

## Effect of NPK and Bio Fertilization on Growth and Oil Yield of Celery (*Apium graveolens*L.) and Dill (*Anethum graveolens* L.) Plants.

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### ABSTRACT

The present study was carried out in two successive seasons of 2014/2015 and 2015/2016 at the experimental farm of Horticultural Research Station, Sabhia. Alexandria governorate, Egypt to study the effect of NPK and biofertilizer on vegetative growth characters, vitamin C content and chemical composition of the essential oil of Celery and Dill plants. Results revealed that the fertilization treatments differently affected the mean values of all studied characters. The application with bio fertilizer (CPP) + 50% NPK significantly increased vegetative growth characters i.e plant height, , fresh and dry weight of aerial parts at two cuts as well as total chlorophyll and carotenoids, Vitamin C % during both seasons. However, the application biofertilizer (CPP) + 50% NPK gave the highest of major's chemical composition of essential oils i.e Apiol, Myristien,  $\beta$ . Pinene, Limonene and  $\beta$ - Phellandrene percentages during first cut of 2014/2015 season. This investigation suggests the need for more studies concerning the effect of NPK fertilizer and biofertilization on Celery and Dill plant under different environments using different types of soil to reach the optimum combination to achieve the best yield

**Keywords:** NPK, biofertilization (CPP), Celery, Dill, Vitamin C, major's compounds oil.

### INTRODUCTION

Celery (*Apium graveolens* L.) and Dill (*Anethum graveolens* L.) is an annual aromatic and medicinal plants belong to family Apiaceae (Umbelliferae) and are a widely cultivated herb used extensively for garnishing and seasoning foods, and for production of an essential oil. The celery and dill leaves are one of the most popular green herbs. It has a mild taste blends other flavors together, and has a high nutrient's content and used in medicinal, household cosmetic and fragrance (Rashed, 2002).

Fertilization is one of the most important factors limiting the productivity of plants. The intensive use of expensive mineral fertilizers in recent gears results in environmental pollution problems. However, Chemical fertilizers at extremely high rates for along period decreased the potential activity of microflora and the stability of soil organic matter (Hussien, 1995).

Recently, great attention has been focused on the possibility of using biofertilizer as a promising component of integrating nutrient supply system in agriculture. The term biofertilizer, represent everything from manures to plant extracts. "Biofertilizer" are those substances that contain living microorganisms and they colonize the rhizosphere of the plant and increase the supply or availability of primary nutrient and/or growth stimulus to the target crop. There are numerous species of soil bacteria that colonize mainly in the rhizosphere of plants. These bacteria are collectively known as plant growth promoting rhizobacteria (PGPR). Some PGPR promote the growth by acting as biofertilizer.

Microorganisms mainly nitrogen fixer, phosphate solubilizer and mycorrhizae are the main sources of biofertilizer. The microorganisms used for the biofertilizer are bacteria of *Bacillus*, *Pseudomonas*, *Lactobacillus*, photosynthetic bacteria, nitrogen fixing bacteria, fungi of *Trichoderma* and yeast. Biofertilizer have shown great potential as a, renewable and environmental friendly source of plant nutrient. Biofertilizer are ready to use and used as a live formulation of beneficial microorganisms, when it amended to seed, root or soil, it mobilizes the availability and utility of the microorganisms and thus

improves the soil health.

In general, bio-fertilizers are microbial preparations containing living cells of different microorganisms which have the ability to mobilize plant nutrients in soil from unusable to usable form through biological process. Bio-fertilizers are used in live formulation of beneficial microorganism which on application to seed, root or soil, mobilize the availability of nutrients particularly by their biological activity and help to build up the lost microflora and in turn improve the soil health in general (Ismail *et al.*, 2014). Their mode of action differs and can be used alone or in combination. Furthermore, biofertilization is an important factor being used to produce products without some mineral fertilizer that cause environmental pollution problems, and high rates of it leads to decrease the potential activity of microflora and the mobility of organic matters. Hence, the attention has been focused on the researches of bio- fertilization to safe alternative to specific chemical fertilizers. Biofertilizer play vital role for increasing the number of microorganisms and accelerate certain microbial process in the rhizosphere of inoculated soil of plants which can change the available forms of some nutrients to be plants (Kandeel *et al.*, 2001, Rashed, 2002; Mohamed and Abdu, 2004).

The present investigation was imposed to study the effects of biofertilizer alone or in combination with inorganic fertilize NPK on growth and oil composition of celery and dill plants. The goal was to minimize inorganic fertilize (NPK) usage.

### MATERIALS AND METHODS

This study was carried out during the two winter seasons of 2014/2015 and 2015/2016 at the Experimental Farm of Sabhia Horticulture Researches Station, Alexandria, Egypt, to study the effect of NPK and bio-fertilization treatments on growth and chemical composition of Celery and Dill plants. The experimental design was a randomized complete block design with three replicates.

The celery and dill fruits were sown on November 11<sup>th</sup> and 14<sup>th</sup> in the two growing seasons, respectively. The plots area of each was 4 m<sup>2</sup> (2.0m

×2.0m) with 3 rows; the distance between the rows was 50 cm and 10 cm between plants.

The used biofertilization of bacteria were phosphorein (*Bacillus megatherium phosphorus* dissolving bacteria P.D.B.), cerealine (*Azospirillum Lipoferum* and *Azotobacter chroococcum*), and potasiomag (*Basillus mucilaginosus*) (CPP) which supplied by ARC, Giza, Egypt. The inoculation, with Cerealine, phosphorein and potasiomag (CPP) was performed by coating celery and dill fruits with each

product individually using a sticking substance (Arabic gum at 5%) just before sowing.

The recommended dose of NPK (2:1:1) was divided in two equal parts, the first one was applied one month after sowing and the second one was applied after the first cut. The chemical fertilizers were applied as ammonium sulphate (20.5%N) at 300 kg/fed, calcium superphosphate (15.5% P<sub>2</sub>O<sub>5</sub>) at 150 kg/fed and potassium sulphate (48% K<sub>2</sub>O) at 150 kg/fed (recommended dose. This experiment included six treatments as follows:

T1:100%recommended dose of NPK (Control)	T4:Cerealine+Phosphorein+Potasiomag (CPP)
T2: 50% recommended dose of NPK	T5:Cerealine+Phosphorein+Potasiomag(CPP)+50%recommended dose of NPK
T3: 25% recommended dose of NPK	T6:Cerealine+Phosphorein+Potasiomag(CPP)+25%recommended dose of NPK

The physical and chemical characteristics of the experimental soil are given in Table (1). The soil was analyzed according to be methods described by Page *et al.* (1982)

At harvest dates on January 11th and February 12th (After 60 and 90 days from sowing) in the two seasons, guarded plants were randomly taken from each plot and the following characteristics were recorded:

- 1-Plant height (cm).
- 2-Fresh and dry weights of aerial parts/plant (g).
- 3-Total chlorophyll and carotenoids concentration (mg/g fresh weight) were determined in celery and dill fresh leaves samples of the fifth leaf from top after 60 and 90 days from sowing for celery and dill, using the method by Moran (1982).
- 4-Vitamin C content was determined in filtered juice samples and expressed as (mg ascorbic acid/100 ml fresh juice) as described by (A.O.A.C., 1965).
- 5-The percentage of major constituents (Apiole, Myristiein, β- pinene, and β- phellandrene).

**Essential oil analysis:**

The volatile oils were extracted separately by hydro-distillation method utilizing apparatus similar to European Pharmacopoeia (EP).The essential oils were analyzed with GC-MS (HP 8644) with flame ionization detector (FID) on a fused silica 132 capillary column DB-5, 25 m in length, 0.32 mm i.d., and 0.5 mm film thickness.133 Helium was used as the carrier gas with a flow rate of 1.6 ml/min; the detector 134 temperature was 260 °C, the oven temperature was programmed to increase from 130 to 260 °C at a rate of 4 °C/min. The split injector was heated at 250 °C, the split 136 ratio was 15:1. Data were processed on a DP 800 integrator. The percentage of major constituents were estimated by measuring the peak area of the different compounds of the chromatogram according to Heftman (1967) and Gunther and Joseph (1978).Sources of the principal components of volatile oils which used as reference for determined essential oil of celery and dill by GC were: Ciba Gigi, NY, USA.

**Table 1. The physical and chemical properties of the experimental soil in 2014/2015 and 2015/2016 seasons**

Season	1. Mechanical analysis							3. Macronutrients analysis		
	Sand	Silt	Clay	Soil texture	Organic matter	Calcium carbonate	pH (1:1)	N%	P (mg/kg)	K (mg/kg)
2014/15	16.00	38.00	46.00	Clay loam	1.01	8.00	7.80			
2015/16	16.80	39.20	44.00	Clay loam	0.92	7.90	7.90			

  

Season	2-Chemical analysis					3. Macronutrients analysis					
	EC (1:1) dS/m	Soluble Cations(1:2) (Cmol/kg soil)				Soluble Anions(1:2) (Cmol/kg soil)			N%	P (mg/kg)	K (mg/kg)
		Ca <sup>++</sup>	Mg <sup>++</sup>	Na <sup>+</sup>	K <sup>+</sup>	CO <sub>3</sub> <sup>+</sup> HCO <sub>3</sub> <sup>-</sup>	SO <sub>4</sub> <sup>-</sup>	Cl <sup>-</sup>			
2014/15	2.10	4.25	3.12	8.12	0.90	2.70	0.47	12.50	0.48	3.79	2.28
2015/16	2.15	4.20	3.15	8.17	0.95	2.57	0.50	12.69	0.45	3.83	2.33

**Statistical analysis:**

The experimental layout was a complete randomized block design with three replicates, each replicate contained 10 plants. Data from all experiments were subjected to analysis of variance using Costat Statistical Software (1986). Means of all data were compared by L.S.D. method according to (Snedecor and Cochran, 1968).

**RESULTS AND DISCUSSION**

**A- Growth parameters and photosynthetic pigments:**

The present results in tables (2& 3) have indicated that growth parameters of celery and dill plants were significantly affected by the application of fertilizer treatments in both seasons. In most cases,

application of treatment 50% NPK + bio fertilizers (CPP) and 100% NPK (CPP) increased significantly plant height, fresh and dry weight as well as total chlorophyll and carotenoids concentration at two cuts in both seasons. While, the lowest amount of growth occurred with the (CPP) treatment alone .The positive effect on growth characters and total chlorophyll and total carotenoids in response to bio fertilizers may be attributed to increasing maentration in plant tissues (Opera and Asigebu 1996). Also, the phosphate solubilizing bacteria (phosphorein) and nitrogen fixing (cerealine) may increase the synthesis of endogenous phytohormones i.e. IAA, GAs and CKs which play an important role in formation of a big active root system which allow more nutrients, uptake from the soil and

finally accelerated plant growth. The previous results agree, more or less, with the findings of Rashed (2002) on parsley, Gad (2001) on *Anelthum graveolens*; Mohammad *et al.* (2012) on *Pimpinella anisum*; Abdel-

Latif (2002) on *Caruim carvi* and Kandeel *et al.* (2001) and Mohamed and Abdu (2004) on *Foeniculum vulgare*.

**Table 2. Effect of fertilization treatments on celery and dill vegetative growth at two cut during 2014/15 and 2015/16 seasons**

Treatments	Plant height (cm)				Fresh weight (g) / plant				Dry weight (g) / plant			
	2014/15		2015/16		2014/15		2015/16		2014/15		2015/16	
	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut
<b>Celery</b>												
100% NPK (Control)	27.47	27.67	25.00	27.73	208.3	224.0	211.2	219.4	40.23	42.32	43.20	45.20
50% NPK	27.23	26.17	24.67	27.33	133.3	139.2	138.5	146.5	24.23	25.14	25.40	26.67
25% NPK	27.17	25.17	24.67	27.17	132.0	135.4	125.7	135.6	21.82	22.27	24.67	25.07
CPP	24.23	25.17	24.33	24.53	128.3	130.0	136.5	133.8	21.97	22.92	20.81	22.81
CPP + 25% NPK	26.73	26.33	25.83	25.73	166.6	170.0	159.3	164.7	33.99	32.33	31.75	33.75
CPP + 50% NPK	28.77	29.67	29.44	28.97	169.6	173.1	156.8	165.4	34.47	33.79	32.21	34.27
L.S.D. (0.05)	0.25	0.30	0.40	0.45	3.80	3.10	4.30	3.90	1.25	1.30	1.35	1.42
<b>Dill</b>												
100% NPK (Control)	29.23	28.67	30.07	29.23	231.6	228.0	225.1	213.4	47.23	44.67	44.20	42.20
50% NPK	25.23	27.81	28.48	27.73	193.3	180.0	190.5	181.6	34.23	33.29	35.40	33.40
25% NPK	26.81	26.57	25.21	26.17	181.1	178.0	184.2	179.2	33.97	34.17	32.81	30.81
CPP	26.53	25.17	26.83	24.53	183.0	178.0	180.7	176.2	33.73	33.03	32.17	31.67
CPP + 25% NPK	27.72	26.91	27.51	25.73	184.6	181.0	185.3	184.1	34.47	34.01	33.75	32.95
CPP + 50% NPK	28.35	27.97	28.44	28.07	191.6	186.1	193.4	185.9	38.77	36.67	37.20	35.20
L.S.D. (0.05)	1.25	0.93	1.08	0.95	4.30	3.19	5.63	4.90	3.25	2.98	2.95	2.77

**Table 3. Effect of fertilization treatments on celery and dill total chlorophyll and carotenoids (mg/g fw) at two cut during 2014/15 and 2015/16 seasons**

Treatments	Total chlorophyll (mg/g f w)				Total carotenoids (mg/g f w)			
	2014/15		2015/16		2014/15		2015/16	
	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut
<b>Celery</b>								
100%NPK (Control)	5.73	5.20	4.90	4.18	1.07	1.06	1.09	1.08
50% NPK	5.81	5.40	4.95	3.38	1.06	1.06	1.09	1.08
25% NPK	5.49	5.71	5.10	3.50	1.07	1.07	1.08	1.07
CPP	4.67	4.67	4.70	3.17	1.09	1.06	1.07	1.06
CPP + 25% NPK	5.75	5.75	5.11	3.36	1.07	1.07	1.09	1.07
CPP + 50% NPK	6.20	5.81	6.50	5.10	1.17	1.08	1.09	1.09
L.S.D. (0.05)	0.35	0.35	0.30	0.40	0.02	0.01	0.01	0.01
<b>Dill</b>								
100% NPK (Control)	5.77	5.76	4.90	3.98	1.08	1.07	1.08	1.07
50% NPK	5.63	5.59	4.95	3.51	1.07	1.06	1.06	1.07
25% NPK	5.59	5.54	5.17	3.44	1.06	1.05	1.05	1.06
CPP	5.33	4.51	4.84	3.14	1.06	1.03	1.05	1.06
CPP + 25% NPK	5.61	5.65	5.19	3.53	1.07	1.06	1.07	1.07
CPP + 50% NPK	5.92	5.81	5.53	4.16	1.10	1.09	1.09	1.08
L.S.D. (0.05)	0.35	0.35	0.30	0.40	0.01	0.01	0.02	0.01

**B- Oil % and vitamin C content:**

The data in Table (4) showed that all treatments of fertilization, affected oil percentage and vitamin C content in both seasons of celery and dill plants. It is clear from data that the highest mean values of oil percentage and vitamin C percentage, in most cases, resulted from the treatments of bio fertilizers (CPP) + 50% NPK in both seasons. The lowest oil % resulted with the (CPP) treatment without NPK.

The increment of vitamin C content of plants leaves using the treatments of biofertilizer (CPP) and half dose of NPK; may be attributed to increase in the occupancy root zone of plant as results of adding fertilization treatments which reflected on nutrients uptake by plants and confirm the previous of vegetative

growth. Similar results, more or less were obtained by Kandeel *et al.* (2001) and Abou El- Maged *et al.* (2008) on fennel plant.

**C- Major components percentage of essential oil:**

The effect of fertilization treatments on celery and dill major's compounds of oils i.e. Apiol, Myristien, β. Pinene, Limonene and β- Phellandrene percentages are shown in Tables (5). The results indicated that using fertilization treatments had significant effect on the studied major's compounds percentage of celery and dill oils. The application of biofertilizer (CPP) and half dose of NPK; gave the highest percentage of majors compounds in first cut of 2014/2015season. Similar results were reported by Darzi *et al.* (2011) on anisum and Ismail *et al.* (2009) on majoram plant.

**Table 4. Vitamin (C) content and oil (%) in celery and dill plants as influenced by fertilization treatments during 2014/15 and 2015/16 seasons.**

Treatments	Vitamin (C) (mg/100ml fresh Juice)				Oil %			
	2014/2015		2015/2016		2014/2015		2015/2016	
	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut
<b>Celery</b>								
100% NPK (Control)	111.9	107.1	111.4	110.3	0.72	0.69	0.76	0.73
50% NPK	109.1	106.3	110.2	107.9	0.66	0.67	0.63	0.62
25% NPK	109.5	105.1	110.7	106.8	0.62	0.64	0.63	0.63
CPP	107.3	102.0	109.3	106.2	0.62	0.63	0.62	0.62
CPP + 25% NPK	112.0	109.4	112.8	110.1	0.65	0.66	0.67	0.69
CPP + 50% NPK	112.4	110.9	114.2	112.0	0.76	0.73	0.78	0.74
L.S.D. (0.05)	2.40	2.02	2.30	2.10	0.02	0.02	0.01	0.02
<b>Dill</b>								
100% NPK (Control)	110.30	109.11	111.40	110.04	0.68	0.64	0.71	0.70
50% NPK	109.10	106.01	110.70	108.23	0.59	0.57	0.62	0.59
25% NPK	109.50	104.02	110.20	107.02	0.57	0.56	0.60	0.58
CPP	107.08	102.05	109.30	106.14	0.57	0.55	0.58	0.55
CPP + 25% NPK	112.30	109.40	112.70	110.14	0.60	0.57	0.63	0.58
CPP + 50% NPK	113.70	109.91	112.80	110.12	0.69	0.66	0.73	0.71
L.S.D. (0.05)	2.40	2.02	2.30	2.10	0.03	0.02	0.04	0.03

**Table 5. Effect of fertilization treatments on major components in Celery oil (%) of first season (1<sup>st</sup>cut)**

Treatments	Apiol (%)	Myristicin (%)	B- Pinene (%)	B- Phellandrene (%)
<b>Celery</b>				
100% NPK (Control)	17.5	34.5	14.1	5.8
50% NPK	17.5	33.4	14.2	5.3
25% NPK	17.4	33.2	12.5	5.3
CPP	17.0	31.4	12.3	5.1
CPP + 25% NPK	18.3	35.3	14.8	5.9
CPP + 50% NPK	18.8	36.2	15.1	6.0
L.S.D. (0.05)	<b>0.35</b>	<b>0.42</b>	<b>0.30</b>	<b>0.25</b>
<b>Dill</b>				
100% NPK (Control)	7.05	42.59	13.79	25.91
50% NPK	5.24	42.10	13.04	25.88
25% NPK	5.23	41.03	12.54	25.21
CPP	5.14	41.02	12.52	25.18
CPP + 25% NPK	6.24	42.29	13.78	25.95
CPP + 50% NPK	7.43	43.00	14.02	26.28
L.S.D. (0.05)	0.35	0.42	0.30	0.25

**CONCLUSION**

This study had shown that using biofertilizer (CPP) plus half a dose of inorganic fertilize NPK have resulted in the greatest plant growth and oil production of celery and dill plants.

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### تأثير السماد المركب NPK والتسميد الحيوي على النمو ومحصول الزيت لنباتى الكرفس والشبث زينب على أحمد قسم بحوث النباتات الطبية والعطرية - مركز البحوث الزراعية

أجريت هذه الدراسة في موسمين متتاليين خلال الموسم الشتوى لعامى من 2015/2014 و2016/2015 في المزرعة التجريبية لمحطة بحوث البساتين بمحافظة الإسكندرية، مصر و ذلك لدراسة تأثير السماد المركب NPK والتسميد الحيوي على صفات النمو الخضري والكيماوي ومكونات محتوى الزيت لنباتى الكرفس والشبث. وقد استخدم في هذه التجارب التصميم الإحصائي القطاعات العشوائية الكاملة لثلاث مكررات. وأظهرت النتائج أن استخدام معاملات التسميد أدى لتأثيرات معنوية مختلفة على أعلى متوسط لقيم جميع الصفات المدروسة أيضاً أدى إضافة المعاملة السماد الحيوى + 50% من السماد المركب (نتروجين- فوسفور- بوتاسيوم) إلى زيادة معنوية لارتفاع النبات ، الوزن الطازج والجاف عند الحشنتين بالإضافة إلى الكلوروفيل والكاروتين والمحتوي من فيتامين سي وكذلك النسبة المئوية للزيت العطري. وبالرغم من أن معاملة إضافة السماد الحيوى + 50% من السماد المركب (نتروجين- فوسفور- بوتاسيوم) تؤدي الى زيادة معنوية للمواد الفعالة الرئيسية لزيت الكرفس و الشبث (النسبة المئوية للأبيول والميريستين و بيتا بينين و الليمونين و بيتا فيلاندين) خلال الحشة الأولى بالموسم الأول 2015/2014. وتشير هذه الدراسة الحاجة الى المزيد من الدراسات بشأن تأثير السماد المركب والسماد الحيوى على نباتات الكرفس و الشبث تحت ظروف مختلفة باستخدام نوعية تربة مختلفة للوصول للخليط المثالى من السماد المركب و السماد الحيوى لتحقيق أفضل محصول

**الكلمات الرئيسية:** NPK - سماد حيوى - كرفس - شبث - محتوى فيتامين س - المكونات الرئيسية للزيت.