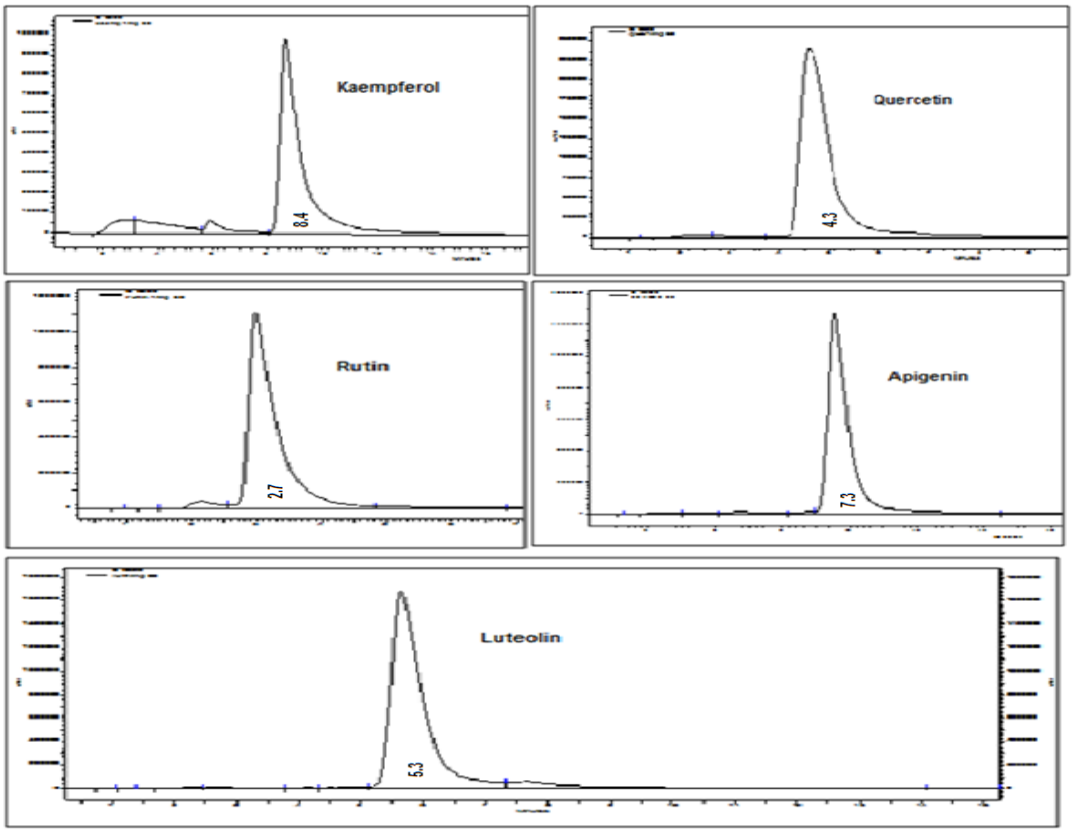


Figure 2- Chromatogram of standard flavonoids.

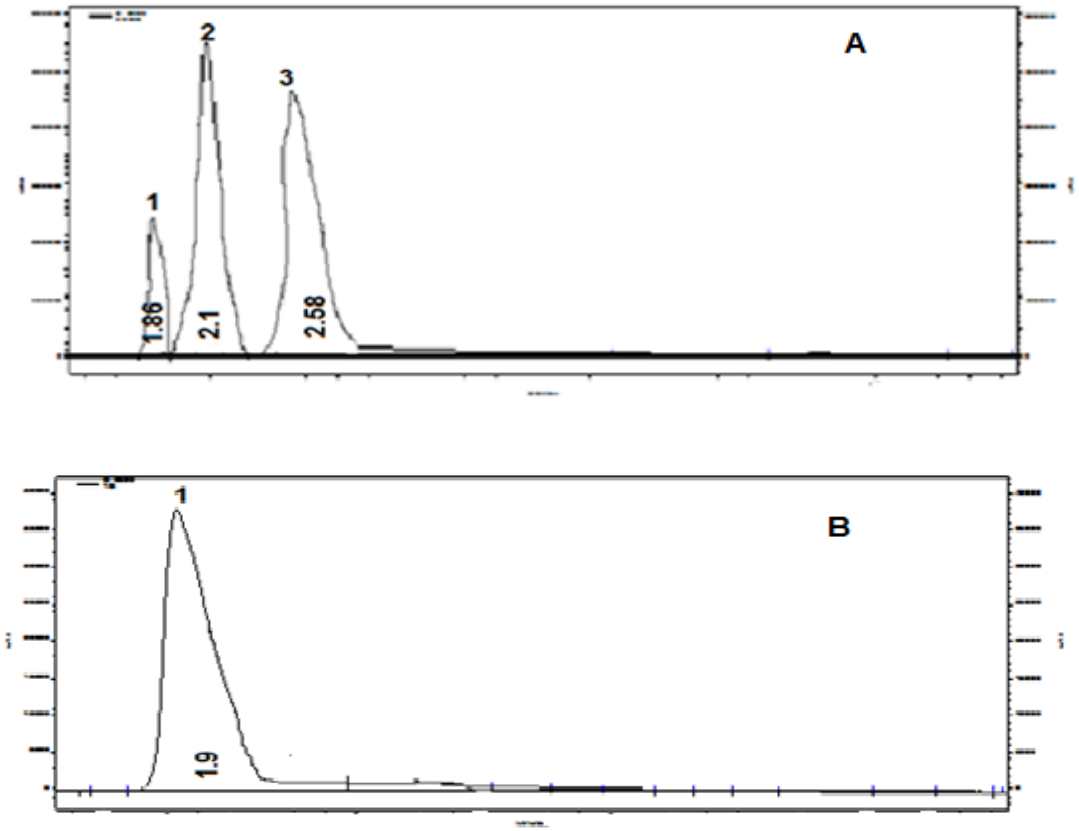


Figure 3 - Representative chromatogram of ethanolic extract of *Arabiscaucasia* A-leaves, B-Stem extract 1-Chlorogenic acid; 2-Caffeic acid; 3-Ferulic acid

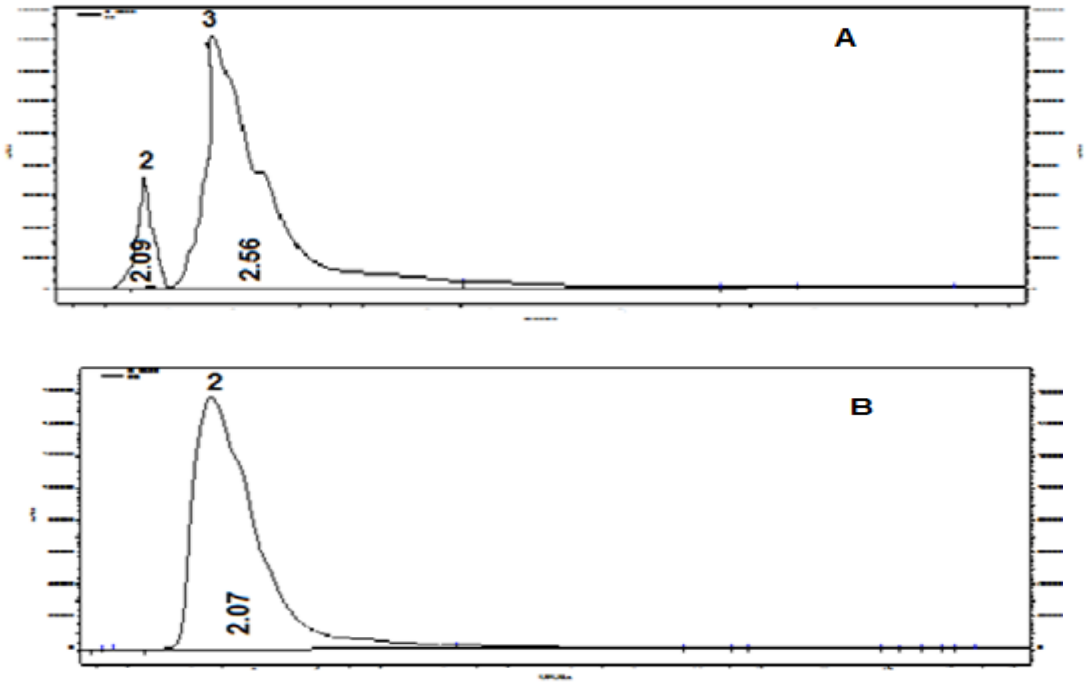


Figure 4 - Representative chromatogram of ethanolic extract of *Arabisaureculata*A-leaves, B-Stem extract 2-Caffeic acid; 3-Ferulic acid.

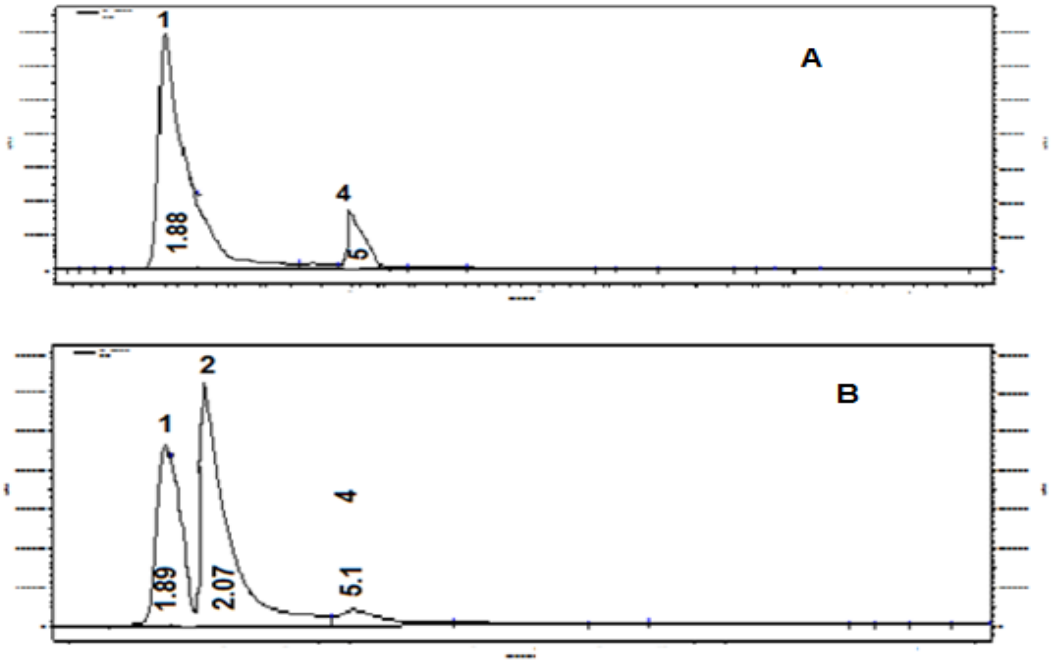


Figure 5 - Representative chromatogram of ethanolic extract of *Arabisaucheri*A-leave, B-Stem extract 1-Chlorogenic acid; 2-Caffeic acid; 4-Luteolin.

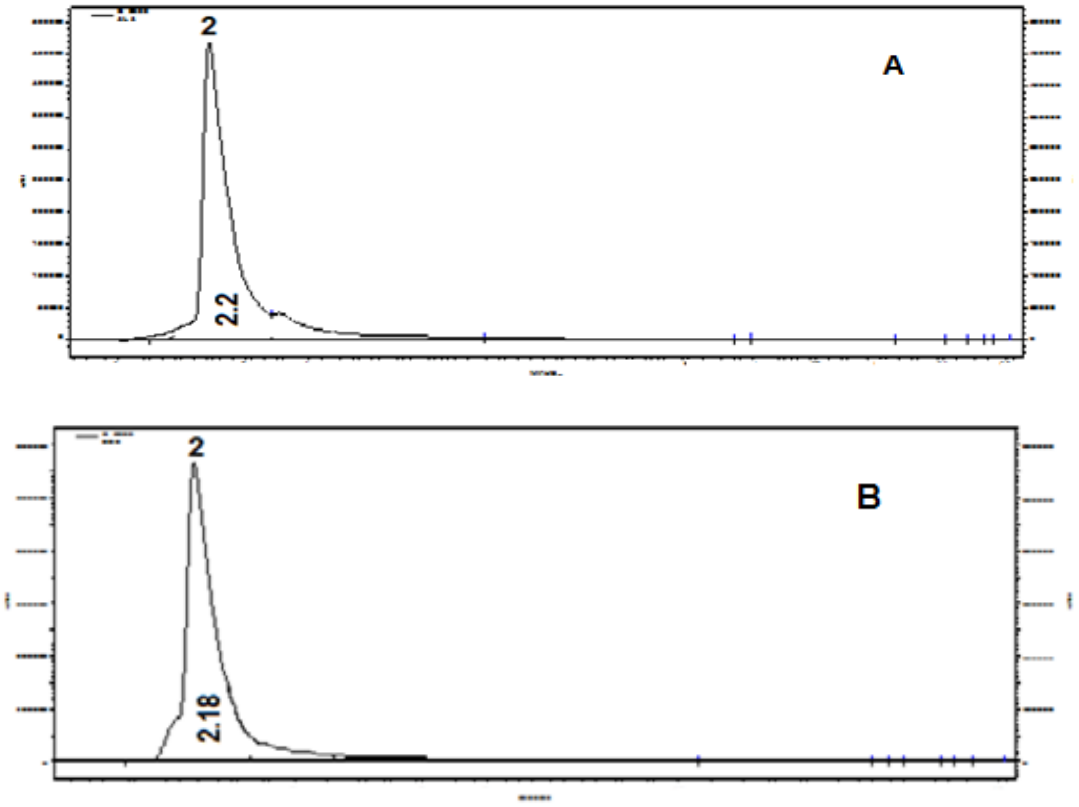
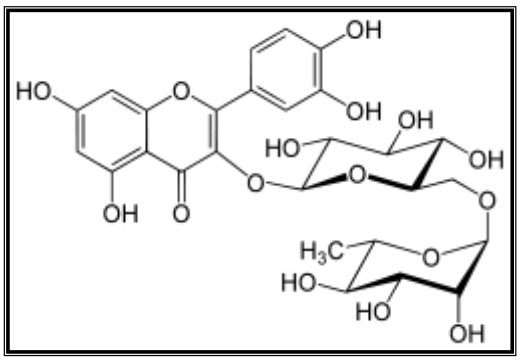
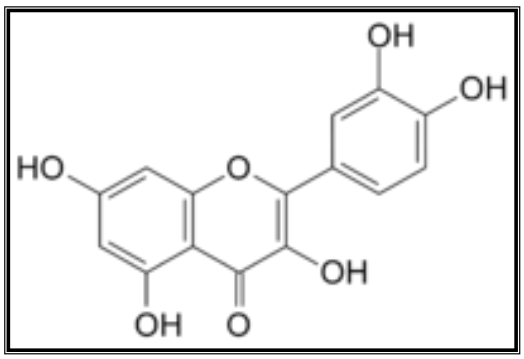


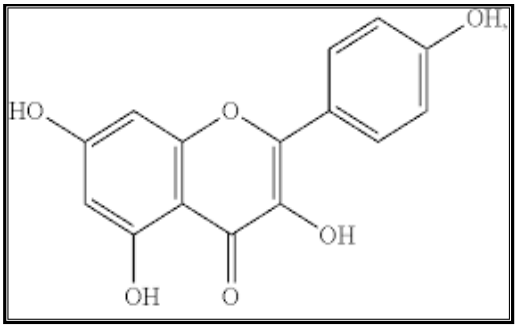
Figure 6 - Representative chromatogram of ethanolic extract of *Arabis nova*A-leave, B-Stem extract 2-Caffeic acid.



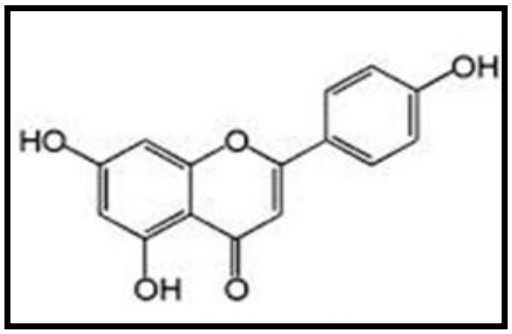
Rutin



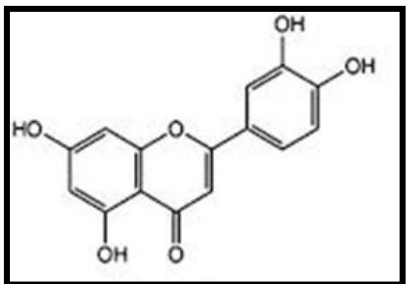
Quercetin



Kameferol



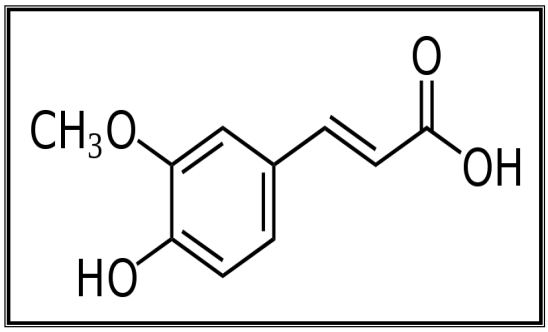
Apeginin

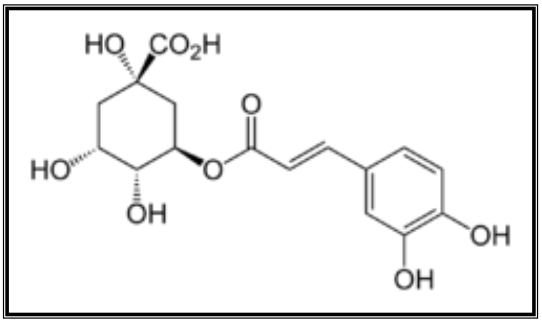


Luteoline

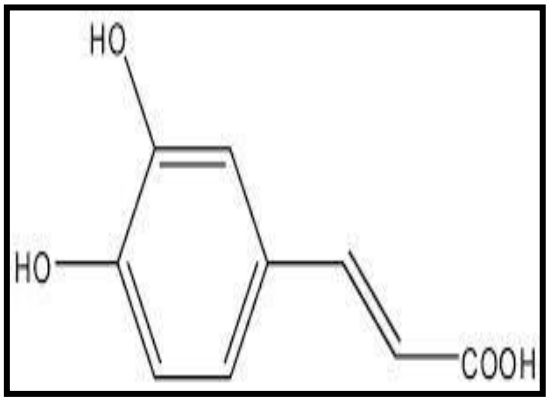
Plate 1- The structure of standard flavnoids*.

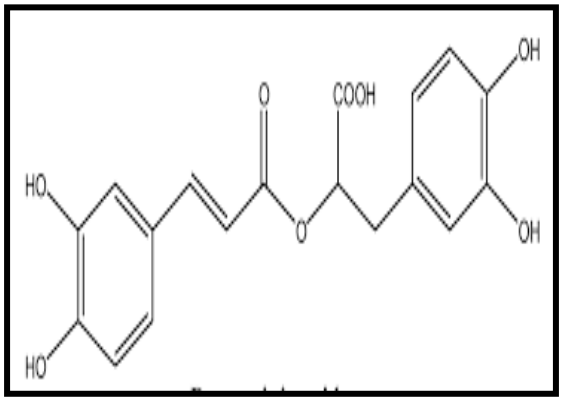
*****the structures taken from [18].





Chlorogenic acid





Caffeic acid **Rosmarinic acid**

Plate 2- The structure of standard phenolic acid compounds*.

* the structures taken from [18].

References

1. Singh, G .**2004**.*plant taxonomy on integrated approach science* puplisher INC USA
2. Singh R. Pande, P.C., Jain,D.K. **2008**. *A Text book of botany (Angio-sperms).* Rastogi publications india
3. Guest , E.**1980**. *Flora of Iraq* .vol 4 part.2.ministary of Agricalture of iraq .
4. Ankanna S., Suhrulatha D., Savithramma N. **2012**., Chemotoxonomical studies of some important Monocotelydon. *Botany Research international*, 2012; 5:90-96.
4. Croncquest, A. **1981**. *An integrated system of classification of flowering plants*, Columbia University press, Newyork.
5. Kumer, S**. 2004.** *Atextbook of plant anatomy*. Compus books international, NewDalhi, India.
6. Pandy, B. P**. 2005**. *Taxonomy of Angiospermae*. Chand and Company Ltd, NewDalhi, India.
7. Stuessy, T. F. **1990**. *Plant Taxonomy*. Columbia Univ. Press, NewYork , United State of America .
8. Scalbert, A. and Williamson G. (2000) Dietary intake and bioavailability of polyphenols. *Journal of nutrition*, 130, 2073S-2085S.
9. Meran , S. H. S. **2009 .** Asystematic study of the Genera Biebersteinina Stephan and Geranium L.(Geraniaceae) in Iraq Kurdistan Region Ph.D Thesis Biology Dep. University of Sulaimani.
10. Faqe-Mahmood, S. A**. 2009**. Systematic study of the genera Cephalaris (L.) Schrad. and Dipsacus L. Dipsacaceae in Kurdistan Region of Iraq .MSc.Thesis Bio. Dep. University of Sulaimani.
11. AL_Jbouri, Kh. Dh. W. **2010**.Acomparative systematic study of Nepeta L.spp (Labiatae)in Iraq.

12. Al-Dalawi, M. T. R. **2011**. Systematic study of the genus Prangos (L.) Lindl. Umbelliferae in Kurdistan Region of Iraq , MSc Thesis , Bio. Dep .University of Salahaddin.
13. Bakr , B. Kh. **2011.** Morphological-systematic study of the genera Scandix L. and Smyrnium L.(Umbelliferae) Apiaceae in Kurdistan Region of Iraq, MSc.Thesis, Bio-Dep. University of Salahaddin.
14. Al-Doskey, Z. A. S. **2014.** A comparative systematic Study of the Genera Rosa L. and Rubus L.(Rosaceae) in Kurdistan Rigion of Iraq .Ph.D thesis Forestry Dep. University of Dohuk .
15. Yonus,Th. F. **2016.** A systematic study of the genus Isatis L.(Brassicaceae) in Kurdistan Region of Iraq , MSc Thesis Agriculture College . University of Salahaddin(in English).
16. Harborne, J. B. **1973.** *Phytochemical method, AGuide to Modern Techniques of plants analysis.*
17. Goodwin and Mercer. **1983.** Introduction *to plant biochemistry*, 2nd edition, pergamon Press Ltd.