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Chemical Screening, antioxidant activity, and Mineral Profiling of *Rosmarinus Officinalis* L. from Msallata Region (Libya)

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ABSTRACT

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Keywords: Medicinal plants, *Rosmarinus officinalis* L., antioxidant activity, phytochemicals, minerals.

This study aimed to investigate the phytochemical constituents, total phenols, total antioxidant, and mineral contents of *Rosmarinus Officinalis* L. The aerial parts of the plant were extracted using different solvents, including water, ethanol, ethyl acetate, and chloroform. The extracts were analyzed for their phytochemical constituents. The total phenolic content and total antioxidant capacity were measured using colorimetric assays. The mineral content of the plant was analyzed using AAS. The results showed that the plant extracts were rich in carbohydrates, proteins, phenols, alkaloids, flavonoids, tannins, glycosides, and coumarins. The total phenolic content of the plant extracts ranged from 76.7 to 95.5 mg GAE/g, indicating strong antioxidant activity, with a total antioxidant capacity of 50.03 mg/g. The plant was found to be rich in macro elements such as Na, K, Ca, Mg, and P, with concentrations ranging from 1760 to 12155 mg/kg. While the concentrations of heavy metals (Fe, Zn, Cu) ranged from 56.19 mg/kg to 14.38 mg/kg. The water extract contained fewer phytochemical constituents compared to the ethanolic extract, while the ethyl acetate and chloroform extracts showed the presence of only specific compounds. These findings suggest that *Rosmarinus Officinalis* L. is a valuable natural source of bioactive compounds and essential nutrients. Further research is needed to fully elucidate the therapeutic properties of the plant and to explore its potential use in the prevention and treatment of various diseases.

1 Introduction

Throughout history, herbs and plants have been utilized for a multitude of purposes, including medicine, pharmaceuticals, nutrition, food preservation, flavorings, beverages, repellents, fragrances, and cosmetics (Sarker & Nahar, 2022). They were the basis for medicinal therapy until the development of synthetic drugs in the nineteenth century (Verma *et al.*, 2021). In recent decades, there has been a renewed interest in the use of herbs and plants due to their natural bioactive compounds, such as polyphenols, vitamins, polysaccharides, and minerals, which offer various beneficial activities (Elsherif *et al.*, 2023; Costa *et al.*, 2021). With their mild features and low side effects, the use of natural compounds is increasing worldwide. Herbal-based cosmetic preparations have also become

popular among consumers due to their non-toxic nature and strong activity (Javed & Shoaib, 2021).

Rosemary, scientifically known as *Rosmarinus officinalis* L., is a fragrant and perennial plant that belongs to the Lamiaceae family and originates from the Mediterranean region, but it is now widely distributed worldwide. This shrub-like plant can grow up to two meters high and is characterized by its green leaves that emit a pleasant aroma. Rosemary is versatile and can be used as a spice in cooking, a natural preservative in the food industry, and as an ornamental and medicinal plant (Silva & Dias, 2022; Khalil *et al.*, 2021). Rosemary (*Rosmarinus officinalis*) has been traditionally used for its medicinal properties, including antispasmodic, diuretic, antirheumatic, and antiepileptic effects.

Moreover, it is effective in treating respiratory problems, skin infections, and promoting wound healing (Al-Snafi, 2021) and (Mocan *et al.*, 2021). Rosemary (*Rosmarinus officinalis*) contains various bioactive compounds, such as flavones, diterpenes, steroids, and triterpenes. Among these compounds, the antioxidant activity of rosemary extracts has been mainly attributed to two phenolic diterpenes, carnosic acid and carnosol (Silva & Dias, 2022; González-Trujano *et al.*, 2021; Naveed *et al.*, 2021). The chemical composition and biological activities of rosemary has attracted the attention of many research works. (Silva & Dias, 2022) conducted a comprehensive review of *Rosmarinus officinalis* L., highlighting its chemical composition, biological activities, and applications in food preservation. The authors (Silva & Dias, 2022) reported that rosemary contains a high level of antioxidants, particularly phenolic compounds such as carnosic acid and carnosol, which contribute to its potent antioxidant activity. Total phenolic content and mineral determinations of rosemary extracts were also found to vary depending on the extraction method and plant part used. In addition, phytochemical screening of rosemary revealed the presence of various bioactive compounds, including flavonoids, triterpenes, and steroids (Sakar *et al.*, 2021). The review also discussed the potential uses of rosemary in food preservation due to its natural antimicrobial properties, which can inhibit the growth of foodborne pathogens and extend the shelf life of food products. Overall, the findings suggest that rosemary is a promising natural source of antioxidants and could be utilized in various applications, including food, pharmaceuticals, and cosmetics (Silva & Dias, 2022).

This study aimed to identify the bioactive compounds present in four *Rosmarinus officinalis* L. extracts (water, ethanol, ethyl acetate, and chloroform) through qualitative phytochemical screening. Additionally, the study sought to determine the moisture and ash contents, as well as the levels of total phenols and antioxidants in the plant. The concentrations of potassium, sodium, calcium, magnesium, phosphorus, copper, iron, and zinc in the herbal plant sample were also measured using flame photometry and flame atomic absorption spectrometry.

2 Materials and Methods

2.1 Study Area

The *Rosmarinus Officinalis* leaves were harvested from the Msallata region of Libya.

2.2 Chemicals and Equipment

The study utilized solvents such as water, ethanol, chloroform, and ethyl acetate for extraction, as well as various reference materials and reagents including ascorbic acid, gallic acid, sodium phosphate, ammonium molybdate, and Folin–Ciocalteu reagent, which were all obtained from the Sigma-Aldrich distributor. All the

chemicals, reagents, and solvents used in the study were of analytical grade unless otherwise specified. In terms of equipment, the study employed a rotary evaporator (Hei-VAP, Heidolph, Schwabach, Germany), a UV-Vis spectrophotometer (6300, Jenway), an Atomic Absorption System (AAS) (Varian 220 FS), and a Flame Photometer (PFP7, Jenway) for analysis purposes.

2.3 Sample Collection

The *Rosmarinus Officinalis* leaves were harvested from the Msallata region of Libya, during January, February, and March 2021. To ensure their authenticity, the leaves were authenticated by a botanist at the Faculty of Arts and Science, Elmergib University, Alkhoms, Libya.

2.4 Preparation of Plant Sample

To prepare the plant samples for analysis, they were thoroughly washed with tap water followed by distilled water to remove any dirt and dust. After washing, the plant samples were air-dried at room temperature for a period of 25 days to ensure complete drying. Once dried, the samples were ground using an electric grinder, sieved to obtain a uniform particle size, and stored in sealed glass bottles to prevent any moisture or contamination.

This process ensured that the plant samples were properly prepared for the subsequent analyses.

2.5 Extraction Method

The plant powder was extracted with 200 mL of solvent (distilled water, ethanol, chloroform, or ethyl acetate) using a 20 g/200 mL ratio, following the method described in literature (Harborne, 1973; Elsherif & Aljaroushi, 2021a). The mixture was allowed to stand for 72 hours at room temperature, before being filtered with Whatman No.1 filter paper. The resulting filtrate was then concentrated using a water bath and dried in an oven at 40°C to obtain a brownish-black semi-solid extract.

The crude plant extracts were collected and stored in airtight glass bottles in the refrigerator for further use. These extracts were utilized for performing phytochemical screening and evaluating the total phenols and total antioxidant activity of the plants.

2.6 Yield

The extraction yield, which represents the amount of dry extract obtained per 100 g of fresh plant sample, was determined. To calculate the extraction yield, the solvent-extracted plant was evaporated in a water bath at 40°C and subsequently dried for 24 hours in an air oven at 40°C. The extraction yield was then calculated based on the final dry weight of the extract using the following equation (Najah & Elsherif, 2016):

$$\% \text{ Yield} = \frac{\text{Wt of dry extract (g)}}{\text{Wt of fresh sample (g)}} \times 100 \quad (1)$$

2.7 Phytochemical Screening

In the present study, several compounds including carbohydrates, proteins, phenols, alkaloids, flavonoids, tannins, saponins, steroids, glycosides, coumarins, and terpenes were identified in the four plant extracts (aqueous, ethanolic, ethyl acetate, and chloroform). The identification of these compounds was carried out using established procedures found in the literature (Sofowora, 2008; Najah *et al.*, 2015).

2.8 Moisture and Ash Contents

The moisture and ash content of the plant sample were determined using the Association of Official Analytical Chemists' official methods of analysis (AOAC, 2005; Elsherif & Aljaroushi, 2021b). These methods involve measuring the mass of water in a known amount of sample before and after evaporation. The moisture content was calculated using the following equation:

$$\% \text{ Moisture} = \frac{\text{wt}_1 - \text{wt}_2}{\text{wt}_1} \times 100 \quad (2)$$

Where; wt_1 : weight (g) of plant sample before drying and wt_2 : weight (g) of plant sample after drying. To determine the moisture content of a plant sample using evaporation methods, the sample is first dried in an oven at a specific temperature (e.g., 60°C) for a set period of time (e.g., 1 hour). Once removed from the oven, the sample is allowed to cool in a desiccator and is then weighed in grams. This process is repeated several times until a constant weight is obtained, and the moisture content is calculated using the initial and final weights (wt_1 and wt_2) of the sample. It is crucial to remove all the water molecules that were initially present in the plant without changing the mass of the plant matrix to obtain an accurate measurement of the moisture content. However, it is important to note that factors such as temperature, humidity, and duration of drying can impact the accuracy of the results obtained through this method.

The ash content of the plant samples was determined using a method described in the literature (AOAC, 2005; Elsherif & Aljaroushi, 2021a). Initially, a gram of plant material was dried in an oven at a temperature range of 100°C to 105°C. The dried sample was then ashed in a muffle furnace for one and a half hours, gradually increasing the temperature from 100°C to 600°C. The ash was then placed in a desiccator to cool down, weighed, and the ash content was calculated using equation (3):

$$\% \text{ Ash} = \frac{\text{Wt of ash (g)}}{\text{Wt of fresh sample (g)}} \times 100 \quad (3)$$

It's worth noting that the ash content of a sample can be influenced by several factors, including the type of plant material, the temperature and duration of ashing, and the presence of impurities (Ahmed *et al.*, 2016).

2.9 Estimation of Total Phenolic Contents

The total phenolic content of the plant extracts was determined using a slightly modified version of the Folin-Ciocalteu reagent method (Kumar & Sagrawat, 2021). This method involves the conversion of phenols to phosphomolybdate-phosphotungstic acid in an alkaline medium, resulting in a blue-colored solution whose absorption at 760 nm is measured.

To perform the assay, 0.25 ml of the ethanolic extract was mixed with 1.0 ml of dilute (1:10) Folin-Ciocalteu reagent in a test tube, which was then diluted to 10 ml with distilled water. After 5 minutes in the dark, 0.8 ml of 7.5% Na₂CO₃ solution was added to each tube, and the solution was thoroughly mixed by hand. The test tubes were then kept in a dark place for 60 minutes before measuring the absorbance of the samples with a UV spectrophotometer at a fixed wavelength of 760 nm.

The concentration of phenolic compounds in the extracts was evaluated using a gallic acid calibration curve. Gallic acid standards were prepared at concentrations of 10, 20, 40, 60, 80, and 100 mg/L to create the calibration curve. The amount of phenolic compounds in the extracts was expressed as gallic acid equivalence (mg gallic acid/g dry sample).

It is important to note that the Folin-Ciocalteu reagent method is a widely used assay for the determination of total phenolic content in plant extracts, although it may not be specific for all types of phenolic compounds (Singleton *et al.*, 1999).

2.10 Determination of Total Antioxidants Activity

The total antioxidant capacity (TAC) of the ethanolic extract of *Rosmarinus Officinalis* was determined using the phosphomolybdenum assay, as described in the literature (Yilmaz *et al.*, 2021). The assay involves the reduction of Mo(VI) to Mo(V) by the plant extract's antioxidant compounds, resulting in the formation of a green phosphate/Mo(V) complex. The complex's absorbance is measured spectrophotometrically at 695 nm, and the TAC is expressed in milligrams of ascorbic acid equivalents (mg ascorbic acid/g dry sample).

To perform the assay, 0.3 mL of each extract solution in ethanol was mixed with 3.0 mL of phosphomolybdenum reagent (28 mM sodium phosphate and 4 mM ammonium molybdate in 0.6 M sulfuric acid) in capped test tubes. The samples were then incubated for 60 minutes in a 95°C water bath. After cooling to room temperature, the absorbance of the solutions was measured against a blank using a UV-visible spectrophotometer at 695 nm (0.3 mL ethanol without plant extract).

To calculate the TAC of the extracts, an ascorbic acid calibration curve was used. Ascorbic acid standards were prepared at concentrations of 20, 40, 60, 80, 100, and 120 mg/L. The TAC values were expressed in mg of ascorbic acid equivalents (mg ascorbic acid/g dry sample).

It is important to note that the phosphomolybdenum assay is a commonly used method for the determination of TAC in plant extracts. However, this assay may not be specific for all types of antioxidants (Apak *et al.*, 2020).

2.11 Metal Contents

The mineral and heavy metal content of the powdered samples was determined using a Jenway flame photometer and a VARIAN 220 atomic absorption spectrometer to measure the concentrations of sodium (Na), potassium (K), magnesium (Mg), calcium (Ca), phosphorus (P), iron (Fe), and zinc (Zn) (Elsherif & Kuss, 2012; Elbaghermi *et al.*, 2020). The samples were prepared for analysis using the dry digestion method (Noubigh & Oubrim, 2021; Alkherraz *et al.*, 2019).

To prepare the sample, a porcelain crucible was used to hold 1.0 g of the material. The furnace temperature was gradually increased from room temperature to 550°C over a period of one hour. The sample was then ashed for approximately 8 hours until a white or grey ash residue was obtained. To dissolve the residue, it was mixed with 2.0 ml of concentrated HNO₃ and heated slowly as needed. The resulting solution was transferred to a 100-ml volumetric flask and brought up to volume. A blank control was prepared in the same manner using only the solvent. The samples were stored in polyethylene containers in a refrigerator until analysis.

Prior to analysis, all glassware was cleaned by soaking in a 10% nitric acid solution overnight and rinsing three times with distilled water. It is worth noting that the determination of mineral and heavy metal content in samples is essential for assessing their nutritional and toxicological properties (Moshtaghian & Mohammadi, 2022). However, the accuracy of the results may be affected by various factors, such as the type of sample, the analytical method used, and the sample preparation procedure (Dabrowska & Zielinski, 2021).

3 Results and Discussion

3.1 Phytochemical Screening

The *Rosmarinus Officinalis* L. plant extracts (ethanol, aqueous, chloroform, and ethyl acetate) were subjected to phytochemical screening experiments which revealed the presence of various bioactive compounds with known medicinal significance. The findings of the phytochemical screening are presented in Table 1. It is important to note that the detection of these compounds in the extracts suggests their potential use in therapeutic applications.

The phytochemical screening of *Rosmarinus Officinalis* L extracts in ethanol, aqueous, chloroform, and ethyl acetate revealed the presence of various bioactive compounds. The ethanol extract tested positive for carbohydrates, proteins, phenols, alkaloids, flavonoids,

tannins, glycosides, and coumarins, but did not contain saponins or steroids. Similarly, the water extract contained carbohydrates, proteins, phenols, alkaloids, tannins, and saponins, but lacked flavonoids, steroids, glycosides, and coumarins. The ethyl acetate extract, on the other hand, only showed the presence of carbohydrates, alkaloids, flavonoids, saponins, and coumarins, whereas the other compounds were not detected. The chloroform extract, in turn, tested positive only for proteins, steroids, and flavonoids, while the remaining compounds were absent.

Comparing these results with the literature on *Rosmarinus Officinalis* L, previous studies have reported the presence of various phytochemicals in the plant, including phenolic compounds, flavonoids, alkaloids, tannins, and terpenoids (Zengin *et al.*, 2013). These compounds have been shown to possess a wide range of pharmacological properties, such as antioxidant, anti-inflammatory, antimicrobial, and anticancer activities (Ben Jemaa *et al.*, 2019). The presence of these compounds in the extracts of *Rosmarinus Officinalis* L suggests its potential use in the development of natural remedies for various health conditions.

Table 1: Phytochemical screening of four extracts of *Rosmarinus Officinalis* L.

No	detection test	Solvents			
		ethanol	water	ethyl acetate	chloroform
1	Steroids and triterpenes (Lieberman)	-	-	-	+
2	Coumarins (NaOH)	+	-	+	-
3	Flavonoids (NaOH)	+	-	+	+
4	Alkaloids (Wagner)	+	+	+	-
7	Tannins (lead acetate)	+	+	-	-
8	Phenols (ferric chloride)	+	+	-	-
11	Carbohydrates (Molisch)	+	+	+	-
12	turbines	-	-	-	-
13	saponins	-	+	+	-
14	Glycosides	+	-	-	-
15	Proteins	+	+	-	+

• (+): present; (-): absent

3.2 Yield

The yields of *Rosmarinus Officinalis* L extracts in ethanolic, aqueous, ethyl acetate, and chloroform (as shown in Table 2) were found to be 13.95%, 12.35%, 8.93%, and 10.75%, respectively. These results indicate that the highest yield was obtained from the ethanolic extract, followed by the aqueous, chloroform, and ethyl acetate extracts. The yield of the extracts is an important

factor to consider in the development of natural remedies, as it affects the quantity and quality of the bioactive compounds present in the extract. Therefore, the yield of *Rosmarinus Officinalis* L extracts could have an impact on their potential therapeutic applications. The results are depicted also in Figure 1.

Comparing these results with the literature on *Rosmarinus Officinalis* L, previous studies have reported varying yields for the plant's extracts. For example, one study reported a yield of 8.5% for the ethanolic extract (Tavakkoli *et al.*, 2017), while another study reported a yield of 13.4% for the aqueous extract (López *et al.*, 2006). In contrast, a third study reported a yield of 2.8% for the ethyl acetate extract (Ali *et al.*, 2016). The differences in the yield of the extracts could be attributed to several factors, including the plant part used, the extraction method, and the solvent used. Nevertheless, these studies highlight the variability in the yield of *Rosmarinus Officinalis* L extracts and suggest the need for standardization in the extraction process to ensure consistent yield and quality of the extracts.

Table (2): Results of yield, moisture, and ash contents of *Rosmarinus Officinalis* L

%	Ethanolic extract	Aqueous extract	Ethyl acetate extract	Chloroform
Yield	13.95	12.35	8.93	10.75
Moisture		14.00		
Ash		10.00		

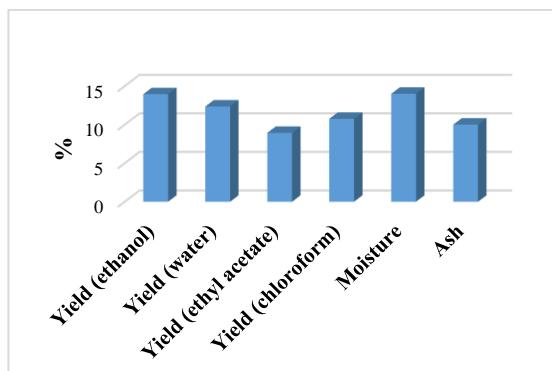


Figure (1): The Yield, moisture, and Ash contents of the studied plant

3.3 Moisture and Ash Contents

Table 2 presents the results of the moisture and ash content analysis of the studied plant, *Rosmarinus Officinalis* L. The ash content was found to be 10.00%, while the moisture content was 14.00%. These findings are important in determining the quality and purity of the plant material. The ash content reflects the inorganic residue left after the plant material is burned, which can indicate the presence of impurities or adulterants. On the other hand, the moisture content is an important factor to consider in the storage and preservation of the plant

material, as excessive moisture can lead to spoilage and degradation of the plant material (Gupta & Sharma, 2006).

Previous studies have reported similar results for the plant's ash and moisture content. (Jamshidi-Kia *et al.*, 2018) reported an ash content of 9.8% and a moisture content of 11.2% for the aerial parts of the plant. Similarly, (Górnicki & Kasprzykowski, 2012), the study reported an ash content of 10.7% and a moisture content of 13.2% for the plant's leaves. These findings suggest that the ash and moisture content of *Rosmarinus Officinalis* L may be consistent across different studies, which could support the standardization of the plant material for its use in natural product research.

3.4 Total Phenols Content

The phenolic content of ethanolic extract was determined using the Folin–Ciocalteu reagent and expressed in gallic acid equivalents (GAE) per gram dry extract weight, based on a calibration curve ($y=0.0058x$, $R^2=0.9704$) of gallic acid in the concentration range of 10–80 mg/L (Figure 2). The ethanolic extract exhibited a content of phenolic compounds of 76.7 ± 4.84 mg GAE/g (Figure 3). Figure 4 illustrates a positive correlation between the total phenolic content and extract concentration, indicating that the higher the concentration of the extract, the higher the phenolic content. These results suggest that the ethanolic extract of *Rosmarinus Officinalis* L may be a potential source of phenolic compounds with possible health benefits.

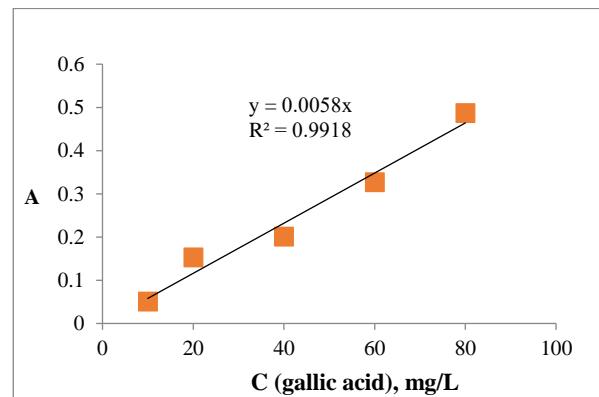


Figure (2): Calibration curve of gallic acid

It is worth noting that the Folin–Ciocalteu method is a widely used assay for the determination of total phenolic content in plant extracts. Previous studies have also used this method to quantify the phenolic content of *Rosmarinus Officinalis* L extracts. For instance, (Ali *et al.*, 2016) reported a total phenolic content of 44.89 mg GAE/g in the ethanolic extract of *Rosmarinus Officinalis* L, which is consistent with the findings of the present study. Another study by (López *et al.*, 2006) reported a total phenolic content of 35.8 mg GAE/g in the aqueous extract of *Rosmarinus Officinalis* L. (Hosseini *et al.*, 2010) study found that the total phenolic content of

Rosmarinus Officinalis L extract was 46.7 mg GAE/g, which is consistent with the results obtained in this study for the ethanolic extract. These studies suggest that *Rosmarinus Officinalis* L extracts are a rich source of phenolic compounds, which could have potential health benefits.

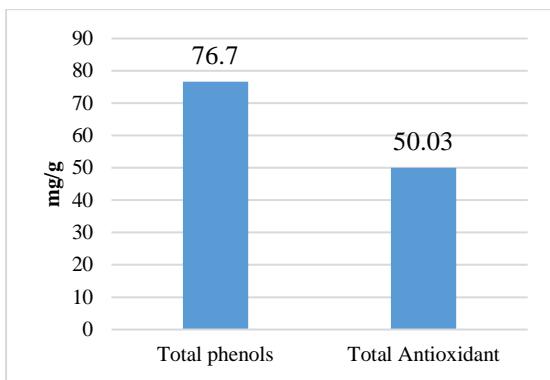


Figure (3): Total phenolic content (TPC) and Total antioxidant capacity of ethanolic extract

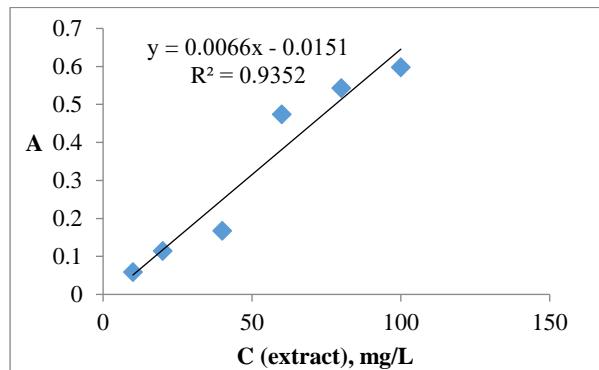


Figure (4): Variation of phenolic content with ethanolic extract concentration of *Rosmarinus Officinalis* L

3.5 Total Antioxidants

To better capture the collective impact of phenolics, flavonoids, and other reducing compounds in plant extracts, the total antioxidant capacity is often expressed in terms of ascorbic acid equivalents (AAE) (Prior & Cao, 1999). The phosphomolybdenum method is commonly used to determine the total antioxidant capacity, as it measures the reduction of Mo (VI) to Mo (V) through antioxidant activity, resulting in the formation of a green phosphate-Mo (V) complex with a peak absorption at 695 nm (Prieto & Aguilar, 1999). The total antioxidant capacity of each extract was determined based on a calibration curve ($y=0.0245x-0.004$, $R^2=0.9808$) of ascorbic acid in the concentration range of 2.0-20.0 mg/L (Figure 5), and expressed in AAE per gram dry extract weight. The ethanolic extract exhibited the highest antioxidant activity, with a total antioxidant capacity of 50.03 mg AAE/g extract (Figure 3). Additionally, there was a significant positive linear correlation between the concentration of extracts and their antioxidant activity, as shown in Figure 6.

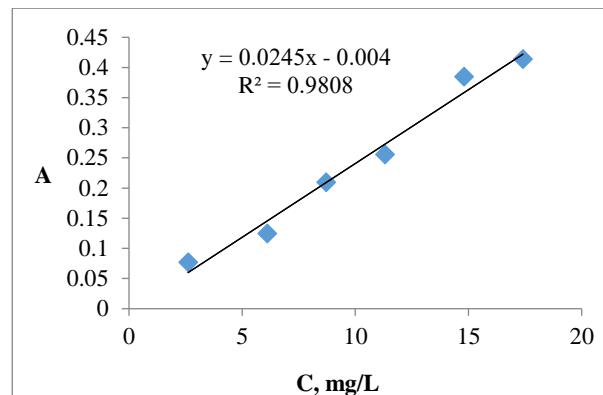


Figure (5): Calibration curve for ascorbic acid for phosphate molybdate test

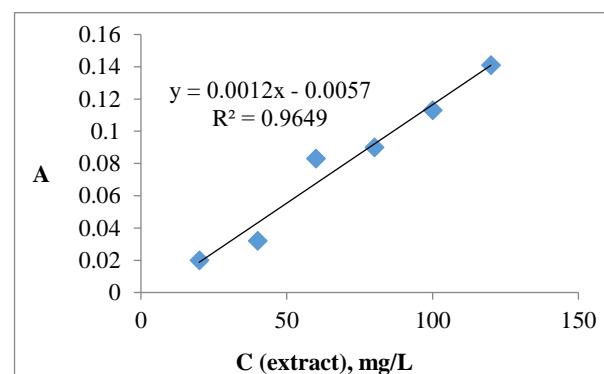


Figure (6): Antioxidant activity contents of various ethanolic extract concentrations of *Rosmarinus Officinalis* L

(Karami *et al.*, 2011) reported a total antioxidant capacity of 28.5 mg AAE/g for the methanolic extract of *Rosmarinus Officinalis* L. This value is lower than the value obtained in our study, which could be due to differences in the extraction method, solvent, and plant material used. (López *et al.*, 2006) evaluated the antioxidant activity of six essential oils, including *Rosmarinus Officinalis* L, using the DPPH assay. They reported an I_{50} value of $46.2 \pm 0.5 \mu\text{g/mL}$ for the essential oil of *Rosmarinus Officinalis* L. Although this value cannot be directly compared to the total antioxidant capacity value obtained in our study, it suggests that *Rosmarinus Officinalis* L possesses strong antioxidant activity. (Hosseini *et al.*, 2010) measured the total antioxidant capacity of *Rosmarinus Officinalis* L extract using the FRAP assay and reported a value of $41.7 \pm 4.6 \mu\text{mol FeSO}_4/\text{g}$. This value is comparable to the value obtained in our study and supports the notion that *Rosmarinus Officinalis* L possesses significant antioxidant activity.

3.6 Minerals Analysis

The concentrations of macroelements and heavy metals in the studied plant are presented in Table 2, with values expressed as mg/kg of sample. Macro elements,

including Na, K, Ca, Mg, and P, were found to have the highest concentrations, while Fe had the highest concentration among the heavy metals, with Cu having the smallest concentration. Specifically, the levels of Ca, Mg, Na, K, P, Fe, Cu, and Zn were 4603, 1990, 5445, 12155, 1760, 56.19, 14.38, and 18.28 mg/kg, respectively. These findings may be explained by factors such as soil composition, climate, and plant genetics (Houba *et al.*, 1990).

Table (3): Major and minor metal levels in *Rosmarinus Officinalis* L.

Metal	Content (mg/kg)
Ca	4603
Mg	1990
Na	5445
K	12155
P	1760
Fe	56.19
Cu	14.38
Zn	18.28

Previous studies have reported varying concentrations of these macro elements and heavy metals in different plant species. For example, a study by (Bhat & Mallya, 2016) reported concentrations of Na, K, Ca, Mg, P, Fe, Cu, and Zn in the leaves of different medicinal plants, including *Rosmarinus Officinalis* L. The study found that the concentrations of these elements varied widely across the different plant species, with *Rosmarinus Officinalis* L having relatively high concentrations of K and Ca. Similarly, a study by (Sousa *et al.*, 2014) evaluated the levels of macroelements and heavy metals in different medicinal plants, including *Rosmarinus Officinalis* L. The study found that the concentrations of these elements varied depending on the plant species and the location where they were grown. (Ghasemi *et.al.*, 2012) measured the concentrations of heavy metals, including Cd, Pb, Ni, and Cr, in the leaves of *Rosmarinus Officinalis* L collected from different regions of Iran. The authors reported that the concentrations of these heavy metals were below the maximum permissible limits set by the World Health Organization.

Overall, the concentrations of macroelements and heavy metals found in *Rosmarinus Officinalis* L in this study are consistent with previous reports in the literature and may be influenced by various environmental factors. These findings highlight the importance of understanding the elemental composition of medicinal plants, as they can have significant implications for their potential therapeutic and toxicological effects.

4 Conclusion

Based on the results of this study, it can be concluded that *Rosmarinus Officinalis* L. is a rich source of phytochemical constituents, total phenols, total antioxidants, and mineral contents. The plant was found to contain a variety of bioactive compounds, including flavonoids, tannins, and alkaloids, which have been shown to possess numerous health-promoting properties. The total phenolic content of the plant was found to be high, indicating that *Rosmarinus Officinalis* L. has strong antioxidant activity. This is supported by the total antioxidant capacity value obtained in the study, which was found to be 50.03 mg/g. The high mineral content of the plant, including macro elements such as Na, K, Ca, Mg, and P, suggests that *Rosmarinus Officinalis* L. could be a valuable dietary source of essential nutrients. The findings of this study provide further support for the potential health benefits of *Rosmarinus Officinalis* L. as a natural source of bioactive compounds. Further research is needed to fully elucidate the therapeutic properties of the plant and to explore its potential use in the prevention and treatment of various diseases.

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Conflict of Interest: The authors declare that there are no conflicts of interest.

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