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Chemical composition as affected by altitudes in *Nepeta septemcrenata* (Ehrenb) growing in Saint Katherine area

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Abstract

This study aims to investigate the impact of altitudinal gradients *viz*., 1600 m, 1850 m, 2100 m, 2350 m and 2600 m on succulence degree, total carbohydrate, soluble and non-soluble carbohydrate, crude protein, total lipids, total ash, total phenols, sodium, potassium, calcium, magnesium and chloride in *Nepeta septemcrenata* as a near endemic growing naturally at Saint Katherin, South Sinai, Egypt. Degree of succulence increased in plants at medium elevations (1850 m, 2100 m, and 2350 m). Total carbohydrate and non-soluble carbohydrate recorded the highest content in those at 2350 m altitude. Soluble carbohydrate and total lipids recorded the highest accumulation values in the plant at the highest altitude (2600 m). The other contents fluctuated among plants at different altitudes. Calcium element was the major among minerals composition in building cell of *N. septemcrenata*.

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Introduction

It is known that Sinai Peninsula has an important strategic location (the linking ring between Africa and Asia continents). Saint Katherine is a protectorate area includes the mountainous region of Southern Sinai. The area has an arid climate with a mean annual rainfall about 60 mm/year plus snow melt on higher mountain peaks which may reach up to 300 mm/year (Grainger, 2003). The altitudinal gradient along a mountain is associated with alterations in a number of environmental factors, such as air temperature, water precipitation, wind exposure, light intensity and partial CO_2 pressure (Kofidis *et al.*, 2003). Average air temperature extremes (1982-1991) ranged from 5.4 °C in January and February to 25.1 °C in July and August. At Gebel Katherine, mean monthly temperature ranges from -1°C to 2°C in winter to 17°C to 19°C in summer and the wind speed does not exceed 6.7 ms^{-1} (Moustafa and Zaghloul, 1996).

Nepeta septemcrenata belonging to Labiatae family is a perennial plant, near endemic, inhabiting stony wadis and mountains in Saint Katherine, South Sinai, Egypt and northwest Saudi Arabia (Boulos, 2002). A number of studies have scanned the phytochemical screening of *N. septemcrenata* and separated many chemical compounds (El-Hamouly and El-Hela, 2004; Abd El-Moaty, 2009; Moustafa *et al.*, 2015; Youssef and Mahgoub, 2015).

But the impact of altitudinal changes on chemical constituent of *N. septemcrenata* has not studied before except that in (Sharaf *et al.*, 2013) who determined the contents of soluble carbohydrates, soluble protein, proline and total phenols. They carried out those determinations through five different altitudes *viz.*, 1800, 2000, 2200, 2400 and 2600 m.a.s.l. at Gebel Saint Katherine for five species (*Nepeta septemcrenata*, *Seriphidium herba-album*, *Tanacetum sinaicum*, *Ballota undulata* and *Teucrium polium*) and found significant variations in the biochemical constituents of the plants among the different elevation ranks in all species.

The present paper will hopefully, through more lights on the chemical composition of *N. septemcrenata* as being affected by altitudinal variation of Saint Katherine Mountain.

Materials and methods

Samples collection of the studied area

Fresh samples of aerial parts of *N. septemcrenata* and associated soil were collected from five different altitudes *viz.*, 1600m, 1850m, 2100m, 2350m and 2600m along Saint Katherine Mountain passed by the beginning of Shaq Mousa till Wadi Arbaen as tributary during April, 2014. The soil samples supporting plants were collected from each level at 0-20 cm depths.These soil samples were air dried and then powdered gently with wooden wallet and passed through 2 mm sieve. The plant samples were weighted freshly then oven dried till constant weight after that ground to fine powder to determine the chemical constituents.

Soil analysis

Electrical conductivity (EC) and pH value were estimated in soil water extract (1:5) and moisture content was determined according to Rowell (1994) and determined as illustrated in Table 1.

Plant analyses

The degree of succulence was calculated according to fresh/dry weight as followed by (Dehan and Tall, 1978). Total and soluble carbohydrates were extracted and estimated calorimetrically by applying the general phenol-sulphoric acid method according to (Chaplin and Kennedy, 1994). Crude protein was estimated by Kjeldahl method as described by A.O.A.C (1990). Total lipids were extracted by equal volumes of petroleum ether: ether using soxhlet apparatus and calculated according to (Christie, 1982). Total phenolics was determined using Folin-Denis reagent as described by (Shahidi and Nacz, 1995). A known weight of plant sample was extracted by 80% ethanol, 1 ml of the extract, 0.5 ml of Folin reagent were mixed well, 1 ml of saturated Na_2CO_3 and mixed well then 3 ml of dist. water were added.

After 1 hour the developed blue color was read at 725 nm by spectrophotometer using catechol as a standard. Total ash content of the dry plant materials was determined after ignition at 500 ˚C in a muffle furnace for 6 hours, and the ash percentage was calculated according to A.O.A.C (1990). Sodium and potassium were measured by flame photometer as described by (Yoshida *et al.*, 1976). Calcium and magnesium were estimated by atomic absorption spectrophotometer using the method of A.O.A.C (1990). The chloride contents of the samples were determined according to (Jackson and Thomas, 1960).

Statistical analysis

The experiment included five altitudes, which were arranged in a randomized complete block design with three replications. Data obtained were analyzed according to COSTAT software program. Means values were differentiated using Duncan at 5% level as mentioned by Duncan (1955).

Results

According to Table 2, degree of succulence in *N. septemcrenata* attained the highest values in those growing at medium elevations (1850m, 2100m and 2350m) without significant change among them and the lowest value in those at 1600m altitude.

Table 1. Some chemical properties of soil supporting *N. septemcrenata*.

Altitudes	pH	EC µmhos	Moisture content%
1600m	7.4	308	3.3
1850m	7.5	240	2.65
2100m	7.5	255	2.64
2350m	7.5	302	2.11
2600m	7.7	292	2.98

Table 2. Degree of succulence, total carbohydrate, soluble and non-soluble carbohydrate in *N. septemcrenata*.

Altitudes	Succulence degree	Total carbohydrate %	Soluble carbohydrate %	Non-soluble carbohydrate %
1600m	3.07^{c}	18.00^{d}	0.83^{e}	17.18^{b}
1850m	4.41^{a}	19.43^{c}	2.38^{d}	17.05^{b}
2100m	4.23^{a}	20.25^{b}	3.90^{b}	16.35^{c}
2350m	4.28^{a}	21.98^{a}	3.76^{c}	18.22^{a}
2600m	3.80^{b}	19.72^{c}	4.96^{a}	14.76^{d}

Total carbohydrate in *N. septemcrenata* recorded the highest value at 2350m altitude followed by those at 2100m, 1850m, 2600m and the lowest value in those at 1600m altitude as shown in Table 2. Soluble carbohydrate recorded the highest value (4.96%) in those at the highest altitude 2600m and the lowest value (0.83%) in those at 1600m. Non-soluble carbohydrate was the highest in those growing at 2350m altitude followed by those at 1600m and 1850m without significant change between them and the lowest value in those at 2600m altitude.

Data in Table 3 indicated that, plants at the elevation of 1850m attained the highest crude protein followed by those at 2100m, while the lowest attained in those at 1600m. Total lipids increased gradually from1.4% at the lowest altitude (1600m) to 3.64% at the highest one (2600m). Total ash content was fluctuated with irregular trend ranged from 13.07% in those growing at 1850m to 11.15% in those growing at 1600m altitude. Also, total phenols fluctuated in plants among different altitudes, the highest value (3.68 mg/g dry wt.) in those at 1600m elevation and the lowest one (2.5 mg/g dry wt.) in those at 2100m elevation.

Concerning minerals composition, all tested elements had fluctuated changes among those growing at different altitudes as illustrated in Table 4. *Nepeta septemcrenata* growing at 1850m elevation recorded the highest accumulation in sodium, potassium and chloride, while those growing at 2100m elevation recorded the highest in calcium and magnesium contents.

Discussion

In the present study it is obvious that the soil is slightly alkaline and non-saline and this may be due to the high elevation with rocky mountain nature. The average moisture content indicates the presence of rainy season. The increase of succulence degree in *N. septemcrenata* growing at medium elevations and the decrease at the lowest elevation indicate that succulence degree is independent on supporting soil moisture content. Regarding the increase in total carbohydrate at high elevation Kumar *et al.* (2006) studied the effect of altitudes on the primary products of photosynthesis in varieties of barley and wheat growing at 1300m (low altitude) and 4200m (high altitude) elevations above sea level in the western Himalayas. They found that plants at high altitude had significant higher activities of phosphoenolpyruvate carboxylase which probably captured CO_2 directly from the atmosphere and/ or that generated metabolically e.g from photorespiration at high altitude.

Table 3. Crude protein, total lipids, total ash and total phenols contents in *N. septemcrenata*.

Altitudes	Crude protein %	Total lipids %	Total ash %	Total phenols mg/g dry wt.
1600m	9.05[e]	1.40[e]	11.15[e]	3.68[a]
1850m	12.93[a]	2.21[d]	13.07[a]	2.58[d]
2100m	11.37[b]	2.47[c]	12.84[b]	2.50[e]
2350m	10.21[d]	2.94[b]	11.85[d]	3.48[b]
2600m	10.49[c]	3.64[a]	12.61[c]	2.85[c]

The highest accumulation of total soluble carbohydrate in plants at the highest altitude agreed with Castrillo (2006) who found that soluble sugars content in *Espeletia schultzii* increased along the altitudinal gradients while protein content did not differ. Soluble sugars can protect the structural integrity of membrane during dehydration (Crowe *et al.*, 1988). A strong correlation between sugars accumulation and osmotic stress has been found (Clifford *et al.*, 1998; Patakas *et al.*, 2002).

Crude protein and total ash contents behave the same behavior, tend to increase at medium altitude and decrease at the lowest altitude. In this trend, Abd El-Maboud (2006) compared the ash content in *Deverra tortuosa* at up-, mid- and down-stream of Wadi El-Gafra and edited that *Deverra* of mid-stream site attained the highest ash content, while the up-stream site behaved intermediately and the lowest ash content was recorded in *Deverra* at down-stream site. Similar results were obtained by Mountousis *et al.* (2008) who found that crude protein contents in grazable herbs were 8.7, 10.9 and 9.73% in the lower, middle and upper altitudinal zone respectively, at NW Greece. On the other hand, the increase of nitrogen has been recorded in plants along the altitudinal gradients (Korner, 1989). Concerning total lipids, it increases directly as altitude increase response to stress resulted in lower temperature and anoxia. Similar results were obtained by (Pantis *et al.*, 1987). Guo *et al.* (2012) studied the changes in chemical composition of *Kobresia littledalei* at different altitudes and found that there was an increase of its crude protein, total lipids and soluble carbohydrate at high altitude. Total phenols in the studied plant have not straight trend along altitudinal gradients. Although, some authors found that defensive chemistry *viz.*, alkaloids, coumarins, phenolics and terpenes, generally decreasing as elevation increase (Erelli *et al.*, 1998; Alonso *et al.*, 2005).

Table 4. Sodium, potassium, calcium, magnesium and chloride contents in *N. septemcrenata*.

Altitudes	Sodium %	potassium %	calcium %	magnesium %	chloride %
1600m	0.633^{b}	0.166^{c}	2.370^{b}	0.760^{d}	0.358^{d}
1850m	0.687^{a}	0.208^{a}	2.580^{a}	0.803^{c}	0.578^{a}
2100m	0.500^{e}	0.125^{d}	2.600^{a}	1.040^{a}	0.423^{c}
2350m	0.566^{d}	0.194^{b}	2.036^{c}	0.810^{c}	0.427^{c}
2600m	0.592^{c}	0.128^{d}	2.393^{b}	0.903^{b}	0.443^{b}

Also, the increase of total phenolic compounds in *Sedum album* transplantation was noticed toward the high altitude site (Bachereau *et al.*, 1998).

The highest sodium, potassium and chloride contents at 1850m was associated with the highest ash content. The significant changes in minerals composition in *N. septemcrenata* were not correlated with the altitudinal gradients. The trivial variations in calcium content between plants growing at the highest altitude (2600m) and those growing at the lowest one (1600m) confirms that finding. In this trend, Soeth *et al.* (2008) found that Ca and Mg concentrations were not affected by altitudes in leaves of trees and shrubs growing in Ecuador at (1900m, 2400m and 3000m), while the concentrations of N and K decreased from 1900m to higher altitudes. It is evident from the results that calcium is the major among minerals composition in *N. septemcrenata* and this adverse the light on the importance of that element in preserving various structural roles in cell wall and membranes and control of ion exchange behavior. Besides its role as a second messenger in signal condition between environmental factors and plant responses (Garg, 1998; White and Broadley, 2003).

Conclusion

It can be concluded that total soluble carbohydrate and total lipids in *N. septemcrenata* are response positively along the altitudinal gradients. Calcium element is the major contribution among elements in building cell of *N. septemcrenata*.

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