Egypt. J. Chem. **Vol. 66,** No. 8 pp. 191 - 208 (2023)

Egyptian Journal of Chemistry

http://ejchem.journals.ekb.eg/

Effect of some mineral nutrients on vegetative growth, chemical constituents, and anatomical structure of Grevillea robusta seedlings.

El-Khateeb, M. A.1; Sakr, S. S.1; Hashish K.I.2, Azza A. M. Mazhar2, and Abdel-Magied, H. M.2

[1]*Department of Ornamental Horticulture, Faculty of Agriculture, Cairo University, Egypt*

[2]Department of Ornamental Plants and Woody Trees, Agricultural & Biological Research Institute, National Research Centre (NRC), 33 El-Buhouth St., 12622 Dokki, Giza, Egypt.

Abstract

This study was carried out in the Experimental Nursery of Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, and the chemical analyses were carried out in laboratories of the National Research Centre, during two seasons the 2014-2015 and 2015-2016 to investigate the response of Grevillea robusta A. Cunn. seedlings to some treatments of mineral nutrients: zinc (Zn) at 50 and 100 ppm, cobalt (Co), nickel (Ni), and selenium (Se) at 5 and 10 ppm. , Uniform seedlings with an average 25cm height and 0.35 cm stem diameter were planted in 35 cm plastic pots, filled with a sandy loam soil were used in this study. The treatments were applied individually as a foliar spray every month, in both seasons throughout the period of growth. Plants were sprayed with Zn at 100 and 50 ppm produced the tallest plants, the thickest stem and main root as well as the heaviest fresh and dry weights of all plant parts. The plants treated with Ni at 10 ppm gave the maximum numbers of leaves and the longest roots. Spraying plants with Ni at 10 and Se at 5 ppm increased the content of chlorophyll-a, b and total carotenoids, while treating seedlings with Zn at 50 and 100 ppm increased the total carbohydrates, N%, P%, K% and Zn (ppm). The plants produced the maximum concentration of Co (ppm) when they were sprayed with Co 10 and 5 ppm, however the concentration of Ni (ppm) were increased markedly in response to 10 and 5 ppm of Ni treatments. Study the anatomical structures of the transverse and the longitudinal sections of main stem of G. robusta were showed an increments by treated with Zn at 100 ppm as compared with untreated plants.
Key words: *Grevillea robusta, zinc, cobalt, nickel, and selenium.*

1. Introduction

Grevillea robusta is an erect, single-stemmed tree typically reaching 20-30m tall and 80cm in stem diameter. It has a conical crown, dense with dark grey to dark brown bark [1]. Its attractive shape and beautiful foliage make *G. robusta* an ideal ornamental tree for landscaping and roadside plantings as a shade or windbreaks tree, the cut leaves are used in flower arrangements, and young plants are grown as indoor pot plants. The tree has economic potential, which produces an attractively figured, easily worked wood and its branches and twigs are used for firewood and charcoal. The leaves contain several useful chemical compounds, in particular rutin, dyes, phenolic compounds[2],[3] and gum resin produced from the wood, which have pharmacological applications.

Several decades ago, there was an urgent requirement to evaluate the different elements or nutrients and their importance for plants like N, P, K, Mg, Ca,…etc., whereas some still need evidence like cobalt (Co), nickel (Ni) and selenium (Se). The importance of the microelements has been well recognized such zinc is required at low levels (e.g., 20-100 mg g-1 dry weight in leaves) for plant enzymatic activity, maintenance of biomembrane integrity, carbohydrate metabolism, and protein synthesis and its participates in the metabolism of indole acetic acid[4],[5]and[6] stated that zinc is essential for the vegetative growth and production of plants, where it is required in all photosynthetic tissues and needed for cell division and development. [7] reported that zinc is a co-factor for several

*Corresponding author e-mail: khaledhashish@yahoo.com.; (Khaled Ismail Hashish).
Received date 14 October 2022; revised date 14 October 2022; accepted date 01 December 2022
DOI: 10.21608/ejchem.2022.168821.7088

enzymes such as anhydrases, dehydrogenases, oxidases and peroxidases. Furthermore, Zn could be also involved in chlorophyll formation by taking part in the regulation of cytoplasmic concentrations of nutrients. Co, Ni and Se are not required by all plants but can promote plant growth and may be essential for particular taxa. These beneficial elements have been reported to improve plant productivity and enhance plant nutritional value by boosting resistance to biotic and abiotic stresses. [8], [9]. [10] indicated that Co is an essential component of several enzymes and co-enzymes that can affect the growth and metabolism of plants, through the retardation of leaf senescence via inhibition of ethylene biosynthesis, and enhancement of drought resistance. Co also can stimulate alkaloids accumulation in medicinal plants, through regulation of the biosynthesis of aromatic amino acid precursors of alkaloids. In some low-Co conditions, a small increase in Co stimulates growth for higher plants. However, high Co concentrations can become toxic to plants. Also, the low concentration of Ni is essential for root growth and hydrogenase activation. The efficiency of nitrogen fixation immediately depends on hydrogenase activity because the oxidation of hydrogen by the latter provides ATP required for N reduction to ammonia[11]. On Grevillea exul var. rubiginosa, [12] found that Ni treatment with 100mg/L suppressed germination and root growth. Furthermore, Ni has been identified as a component of a number of enzymes, including glyoxalases, peptide deformylases, and ureases, and a few superoxide dismutases and hydrogenases [13].Selenium is a cofactor of the enzyme glutathione peroxidase, which is a catalyzer in the reduction of peroxides that can damage cells and tissues, and can act as an antioxidant which can improve antioxidative status and to a reduced biosynthesis of ethylene, which is the hormone with a primary role in plant senescence [14],[15]. The first positive effect of Se on plant growth was reported by [16] who showed that the application of 0.5 mg kg-1 Se stimulated growth and dry-matter yield of Indian mustard (*Brassica juncea L.*).[17] showed that the positive effects of low Se concentrations on the photosynthetic process may be explained via the enhancement of the antioxidant activity in cells.

2. Materials and methods

The present study was carried out in the Experimental Nursery of Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, and the chemical analyses were carried out in laboratories of the National Research Centre, during two seasons the 2014-2015 and 2015-2016. The objective of this study was to investigate the effect of some mineral nutrients (zinc, cobalt, nickel, and selenium) on vegetative growth, chemical constituents, and anatomical structure of *Grevillea robusta* A.Cunn. seedlings.

2.1. Plant materials

Seedlings of *Grevillea robusta* A. Cunn. were obtained from El Bably farm-Qanater khairia, Cairo, Egypt, with an average 23-25 cm height, 10 No. of leaves, and 0.35 cm stem diameter were used in this study.

2.2. Experimental procedures

On the 10th of February, during both seasons the seedlings were transplanted in plastic pots of 35cm diameter, filled with sandy loam soil, the physical and chemical analysis of these growing media is presented in Table (1). The commercial fertilizer "Kristalon®" (N: P: K 19:19:19) at 2.5 g/pot was added twice every season; the first was applied after a month from transplanting, and second was added after 5 months. In the second week of January, every season was the end of the growing period. All seedlings were irrigated regularly.

Table 1. Physical and chemical analysis of the growing media.

Physical analysis								
Clay%		Sand %			Silt %			
19.6		66.8			13.6			
Chemical analysis								
E.C. dS/m	pH	Anion (meq/L.)			Cation (meq/L.)			
		HCO3-	Cl-	SO4--	Ca++	Mg++	Na+	K+
0.30	8.01	1.1	1.2	0.70	1.0	0.6	1.2	0.2
DTPA-extractable-elements (ppm)								
Zn		Co			Ni			
0.07		Not detected			Not detected			

The layout of the experiment was a completely randomized design and each treatment had 3 replicates (3 seedlings per replicate)Mineral nutrients of zinc, as well beneficial elements of cobalt, nickel, and selenium were applied as a foliar spray every month, in both seasons throughout the period of growth individually as zinc at the rate of 50 and 100 ppm, while cobalt, nickel, and selenium were applied at the rates of 5 and 10 ppm, using Bio-film as a wetting agent at 1 ml/l of the solution, until the solution runoff point. Seedlings were irrigated regularly. Zinc as Zn-chelated (EDTA 13%), cobalt as cobalt sulfate, extra pure AR (Co So4.7H2O 99%), Ni as nickel sulfate hexahydrate

(Ni So4.6H2O), and Se as selenium dioxide (Se O_2 99.8%).

The following data were recorded from *Grevillea robusta* seedlings at the end of each season:

2.3. Vegetative growth characteristics

Plant height (cm), number of leaves/plant, stem diameter (cm) (about 3cm from the seedling base), root length (cm), fresh weights (F. W.) of leaves, stems, and roots (g) and dry weights (D. W.) of leaves, stems, and roots (g).

2.4. Chemical composition

Photosynthetic pigments content (chlorophyll a, b, and carotenoids, mg/g. F.W.), determined according to [18], total carbohydrates in leaves, stems, and roots (%D.W.), determined according to[19], nitrogen and phosphorus in leaves, stems, and roots (%D.W.), determined according to[20], potassium, zinc, cobalt, and nickel in leaves, stems and roots (%D. W.), determined according to [21].

2.5. Anatomical structure

Anatomical studies (transverse sections and longitudinal sections) were conducted on the main stem of *Grevillea robusta* A. Cunn. treated with 100 ppm of zinc as compared to the control.Samples were taken from the main stem of *Grevillea robusta* A. Cunn. plants were cleaned with tap water and cut into suitable parts, killed and fixed in FAA, dehydrated in different concentrations of ethyl alcohol, clearing in different concentrations of ethyl alcohol+xylene, infiltrated and embedded in pure paraffin wax (M.P.58-60cº) [22]. Sectioning at a thickness of 20µm was performed by using a rotary microtome. Paraffin ribbons were mounted on slides and sections stained with safranine and fast green. Sections were mounted in Canda balsam and then examined microscopically and in microphotography.

2.6. Statistical analysis

Analysis of the variance of the obtained data from each attribute was computed using the MSTAT Computer Program [23]. Duncan's New Multiple Range test at a 5% level of probability was used to test the significance of differences among mean values of treatments [24].

3. RESULTS AND DISCUSSION

3.1. Vegetative growth

The results in Table (2) showed that applications of Zn, Co, Ni, and Se at different concentrations positively affected the height of *Grevillea robusta* A. Cunn. seedlings in two seasons. Sprayed plants with Zn at 100 and 50 ppm were the tallest plants (80.73 and 78.56 cm), followed by Co at 10 ppm (75.10 cm) which significantly differed from other treatments. Whereas, the shortest plant was recorded by untreated plants, followed by Se at 10 ppm giving 58.63 and 63.22 cm in ascending order.

The obtained results of the second season revealed that, the average values of plant height ranged from 51.73 to 79.02 cm. Foliar spraying with 50 ppm of Zn gave the tallest plant (79.02 cm) followed by Zn at 100 ppm (76.00 cm) compared with the other treatment. However, the shortest plant was recorded by untreated plants (51.73 cm), followed by plants treated with Se at 5 ppm (54.72 cm). A similar trend was obtained by [25], [26], [27], and [28]. Likewise, enhancing effect of lower doses of Co on plant height had previously been reported by [29]. Whereas, cobalt is necessary for the processes of stem growth and elongating the coleoptiles. Moreover, cobalt reduces the peroxidase activity, which is known to affect the breakdown of Indole acetic acid (IAA). Cobalt, a transition element, is an essential component of several enzymes and coenzymes. It has a role in affecting the growth and metabolism of plants in different stages, depending on the concentration and status of cobalt in the rhizosphere and soil. The beneficial effects of cobalt include retardation of senescence of leaf, increase in drought resistance in seeds, regulation of alkaloid accumulation in medicinal plants, and inhibition of ethylene biosynthesis[30].

The results in Table (2) indicated that treating Grevillea robusta seedlings with Ni and Co at 10 ppm and Zn at two levels (50 and 100 ppm, each in order), as well as Co and Ni at 5ppm, significantly increased the number of leaves/plant giving 30.80, 27.73, 25.50, 23.06, 25.11 and 22.82 leaves/plant, respectively, as compared with that untreated plants, which obtained the least number of leaves/plant (18.13). Regarding the second season, the maximum numbers of leaves were produced from Zn at 100 ppm (37.67 leaves/plant), followed by Zn at 50 ppm and Ni at 10 ppm giving 31.56 and 30.13, respectively. Whereas the minimum numbers of leaves/plant was obtained from untreated plants (23.08 leaves/plant). Depending on its concentration, nickel can both stimulate and inhibit enzyme activities in plant tissues, where, the activity of the antioxidant enzymes such as catalase, peroxidase, superoxide dismutase, glutathione reductase, and ascorbate oxidase, enzyme activities considerably declined at higher Ni concentration and were promoted at a lower Ni concentration which protect plant cells against free radicals and lead to vigor seedlings growth [31].

Data presented in Table (2) showed that in the first season. The stem thicknesses of 0.83 and 0.80 cm were recorded on plants sprayed with Zn at 100 and 50 ppm, respectively. While thinness of 0.58 and 0.60 cm was found with the untreated plants and seat 10 ppm, respectively.

The results in the second season were similar to those obtained in the first one. The data showed that Zn at 100 produced the thickest stem (0.80 cm), followed by Zn at 50 ppm and Co at 10 ppm (0.78 and 0.73 cm), meanwhile, the thinnest stem was obtained by the control plants which gave (0.52 cm). Generally, the addition of Zn at different concentrations had a great effect on increasing the thickness of plant stems, as compared with the other treatments, where an ideal fertilization regime promotes seedling growth and nutrient loading without toxic effects [32].

Table 2. Effect of some mineral nutrients(Zn, Co, Ni and Se)on some vegetative growth parameters of *Grevillea robusta* A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

Treatments	Plant height (cm)	No of leaves /plant	Stem diameter (cm)	Root diameter (cm)	Root length (cm)
			First season		
Control	58.63f	18.13e	0.58e	0.41g	14.05e
Zn 50ppm	78.56ab	25.50bc	0.80ab	0.62a	26.83a
Zn 100ppm	80.73a	25.11bc	0.83a	0.52cde	23.51b
Co 5ppm	71.32c	23.06cd	0.75bc	0.48ef	21.22bc
Co 10ppm	75.10b	27.73b	0.77bc	0.50def	19.41cd
Ni 5ppm	66.41de	22.82cd	0.68d	0.55bcd	20.60c
Ni 10ppm	69.09cd	30.80a	0.73cd	0.60ab	28.02a
Se 5ppm	65.00e	20.70de	0.60e	0.56bc	17.27d
Se 10ppm	63.22e	21.03de	0.61e	0.45fg	17.13d
			Second season		
Control	51.73f	23.08f	0.52f	0.35f	11.51f
Zn 50ppm	79.02a	31.56b	0.78ab	0.57a	17.01c
Zn 100ppm	76.00a	37.67a	0.80a	0.56a	17.31bc
Co 5ppm	60.50c	28.36cd	0.70c	0.51bc	14.20de
Co 10ppm	67.81b	26.73de	0.73bc	0.40ef	15.71cd
Ni 5ppm	60.00cd	26.00e	0.64d	0.47cd	19.00ab
Ni 10ppm	63.17c	30.12bc	0.71c	0.54ab	20.21a

Se 5ppm	54.72ef	25.06ef	0.56ef	0.43de	16.00cd
Se 10ppm	56.33de	24.81ef	0.58e	0.52abc	13.46e

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

The results presented in Table (3) revealed that treating the plants with all treatments significantly increased the root length in both seasons as compared with the control. In the first season, the plants treated with Ni at 10 ppm produced the longest roots (28.02 cm) as compared with the other treatments. In the second season, the plants sprayed with Ni at 10 and 5 ppm as well as Zn at 100 and 50 ppm produced maximum values of the root length which gave 20.21, 19.00, 17.31, and 17.01 cm, respectively, as compared with the other treatments. While, the unsprayed plants produced the shortest roots with 14.05 and 11.51 cm, respectively in the two seasons.

Data on fresh and dry weights of leaves as affected by different concentrations of Zn, Co, Ni, and Se are presented in Table (3). It can be discussed as follows, all treatments significantly increased leaves fresh weight.

In the first season, Zn at 50 ppm gave the heaviest fresh leaves with 44.71 g, as compared with the untreated plants which gave 20.62 g, while, in the second season, Zn at 100 significantly increased the weight of fresh leaves giving 41.63, as compared with the control plants (26.67 g). The lightest fresh leave was obtained by Se at 10 which gave 25.00 g.

Concerning the effect of different concentrations of Zn, Co, Ni, and Se on dry weights of Grevillea robusta leaves (Table, 3). The effects had the same trend, since the application of Zn at 50 ppm significantly increased leaves' dry weight which gave 18.78, consecutively, in the first season. On the other hand, the lightest leaves dry weight was recorded by untreated plants (8.25 g). In the second season, it is obvious that the weight of the grevillea dry leaves had almost the same trend as the fresh weights as shown in Table (3).

Data presented in Table (3) show that all used mineral nutrients produced significant increments in fresh and dry weight of stem in comparison to control in two seasons. Application of Zn at 100 ppm significantly increased the fresh weights of grevillea stem in the first season, giving the heaviest stem fresh weights (29.38g), as compared with the other treatments, where the lightest fresh stems were recorded by using Se at 10 ppm (20.00 g). In the second season, plants treated with Zn at a low rate (50 ppm) produced heavy fresh stems (26.71g), compared with other treatments. Meanwhile, the untreated plants gave the lightest value of the fresh weight of stem (18.15 g).

Regarding the effect of the two concentrations of Zn, Co, Ni, and Se on stem dry weights of *Grevillea robusta* A. Cunn. seedlings, the data in Table (21) indicated that there was a similar trend with the values of stem fresh weights in the first and second seasons.

The effect of two levels of Zn, Co, Ni, and Se on the fresh weight of roots was shown in Table (3). It is clear from the obtained data that the root fresh weights of *Grevillea robusta* A. Cunn. seedlings were markedly increased when seedlings were treated with Zn at 100 and 50 ppm and Ni at 10 ppm in both seasons

As for the weight of dry roots, data obtained in Table (3) show clearly that all treatments showed a remarkable increment with a significant effect on dry weights of roots in the first season. This effect was more pronounced when Zn at 100 and 50 ppm was sprayed, which increased the values to reach their maximum (13.37 and 11.90 g), in contrast to those previous treatments the untreated plants recorded the lightest dry roots with 5.62 g. In the second season, the same trend of the fresh weight was shown. So, the heaviest dry roots were recorded when the plants were sprayed with Zn at 100 and 50 ppm (11.82 and 9.87 g) followed by Ni at 10 ppm (8.75 g), meanwhile, the lightest dry roots were reduced by the untreated plants.

Table 3. Effect of some mineral nutrients (Zn, Co, Ni and Se) on fresh and dry weights of leaves, stems and roots of *Grevillea robusta* A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

Treatments	Leaves fresh weight (g)	Leaves dry weight (g)	Stem fresh weight (g)	Stem dry weight (g)	Root Fresh weight (g)	Root dry weight (g)

			First season			
Control	20.62h	8.25f	23.00d	10.22d	10.52g	5.62g
Zn 50ppm	44.71a	18.78a	27.41b	13.81ab	23.60b	11.90b
Zn 100ppm	30.53c	11.91c	29.38a	14.60a	26.70a	13.37a
Co 5ppm	24.11f	9.66def	20.82ef	10.35d	20.00de	9.95de
Co 10ppm	27.60d	10.85cd	23.20d	11.60cd	20.83cd	10.40cd
Ni 5ppm	26.00e	10.41cde	22.16de	11.13cd	18.56e	9.38e
Ni 10ppm	38.30b	16.85b	25.60c	12.80bc	22.01bc	11.08bc
Se 5ppm	22.47g	8.90ef	29.11a	14.06ab	15.33f	7.91f
Se 10ppm	22.06g	8.83ef	20.00f	11.55cd	14.76f	7.36f
			Second season			
Control	26.67ef	10.75de	18.15f	9.15e	12.07f	6.40d
Zn 50ppm	36.41b	14.57b	26.71a	13.39a	19.73b	9.87b
Zn 100ppm	41.63a	16.64a	26.00ab	13.21ab	23.66a	11.82a
Co 5ppm	27.23e	11.21de	21.03e	10.62d	13.11ef	6.70d
Co 10ppm	25.26fg	10.84de	22.06de	11.00cd	14.36de	7.55cd
Ni 5ppm	28.10e	11.75de	22.00de	11.49cd	16.06cd	8.35c
Ni 10ppm	30.70d	12.02cd	23.31cd	12.12abc	16.32c	8.75bc
Se 5ppm	34.12c	13.30bc	24.63bc	11.96bcd	13.92e	6.79d
Se 10ppm	25.00g	10.28e	21.50e	10.88cd	12.80ef	6.48d

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

3.2. Chemical constituents

3.2.1. Photosynthetic pigments content (mg/g F. W)

Data in Table (4) declared that Ni at 10 ppm produced the greatest content of chlorophyll-a (0.93 and 0.90 mg/g F.W), followed by Zn at 100 ppm (0.90 and 0.81 mg/g F.W.), in the first and second seasons, respectively. On the other hand, the lowest chlorophyll-a content was recorded by untreated plants (0.54 and 0.50 mg/g F.W.) in both seasons.

Concerning the effect of Zn, Co, Ni, and Se at two rates on chlorophyll- b content, the obtained results clear that spraying grevillea plants with Se at 5 ppm and Ni at 10 and Zn at 100 ppm were more effective treatments on increasing the chlorophyll-b content in the leaves giving 0.47, 0.46 and 0.43 mg/g F.W., than other treatments in the first season. While, the untreated plants produced the minimum value of chlorophyll-b content (0.27 mg/g F.W.) in the first season. In the second season, similar results were obtained in a different order.

Table 4. Effect of some mineral nutrients (Zn, Co, Ni and Se) on the content of chlorophylls and total carotenoids (mg/g F.W.) Grevillea robusta A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

Treatments	Chl. a	Chl. b	Carotenoids
		First season	
Control	0.54e	0.27e	0.39e
Zn 50ppm	0.76c	0.38bc	0.67c
Zn 100ppm	0.87b	0.43ab	0.71bc
Co 5ppm	0.56e	0.29de	0.44e
Co 10ppm	0.65d	0.33cd	0.54d
Ni 5ppm	0.70d	0.34cd	0.59d
Ni 10ppm	0.93a	0.46a	0.79a
Se 5ppm	0.80c	0.47a	0.74ab
Se 10ppm	0.59e	0.31de	0.41e
		Second season	
Control	0.50e	0.24e	0.36e
Zn 50ppm	0.73c	0.39b	0.59c
Zn 100ppm	0.81b	0.46a	0.70b
Co 10ppm	0.61d	0.31cd	0.50d
Co 5ppm	0.54e	0.28de	0.42e
Ni 5ppm	0.63d	0.34bc	0.50d
Ni 10ppm	0.90a	0.49a	0.78a
Se 5ppm	0.70c	0.37b	0.63c
Se 10ppm	0.53e	0.23e	0.39e

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

Whereas, the lowest content of chlorophyll-b in grevillea leaves was obtained with plants treated with Se at 10 ppm resulting in 0.23 mg/g F.W. followed by the untreated plant (0.24 mg/gm F.W.) in the second season.

It is well known that zinc participates in chlorophyll synthesis and protects chlorophyll from decomposition and it affects photosynthesis by influencing the carbon anhydride enzyme activity and the chlorophyll content. It also influences nitrogen assimilation [33], [34]. Likewise, cobalt performs biological functions in plants as the synthesis of chlorophyll- b.[35].

Regarding the effect of different concentrations of Zn, Co, Ni, and Se on total carotenoids content, the obtained data indicated that the maximum values of the carotenoids content were recorded by plants treated with Ni at 10 ppm and Se at 5 ppm (0.79 and 0.74 mg/g F.W.), followed by Zn at 100 ppm (0.71 mg/g F.W.) in the first season. Meanwhile, the lowest value of the total carotenoids was found in the untreated plants (0.39 mg/g F.W.) in the first season.

In the second season, spraying plants with all treatments increased the total carotenoid content in grevillea leaves spraying the plants with Ni at 10, Zn at 100 ppm and Seat 5 ppm produced 0.78, 0.70, 0.63mg/g F.W., respectively, as compared with the control plants which produced 0. 36 mg/g F.W of total carotenoids content.

3.2.2. Total carbohydrates percentage (% D.W.)

Data in Table (5) and showed the effect of various concentrations of Zn, Co, Ni, and Se on total carbohydrates in the different organs of *Grevillea robusta* A. Cunn. seedlings.

The results declared that treating seedlings with Zn at 100 and 50 ppm and Ni at 10 ppm was the most effective in increasing the total carbohydrates percentage in grevillea leaves giving 30.06, 28.22 and 27.35%, respectively, compared with other treatments, which the lowest leaves of total carbohydrates percentage were obtained by untreated plants (21.24%) followed by plants treated with Se at 10 ppm (23.71%) in the first season. Also, Zn at 50 ppm and Ni at 10 ppm applications caused a remarkable increment of total carbohydrates percentage in leaves giving 29.12% and 27.76%, respectively, in the second season. Whereas, the total carbohydrates percentage in grevillea leaves declined to reach 21.39% in control plants and 22.51% in plants inoculated with Se at 10 ppm, in the second season.

Regarding, total carbohydrates in stems data declared that, the stem carbohydrates percentage reached the maximum values when plants were sprayed with Zn at 100 and 50 ppm and Ni at 10 ppm in the first seasons, giving 44.84, 42.16, and 41.43%, respectively. However, grevillea plants contained low values of total carbohydrates in their stems, when treated with Se at 10 ppm (34.13%) in the first season. While, in the second season treatments of Ni at 10 ppm and Zn at 50 and 100 ppm produced stems containing high total carbohydrates percentage (45.76, 43.31 and 42.85%, respectively), on the other hand, the untreated plants recorded the lowest content of total carbohydrates (35.00%).

Concerning the effect of different concentrations of Zn, Co, Ni, and Se on total carbohydrates in *Grevillea robusta* roots, the data indicated that the application of Zn at 50 ppm resulted in the highest content of total carbohydrates (40.00%) followed by Zn at 100 ppm and Ni at 5 ppm treatment giving 38.50 and 36.12% in the first season as compared to the control plants (29.08%).

Table 5. Effect of some mineral nutrients (Zn, Co, Ni and Se) on total carbohydrates percentage (% D. W.) in the different organs of *Grevillea robusta* A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

Treatments	Leaves	Stems	Roots
		First season	
Control	21.24f	34.83de	29.08e
Zn 50ppm	28.22b	42.16b	40.00a
Zn 100ppm	30.06a	44.87a	38.50a
Co 5ppm	24.51cde	36.46d	32.26c
Co 10ppm	25.09cd	35.54de	31.76cd
Ni 5ppm	25.75c	39.86c	36.12b
Ni 10ppm	27.35b	41.43bc	35.30b
Se 5ppm	23.71de	34.13e	30.10de
Se 10ppm	23.22e	34.73de	31.05cde
		Second season	
Control	21.39d	35.00e	29.24e
Zn 50ppm	29.12a	43.31b	39.16ab
Zn 100ppm	26.42b	42.85b	40.03a
Co 5ppm	23.33c	36.26e	31.11de
Co 10ppm	23.87c	39.81cd	32.73d
Ni 5ppm	26.12b	39.76cd	35.47c
Ni 10ppm	27.76ab	45.76a	41.24a
Se 5ppm	24.10c	40.33c	37.53bc
Se 10ppm	22.51cd	38.46d	32.00d

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

However, Ni at 10 ppm and Zn at 100 and 50 ppm gave the maximum values of carbohydrates content resulted in 41.24, 40.03 and 39.16%, respectively, in the second season, compared with untreated plants which produced the minimum values of carbohydrates content (29.24%) in the second season.

3.3. Mineral concentrations

3.3.1. Nitrogen percentage (% D.W.)

Data presented in Table (6) showed clearly that the nitrogen concentration in *Grevillea robusta* leaves was increased when plants were treated with all tested mineral nutrients. In the first season, the maximum values of nitrogen concentration were in grevillea leaves treated with Co at 10 ppm giving 2.02%and Zn at 100 ppm resulting in 1.85%. Whereas, the lowest concentration of nitrogen was recorded by untreated plants (1.01%) followed by Se at 10 ppm (1.39%) compared with other fertilizers treatments.

Results of the second season revealed that the average N% ranged from 1.05% to 1.97%. The plants sprayed with Zn at 50 ppm gave the highest values (1.97%) followed by those treated with Zn at 100 ppm (1.90%). The low values of N% (1.05 and 1.40%) were recorded with unsprayed and sprayed plants with Co at 5 ppm, in ascending order. The data in Table (6) showed that the concentrations of nitrogen in *Grevillea robusta* stems had a great response to the application of some mineral element treatments. In the first season, N% of stems was insignificantly affected by all tested elements. grevillea stems obtained the highest nitrogen concentrations when treated with Co at 10 and Zn at 100 ppm giving 1.53 and 1.49%, respectively. While, untreated plants gave the lowest content of nitrogen (0.85%).

Regarding the effect of Zn, Co, Ni, and Se on nitrogen concentration in grevillea stems, it was found that in the second season, all treatments significantly increased N% compared with the untreated plants. In this respect, the determined values showed that plants treated with Zn at 50 and 100 ppm gave 1.47and 1.42%, respectively, While, untreated plants gave the lowest content of nitrogen (0.79%).

Table 6. Effect of some mineral nutrients (Zn, Co, Ni and Se) on nitrogen percentage (% D. W.) in the different organs of Grevillearobusta A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

	Nitrogen (% D. W.)		
Treatments	Leaves	Stems	Roots
	First season		
Control	1.01e	0.85a	0.63de
Zn 100ppm	1.85a	1.49a	1.11a
Zn 50ppm	1.64b	1.41a	0.91b
Co 10ppm	2.02a	1.53a	1.16a
Co 5ppm	1.43cd	1.16a	0.60e
Ni 10ppm	1.61bc	1.37a	0.86b
Ni 5ppm	1.51bcd	1.34a	0.70c
Se 10ppm	1.39d	1.09a	0.66cd
Se 5ppm	1.56bcd	1.40a	0.71c
	Second season		
Control	1.05b	0.79d	0.60f
Zn 100ppm	1.90a	1.42a	0.97ab
Zn 50ppm	1.97a	1.47a	1.01a
Co 10ppm	1.71ab	1.30abc	0.80cd
Co 5ppm	1.40ab	1.17c	0.64ef
Ni 10ppm	1.75a	1.31abc	0.84c
Ni 5ppm	1.60ab	1.29abc	0.76d
Se 10ppm	1.46ab	1.21bc	0.69e
Se 5ppm	1.63ab	1.38ab	0.92b

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

Data in Table (6) showed that using various concentrations of Zn, Co, Ni and Se affected nitrogen content in the roots of grevillea seedlings. Zinc at 100 and 50 ppm produced the greatest content of N resulted in respectively, in the first season and respectively in the second season. On the other hand, Se at 10 ppm gave the lowest value of N content (0.60%) in the first season. Also, the untreated plants produced the minimum content of N in the second season, the concentration of nitrogen in roots of *Grevillea robusta* A. Cunn. seedlings were affected in both seasons by different levels of Zn, Co, Ni, and Se as shown in Table (6). Spraying plants with different concentrations of Zn, Co, Ni, and Se significantly increased N% in two seasons. The plants produced the maximum concentration of N% in root in the first season when they were sprayed with Co at 10(1.16%) and Zn at 100 ppm (1.11%), followed by Zn at 50 ppm and Ni at 10 ppm (0.91

Data presented in Table (7) showed the effect of different treatments on P% in *Grevillea robusta* stems. In this respect, treating the seedling with Zn at 100 and 50 ppm, gave the greatest concentration of phosphorus in the stems (0.21 and 0.20%) in the first season, as compared with the control plants (0.10%). Whereas, spraying plants with Zn at 50 and 100 ppm resulted in the maximum phosphorus percentage in stems (0.22 and 0.20%) in the second season, respectively, compared with untreated plants (0.12%).

In the case of the roots, there was a remarkable increase in the concentrations of phosphorus as a

and 0.86%). In the second season, the maximum concentration of nitrogen was recorded with Zn at 50 and 100 (1.01 and 0.97%), followed by Se at 5 and Ni at 10 ppm (0.92 and 0.85%). On the other hand, the untreated plants gave the minimum value of N% (0.60%).

3.3.2. Phosphorus percentage (% D.W.)

As for the effect of Zn, Co, Ni, and Se treatments on phosphorus concentration in the different organs, of Grevillea robusta A. Cunn. seedlings, the results in Table (7) revealed that, treating the plants with those treatments increased the phosphorus percentage in the leaves over the control plants. The maximum values (0.29 and 0.25%) in the first season and (0.32 and 0.30%) in the second one were obtained with the application of Zn at 100 and 50 ppm, respectively, as compared with the control plants, which produced (0.17 and 0.18%) in both seasons.

result of treating Grevillea robustaA. Cunn. seedlings with Zn at 100 and 50 ppm and Co at 10 ppm giving 0.26, 0.23, and 0.20% in the first season, respectively. However, in the second season, the same results were obtained but in different orders. The maximum phosphorus concentration in roots was obtained by Zn at 50 and 100 ppm and Co at 10 ppm resulting in (0.27, 0.24, and 0.21%), respectively. On the other hand, the lowest values of phosphorus concentration in roots resulted from untreated plants in the two seasons (0.15 and 0.14%).

Table 7. Effect of some mineral nutrients (Zn, Co, Ni and Se) on the contents of phosphorus (% D. W.) in the different organs of *Grevillea robusta*A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

	Phosphorus (% D. W.)					
	Leaves	Stems	Roots	Leaves	Stems	Roots
Treatments	First season			Second season		
Control	0.17c	0.10d	0.15c	0.18c	0.12c	0.14d
Zn 50ppm	0.25ab	0.20ab	0.23ab	0.30a	0.22a	0.27a
Zn 100ppm	0.29a	0.21a	0.26a	0.32a	0.20ab	0.24ab
Co 5ppm	0.19c	0.15a-d	0.19bc	0.21c	0.18abc	0.18cd
Co 10ppm	0.23bc	0.17abc	0.20bc	0.27ab	0.19ab	0.21bc
Ni 5ppm	0.19c	0.16a-d	0.18bc	0.20c	0.17abc	0.17cd
Ni 10ppm	0.20bc	0.17abc	0.19bc	0.24bc	0.17abc	0.20bcd
Se 5ppm	0.16de	0.11cd	0.08ab	0.17de	0.09c	0.06cd
Se 10ppm	0.18c	0.12cd	0.17bc	0.21c	0.16abc	0.17cd

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

3.3.3. Potassium percentage (% D.W.)

The obtained data in Table (8) revealed the concentration of potassium in all plant organs of *Grevillea robusta*A. Cunn. seedlings were increased by treating with Zn, Co, Ni, and at two rates as compared with the control plants.

The maximum values of potassium content in leaves in both seasons were obtained by Zn at 100 and 50 ppm and Co at 10 ppm, giving (0.74, 0.71, and 0.63%) in the first season, respectively, and (0.68, 0.63, and 0.61%) in the second one, respectively. The untreated plants gave the minimum concentrations of potassium in both seasons (0.41 and 0.40%), respectively.

Table 8. Effect of some mineral nutrients (Zn, Co, Ni and Se) on potassium percentage (% D. W.) in the different organs of Grevillea robusta A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

Treatments	Potassium (% D. W.)					
	Leaves	Stems	Roots	Leaves	Stems	Roots
	First season			Second season		
Control	0.41f	0.21c	0.16d	0.40f	0.19g	0.15d
Zn 50ppm	0.71a	0.37a	0.27ab	0.63ab	0.40a	0.30a
Zn 100ppm	0.74a	0.42a	0.29a	0.68a	0.36ab	0.25ab
Co 5ppm	0.60bc	0.27bc	0.23bc	0.53cde	0.30cd	0.26ab
Co 10ppm	0.63b	0.31b	0.20cd	0.61abc	0.33bc	0.24ab
Ni 5ppm	0.55cd	0.25bc	0.20cd	0.57bcd	0.26def	0.22bc
Ni 10ppm	0.50de	0.30b	0.21cd	0.60abc	0.29cde	0.27ab
Se 5ppm	0.49e	0.23c	0.18cd	0.50de	0.21fg	0.17cd
Se 10ppm	0.51de	0.26bc	0.19cd	0.48e	0.24efg	0.18cd

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

All treatments with Zn, Co, Ni, and Se at two levels resulted in higher K concentrations in stems of *Grevillea robusta* A. Cunn. seedlings as compared with control plants in two seasons.

Regarding, the results in the first season, the application of Zn at 100 and 50 ppm and Co at 10 ppm gave the greatest values of K% in stems (0.42, 0.37, and 0.31%, respectively), whereas the lowest value of K% (0.21%) was produced with the untreated plants, followed by plants sprayed with Se at 5 ppm (0.23%).

Results in the second season were similar to those obtained in the first season but in different orders. The data showed that treating plants with Zn at 50 and 100 ppm and Co at 10 ppm gave the maximum values of K% (0.40, 0.36, and 0.33%), respectively, but the lowest value of K% (0.19%) was obtained from untreated plants, followed by Se at 5ppm with (0.21%).

In the first season, the data presented in Table (8) showed that K concentrations of *Grevillea robusta* roots ranged from 0.16 to 0.29%. The greatest percentage of K was recorded by plants treated with Zn at 100 ppm (0.29%), whereas untreated plants resulted in the lowest K percentage (0.16%). In the second season, the maximum K% (0.30 and 0.27%) were obtained from plants sprayed with Zn at 50 ppm and Ni at 10 ppm. On the other hand, the untreated and treated plants with Se at 5ppm resulted in the lowest K% values (0.15 and 0.17%), respectively.

3.3.4. Zinc concentration (ppm)

The response of *Grevillea robusta* A. Cunn. to the different treatments of Zn, Co, Ni, and Se is presented in Table (9). Data revealed that spraying plants with all treatments increased the percentage of zinc in leaves as compared with untreated plants in the two seasons. Concerning, the first season, spraying the seedlings with Zn at 100 and 50 ppm and Co at 10 ppm produced the greatest increase in the Zn concentration (57.13, 54.00, and 41.06 ppm, respectively) over the control (25.60 ppm). Whereas, in the second season, using Zn at 50 and 100 ppm, followed by Ni at 10 ppm produced the greatest Zn concentration in *Grevillea robusta* leaves, resulting in 53.36, 50.78, and 40.22 ppm, respectively, as compared with the control plants (27.41 ppm). Concerning the effect of Zn, Co, Ni, and Se at two levels on Zn concentration in *Grevillea robusta* stems, data in Table (27) revealed that plants fertilized with Zn at 100 and 50 ppm and Co at 5 and 10 ppm resulted in 43.58, 40.63, 32.30 and 30.46 ppm, respectively in the first season. Meanwhile, the control plants obtained 18.76 ppm of Zn concentration in grevillea stems. In the second season, the maximum values of Zn concentration in *Grevillea robusta* stems were obtained by Zn at 50 and 100 ppm and Co at 10 and 5ppm giving 41.39, 40.61, 34.00, and 31.73 ppm as compared with the untreated plants (19.25 ppm).

Results in Table (9) showed that Zn concentration in *Grevillea robusta* roots was affected by different two levels of Zn, Co, Ni, and Se. In the first season, the seedlings fertilized with Zn at 50 and 100 ppm produced the greatest percentage of Zn concentration (30.72 and 30.15 ppm), followed by Co at 10 and 5 ppm resulted in 23.66 and 21.70 ppm, as compared to the control (10.57 ppm). In the second season, a similar trend was observed and the maximum Zn concentration in *Grevillea robusta* roots was obtained by Zn at 100 and 50 ppm (32.30 and 30.50 ppm), followed by Co at 5 and 10 ppm (24.00 and 22.00 ppm), as compared with the control plants (13.21 ppm). Similar data were also reported by [36], who found that total Zn contents in the shoots increased with the increased application of external zinc.

Table 9. Effect of some mineral nutrients (Zn, Co, Ni and Se) on zinc concentrations (ppm) in the different organs of *Grevillea robusta* A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

Treatments	Zinc (ppm)					
	Leaves	Stems	Roots	Leaves	Stems	Roots
	First season			Second season		
Control	25.60h	18.76f	10.57f	27.41f	19.25f	13.21h
Zn 50ppm	54.00b	40.63b	30.72a	53.36a	41.39a	30.50b
Zn 100ppm	57.13a	43.58a	30.15a	50.78b	40.61a	32.30a
Co 5ppm	38.73d	32.30c	21.70c	37.80d	31.73c	24.00c
Co 10ppm	41.06c	30.46c	23.66b	39.40cd	34.00b	22.00d
Ni 5ppm	31.82f	26.60d	17.26d	33.65e	28.87d	19.07e
Ni 10ppm	35.30e	28.13d	18.00d	40.22c	27.21d	19.23e
Se 5ppm	27.94g	20.16ef	15.43e	29.58f	25.15e	17.29f
Se 10ppm	27.10gh	21.40e	14.80e	28.75f	23.53e	15.63g

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

3.3.5. Cobalt concentration (ppm)

Data in Table (10) showed that the application of Zn, Co, Ni, and Se at two rates had appositive effects with clear significant differences in cobalt concentration in the different organs of *Grevillea robusta* A. Cunn. seedlings.

Concerning the results of *Grevillea robusta* leaves, the determined values, showed that application of Co at 10 and 5 ppm and Zn at 50 ppm gave the highest concentration of

Co resulting in 4.45, 4.21, and 3.06 ppm, respectively, in the first season. Also, in the second season, Co at 10 and 5 ppm produced the greatest Co concentration (4.17 and 4.02 ppm), followed by Zn at 100 ppm (3.11 ppm). On the other hand, the lowest Co concentration was obtained by the untreated plants (0.79 and 0.73), followed by Se at 10 ppm (1.30 and 1.17 ppm) in the first and second seasons, respectively.

Table 10. Effect of some mineral nutrients (Zn, Co, Ni and Se) on cobalt concentrations (ppm) in the different organs of Grevillea robusta A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

	Cobalt (ppm)					
	Leaves	Stems	Roots	Leaves	Stems	Roots
Treatments	First season			Second season		
Control	0.79i	0.31h	0.20g	0.73i	0.33h	0.21g
Zn 50ppm	3.06c	2.03c	0.75c	2.79d	1.89c	0.62c
Zn 100ppm	2.87d	1.71d	0.62d	3.11c	1.00e	0.57cd
Co 5ppm	4.21b	3.16a	1.00b	4.02b	2.96a	1.45a
Co 10ppm	4.45a	2.75b	1.10a	4.17a	2.84b	1.33b
Ni 5ppm	2.19f	1.16f	0.57d	2.33e	1.22d	0.47ef
Ni 10ppm	2.63e	1.40e	0.60d	2.21f	1.19d	0.50de
Se 5ppm	1.73g	0.77g	0.37f	1.80g	0.92f	0.40f
Se 10ppm	1.30h	0.83g	0.44e	1.17h	0.80g	0.39f

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

Regarding the concentration of cobalt in *Grevillea robusta* A. Cunn. stems, the application of mineral nutrients (Zn, Co, Ni, and Se) increased the Co concentration, giving its highest values with Co at 5 and 10 ppm and Zn 50 ppm recorded at 3.16, 2.75, and 2.03 ppm, respectively, in the first season and 2.96, 2.84 and 1.89 ppm. in the second one, in descending order, as compared with (0.31 and 0.33 ppm) for the untreated plants in both seasons, respectively.

The concentration of cobalt in roots of Grevillea robusta A. Cunn. seedlings were affected in both seasons by different levels of Zn, Co, Ni, and Se as shown in Table (10). The results were similar to those observed in either leaves or stems. Spraying plants with different concentrations of Zn, Co, Ni, and Se significantly increased Concentration in two seasons. The plants produced the maximum concentration of Co in roots in the first season when they were sprayed with Co 10 and 5 ppm (1.10 and 1.00 ppm). On the other hand, the untreated plants gave the minimum value of Co concentration (0.20 ppm). In the second season, the results were almost similar to those of the first one, the plants sprayed with Co at 5 and 10 ppm gave the greatest Co concentration in grevillea roots (1.45 and 1.33 ppm), while the unsprayed plants recorded the minimum concentration of Co in the root (0.21 ppm). In this connection, Co concentration in plants is typically in the range of 0.1-10 ppm on a dry weight basis [30].

3.3.6. Nickel concentration (ppm)

The response of nickel concentration in all plant organs of *Grevillea robusta* A. Cunn. seedlings to the different treatments of Zn, Co, Ni, and Se were presented in table (11). Data revealed that treating the plants with Zn, Co, Ni, and Se at two levels increased Ni concentration in grevillea leaves as compared with the untreated one in the first season. Spraying the seedlings with Ni at 10 and 5 ppm and Zn at 100 ppm, produced the greatest increase in nickel concentration (5.98, 4.55, and 3.63 ppm), as compared with unsprayed seedlings which recorded the lowest concentration of Ni in leaves (1.21 ppm), followed by plants treated with Se at 10 and 5 ppm giving 1.52and 1.95 ppm in ascending order. In the second season, spraying plants with all treatments increased nickel concentration in grevillea leaves.

The values of 6.64, 6.27, and 4.16 ppm were obtained with the application of Ni at 10 and 5 ppm as well as Zn at 100 ppm, respectively, as compared with the control plants, which produced 1.43 ppm. In this respect, [37] pointed out that, Ni concentration in most plant leaf material normally ranges from about 0.1 to 5 ppm (in dry weight), but can be highly variable depending on its availability in soils, plant species, plant part, and the season. Tissue Ni concentrations greater than 10 ppm are considered toxic in sensitive plant species.

Table 11. Effect of some mineral nutrients (Zn, Co, Ni and Se) on nickel concentrations (ppm) in the different organs of Grevillearobusta A. Cunn. seedlings during 2014-2015 and 2015-2016 seasons.

Treatments	Nickel (ppm)					
	Leaves	Stems	Roots	Leaves	Stems	Roots
	First season			Second season		
Control	1.21i	0.48g	0.36f	1.43g	0.55e	0.39f
Zn 50ppm	3.34d	2.49c	1.21c	3.90d	2.67b	1.44c
Zn 100ppm	3.65c	2.60c	1.39c	4.16c	2.55b	1.60bc
Co 5ppm	3.12e	1.35de	0.84de	3.20e	1.46c	0.82de
Co 10ppm	2.82f	1.51d	0.94d	3.03e	1.30cd	0.88d
Ni 5ppm	4.55b	3.22b	1.73b	6.27b	3.85a	1.72b
Ni 10ppm	5.98a	3.90a	1.98a	6.64a	3.68a	1.90a
Se 5ppm	1.95g	1.03f	0.68e	2.64f	1.17cd	0.65e
Se 10ppm	1.52h	1.23ef	1.62b	2.51f	1.09d	0.73de

Means within a column having the same letters are not significantly different according to Duncan's Multiple Range Test (DMRT) at 5% level.

Results in Table (11) indicated that nickel concentrations in *Grevillea robusta* stems were significantly affected by different levels of Zn, Co, Ni, and Se. In the first season, the results revealed that the average nickel concentration in grevillea stems ranged between 0.48 and 3.90 ppm. All treatments had a pronounced effect on increasing nickel concentration compared with the untreated plants which produced the lowest value (0.48 ppm), Meanwhile, the maximum concentrations of nickel (3.90, 3.22, and 2.60 ppm) were recorded in plants sprayed with Ni at 10 and 5 ppm as well as Zn at 100, respectively, as compared with the control plants (0.48 ppm). Whereas, in the second season Ni at 5 and 10 ppm and Zn at 50 ppm produced the highest values of Ni content (3.83, 3.68, and 2.67 ppm, respectively), as compared with the untreated plants, which gave 0.55 ppm.

The effect of mineral nutrients (Zn, Co, Ni, and Se at two levels) on nickel concentration in roots of *Grevillea robusta* A. Cunn. seedlings are shown in Table (11). It is obvious from data in the first season that the Ni concentration (ppm) in roots increased markedly in response to 10 and 5 ppm of Ni treatments and Se at 10 ppm (1.98, 1.73, and 1.62 ppm) in the first season, respectively as compared with the control plants and other treatments. Likewise, in the second season, the results of nickel concentration in roots of *Grevillea robusta* A. Cunn.

were almost similar to those in the first one, where nickel at 10 and 5 ppm gave the maximum concentrations of Ni (1.90 and 1.72 ppm), followed by Zn at 100 and 50 ppm resulted in 160 and 1.44 ppm, as compared with the control plants (0.39 ppm). The mechanisms behind the beneficial effects of Zn, Ni, Co, and Se on the plant are relatively under investigated, compared to the toxic effects that many beneficial elements have at high levels[8].

3.4. Anatomical studies

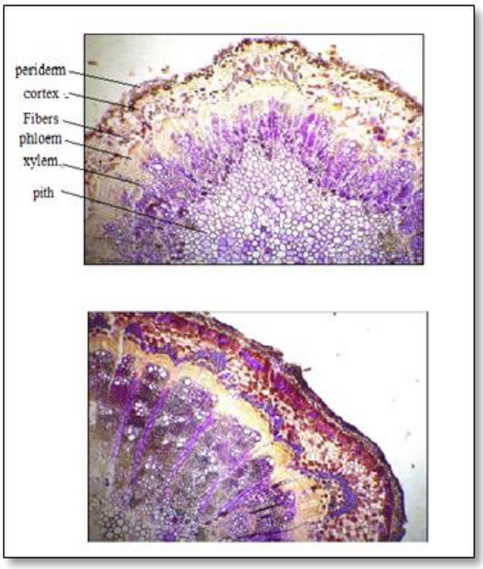
3.4.1. Effect of some mineral nutrients on stem anatomical structure of Grevillea robusta A. Cunn. Seedlings

Data presented in Table (12) stated that transverse sections of the main stem of *Grevillea robusta* A. Cunn. exhibited a clear increase with the treatment of zinc at 100 ppm for all histological characters understudies as compared with untreated plants. It is obvious from Table (12) and microphotographs in Fig. (1) that the thickness of periderm in *Grevillea robusta* stem treated with 100 ppm of zinc showed a response to control, which recorded an increase of 33.3% over the untreated plant. Meanwhile, cortex thickness showed an increase of 100% in plants sprayed with Zn at 100 ppm more than those in the control. The data presented in Table (12) and Fig (1) cleared that the thickness of the fibers verified in the grevillea stem was recorded an increase by 11.1% in treated plants over the control ones, while the thickness of phloem was cleared appositive effect in the treated plant by 20% over control.

Table 12. Measurements in micrometer (μ) of certain histological features in transverse section and longitudinal section through the main stem of *Grevillea robustaA*. Cunn. plant treated 100 ppm of zincand control plants.

	Transverse section characters						
Treatments	Periderm thickness	Fibers thickness	Cortex thickness	Phloem thickness	Xylem thickness	Pith thickness	Stem thicknes
Control	37.5	112.5	150	112.5	350	1250	5250
Zn 100ppm	50.0	125	75	125.0	625	750	6000

	Longitudinal section characters	
	Cortex parenchymatous cell length	Pith parenchymatous cell length
Control	37.5	37.5
Zn 100ppm	50	50



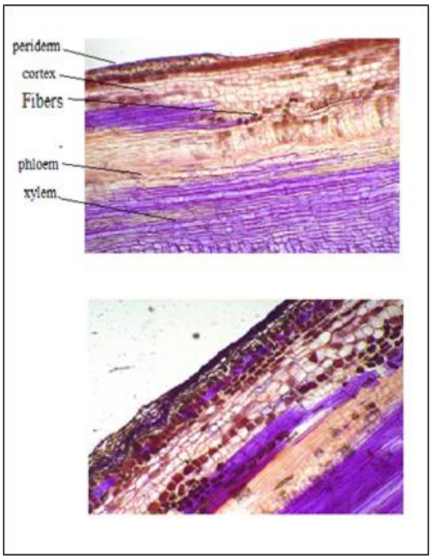


Fig. 1. Transverse section through the main stem of *Grev illea robustaplant* (x100).

a. Untreated plant (control)

Fig. 2. Longitudinalsectionthrough the main stem of Grevillea robusta plant (x100).

b. plant treated with 100 ppm of Zn

However, the xylem thickness of the main stem of *Grevillea robusta* was verified a sharp increase for treated grevillea plants by 78.5% over control. This result might lead to increasing the support of the xylem to the plant growth of the treated plants through the absorption of water and nutrients. Meanwhile, pith thickness exhibited an increase for the untreated plant which showed an increase of 16.6% over *Grevillea robusta* plants treated with 100 ppm of Zn.

The main stem thickness was increased by 14.3% over the thickness of the stem of control plants due to increments in most of the included tissues as well as the effects of Zn treatment on cell division and enlargement that led to an increase in the stem thickness.

The obtained and illustrated data in Table (12) and Fig. (2) concerning longitudinal sections of the main stem of *Grevillea robusta* plant revealed that the length of the parenchymatous cell of both cortex and pith were increased when grevillea plants treated with Zn at 100 ppm, where cortex parenchymatous cell length had an increment by 33.3% over untreated plants. While the length of the grevillea pith parenchymatous cell in longitudinal sections recorded 33.3% over untreated plants. Zinc plays a fundamental role in several critical cellular functions[38] and it can interact with various biochemical and metabolic processes in plants leading to structural cell modifications [39],[40], thus it has been reported to affect cell division and elongation[41], which may explain the results observed.

4. Conclusion

In conclusion, sprayed *Grevillea robusta* plants with Zn at 50 and 100 ppm produced the tallest plants, the thickest stem and main root as well as the heaviest fresh and dry weights of all plant parts, also produced the maximum values of total carbohydrates (% D.W.), N%, P%, K% and Zn (ppm) in all parts. Whereas, the plants treated with Ni at 10 ppm gave the maximum numbers of leaves and the longest roots. Concerning the photosynthetic pigments content, spraying plants with Ni at 10 ppm and Se at 5 ppm increased the content of chlorophyll-a, b and total carotenoids. The transverse and longitudinal sections of main stem were increased by treated with Zn at 100 ppm as compared with untreated plants.

5. Conflicts of interest

The author declares there is no conflict of interest.

6. References

[1]. McGillivray, D.M.; Makinson, R.O. (1993). Grevillea, Proteaceae, a taxonomic revision. Melbourne University Press Molecules, 16:9331-9339.

[2]. Cannon, J.R.; Chow, P.W.; Fuller, M.W.; Hamilton, B.H.; Metcalf, B.W. and Power, A.J. (1973). Phenolic constituents of Grevillea robusta (Proteaceae). The structure of robustol, a novel macrocyclic phenol. Australian Journal of Chemistry, 26(10):2257-2275.

[3]. Ahmed, A.S.; Nakamura, N.; Meselhy, M.; Makhboul, M.; El-Emary, N. and Hattori, M. (2000). Phenolic constituents from Grevillea robusta. Phytochemistry, 53(1):149-154.

[4]. Kaur, H., Kaur, H., Kaur, H. and Srivastava, S., 2022. The beneficial roles of trace and ultratrace elements in plants. Plant Growth Regulation, pp.1-18. doi.org/10.1007/s10725-022-00837-6.

[5]. Cakmak, I. (2000). Possible roles of zinc in protecting plant cells from damage by reactive oxygen species. New Phytologist, 146(2):185-205.

[6]. Mattiello, E.M.; Ruiz, H.A.; Neves, J.C.; Ventrella, M.C.; Araujo, W.L. (2015). Zinc deficiency affects physiological and anatomical characteristics in maize leaves. J. Plant Physiol., 183, 138–143.

[7]. Aravind, P. and Prasad, M. (2005). Zinc protects chloroplasts and associated photochemical functions in cadmium exposed Ceratophyllum demersum L., a fresh water macrophyte. Plant Science, 166:1321–1327.

[8]. Pilon-Smits, E.A.; Quinn, C.F.; Tapken, W.; Malagoli, M. and Schiavon, M., (2009). Physiological functions of beneficial elements. Current Opinion in Plant Biology, 12(3):267-274.

[9]. Marschner, H. (2012). Marschner's Mineral Nutrition of Higher Plants, 3rd Ed.: Academic Press Inc.London, 684 p.

[10]. Chang, J., Clay, D.E., Arnall, B. and Reicks, G. (2017). Essential plant nutrients, fertilizer sources, and application rates calculations. In: Practical Mathematics for Precision Farming (Eds. Clay et al.), American Society of Agronomy, Crop Science Society of America, and Soil Science Society of America, Inc., pp. 167-180.

[11]. Weisany, W.; Raei, Y. and Allahverdipoor, K.H. (2013). Role of some of mineral nutrients in biological nitrogen fixation. Bulletin of Environment, Pharmacology and Life Sciences, 2(4):77-84.

[12]. Leon, V.; Rabier, J.; Notonier, R.; Barthelemy, R., Moreau, X.; Bouraima-Madjebi, S.; Viano, J. and Pineau, R. (2005). Effects of threenickel salts on germinating seeds of Grevillea exul var. rubiginosa, an endemic serpentine proteaceae. Annals of Botany, 95(4):609-618.

[13]. Awasthi, S., Chauhan, R. and Srivastava, S., (2022). The importance of beneficial and essential trace and ultratrace elements in plant nutrition, growth, and stress tolerance. In Plant Nutrition and Food Security in the Era of Climate Change (pp. 27-46). Academic Press.

[14]. Ikram, M., Javed, B. and Raja, N.I., 2021. Biomedical potential of plant-based selenium nanoparticles: a comprehensive review on therapeutic and mechanistic aspects. International Journal of Nanomedicine, 16, p.249.

[15]. Puccinelli, M.; Malorgio, F. and Pezzarossa, B. (2017). Selenium enrichment of horticultural crops. Molecules, 22(6):923-933.

[16]. Singh, M.; Singh, H. and Bhandari, D.K. (1980). Interaction of selenium and sulphur on the growth and chemical composition of raya. Soil Science, 129:238-244.

[17]. Schiavon, M.; Lima, L.W.; Jiang, Y. and Hawkesford, M.J. (2017). Effects of selenium on plant metabolism and implications for crops and consumers. In: Selenim in plants (Eds. Pilon-Smits, E.A.H.; Winkel, L.H.E. and Lin, Z.Q.), Springer International Publishing AG, Cham (ZG), Switzerland, pp. 257-275.

[18]. Nornai, R. (1982). Formula for determination of chlorophylls pigments extracted with N. N. dimethyl formamide. Plant Physiol., 69: 1371-1381.

[19]. Herbert, D. P.; Phlipps, P. and Styrangl, R. (1971). Determination of carbohydrates method on microbial. In: Methods in Microbiology, (Eds. Norris, J.R. and Ribbons, D.W.), Academic Press, London, 5:209-344.

[20]. Cottenie, A.M.; Verloo, M.; Kiekens, L.; Velghe, G. and Camerlynck, R. (1982). Chemical Analysis of Plant and Soil. Laboratory of Analytical and Agrochemistry. State Univ. Ghent, Belgium, pp.100-129.

[21]. Chapman, H.D. and Pratt, P.F. (1961). Methods of Analysis for Soils, Plant and Water. Div. of Agric. Sci.,Univ. of California, 309 p.

[22]. Johansen, D.A. (1940). Plant Microtechique. McGraw-Hill Book Company, Inc., London, 523p.

[23]. MSTAT Development Team (1989). MSTAT user's guide: A microcomputer program for the design, management and analysis of agronomic research experiments. Michigan State University East Lansing, U.S.A.

[24]. Steel, R.G. and Torrie, J.H. (1980). Principles and Procedures of Statistics A Biometrical Approach. McGraw Hill, New York, 633p.

[25].Gad, N.; Abd El-Moez; Bekbayeva, L.K.; Karabayeva, A.A. and Surif, M. (2013). Effect of cobalt supplement on the growth and productivity of soybean (Glycine max L. Merril). World Appl. Sci. J., 26(7): 926-933.

[26]. Sahito, H.A.; Solangi, A.W.; Lanjar, A.G.; Solangi, A.H. and Khuhro, S.A. (2014). Effect of micronutrient (zinc) on growth and yield of

mustard varieties. Asian Journal of Agriculture and Biology, 2(4):105-113.
[27]. Sarma, B.; Devi, P.; Gogoi, N. and Devi, Y.M. (2014). Effects of cobalt induced stress on triticum aestivum L. crop. Asian J. Agri. Biol, 2(2):137-147.
[28]. Minz, A.; Sinha, A.K.; Kumar, R.; Kumar, B.; Deep, K.P. and Kumar, S.B. (2018). A Review on importance of cobalt in crop growth and production. International Journal of Current Microbiology and Applied Sciences, 7(2):2978-2984
[29]. Kadhim, R.E. (2011). Effect of Pb, Ni and Co in growth parameters and metabolism of Phaseolus aureus Roxb. Euphrates J. of Agri. Sci., 3(3):10-14.
[30]. Palit, S.; Sharma, A. and Talukder, G. (1994). Effects of cobalt on plants. Botanical Review, 60(2):149-181.
[31]. Seregin, I.V. and Kozhevnikova, A.D. (2006). Physiological role of nickel and its toxic effects on higher plants. Russian Journal of Plant Physiology, 53(2):257-277.
[32]. Uscola, M.; Salifu, K.F.; Oliet, J.A. and Jacobs, D.F. (2015). An exponential fertilization dose-response model to promote restoration of the Mediterranean oak (Quercu ilex). New Forests 46(5):795-812.
[33].Han J,Lee Y,Yeom K.H,Kim Y.K, Jin H and Kim V.N.(2004).The Drosha-DGCR8 complex in primary microRNA processing Genes Dev. 2004; 18: 3016-3027

[34]. Alekhina N D, Kharitonashvili E V (2005) Mineral Nutrition In: Ermakov I P (ed)Fiziologiya rastenii (Plant Physiol),Moscow, pp: 306-415.
[35]. Dennis, M. and Kolattukudy, P.E. (1992). A cobalt-porphyrin enzyme converts a fatty aldehyde to a hydrocarbon and Co. Proceedings of the National Academy of Sciences, 89(12):5306-5310.
[36]. Zhao, F.J.; Shen, Z.G. and McGrath, S.P. (1998). Solubility of zinc and interactions between zinc and phosphorus in the hyperaccumulator Thlaspi caerulescens. Plant, Cell and Environment, 21(1):108-114.
[37]. Barker, A.V. and Pilbeam, D.J. (2015). Handbook of Plant Nutrition. CRC press, 76p.
[38]. Marschner, H. (1995). Mineral Nutrition of Higher Plants. Academic Press Inc. London, 889 p.
[39]. Heumann, H.G. (2002). Ultra structural localization of zinc in zinc-tolerant Armeria maritima Halleri. by autometallography. J. Plant Physiol., 159:191-203.
[40]. Di Baccio, D.; Tognetti, R.; Minnocci, A. and Sebastiani, L. (2009). Responses of the Populus × euramericana clone I-214 to excess zinc carbon assimilation, structural modifications, metal distribution and cellular localization. Environ. Exp. Bot., 67: 153-163
[41]. Seregin, I.V.; Kozhevnikova, A.D.; Gracheva, V.V.; Bystrova, E. and Ivanov, V. (2011). Tissue zinc distribution in maize seedling roots and its action on growth. Russian Journal of Plant Physiology, 58(1):109-117.