Effects of nitrogen source and effective micro-organisms on vegetative growth of shrubs of selected grapefruit (*Citrus paradisi* Macf) cultivars

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ABSTRACT

Grapefruit (Citrus Paradisi Macf.) is one of the most important citrus fruits in the Sudan. It can be successfully grown throughout the country where there are suitable soils and sufficient water to sustain tree growth. The objectives of this study were to find out the effects of different nitrogen sources, effective microorganisms (EM) and their combinations on the vegetative growth of shrubs of selected grapefruit cultivars. A field experiment was carried out at Umbarona, Wad Medani, Gezira State, Sudan, latitude 14.38 N, longitude 33.48 E and altitude 409 masl, in June 2012. Treatments consisted of three grapefruit cultivars: Marsh Seedless, Red Blush and Shamber. Fertilizer treatments were urea (100%), chicken manure (100%), urea with chicken manure (50% each), urea with EM, chicken manure with EM, urea (50%)+ chicken manure (50%)+ EM and unfertilized control. All fertilizer treatments were applied at the rate of 43 kg N/ha. Treatments were arranged in a split-plot design with two replicates. Grapefruit cultivars were allotted to the main plots and fertilizer treatments to the sub-plots. Data were collected on plant height (cm), stem diameter (cm), number of new branches, length of new branches (cm) and diameter of new branches (cm). Results showed that Marsh Seedless cultivar had the most vigorous vegetative growth compared to the other two cultivars. The results showed that the combination of urea, chicken manure and EM significantly gave the highest vegetative growth values, followed by chicken manure with EM, urea with chicken manure, urea with EM, urea alone, chicken manure alone and the least values of growth parameters were recorded for the unfertilized control. It is recommended to grow Marsh Seedless grapefruit cultivar and fertilize it with a combination of urea, chicken manure and EM.

INTRODUCTION

The grapefruit (*Citrus Paradisi* Macf.) is one of the most important citrus fruits in the Sudan. It can be successfully grown throughout the country where there are suitable soils and sufficient water to sustain tree growth. The quality of Sudanese grapefruits has been commended as superior to fruits in other citrus producing countries. The total production of grapefruit in the Sudan increased from 183000 tons in 2010 to 206000 tons in 2012 (FMAI, 2013).

Most of the traditionally grown grapefruit cultivars in the Sudan (Foster and Duncan) are seedy and do not compete in international markets. In the seventies of the last century, several grapefruit cultivars have been introduced to the Sudan and evaluated for their growth, yield and fruit quality (Ali Dinar and Osman, 1984; Sidahmed and Geneif, 1984b). Results indicated that the commercial seedless cultivars such as Miami and Red Blush were among the high yielding cultivars with an excellent fruit quality under Sudan conditions.

In recent years, citrus orchards in the Sudan have suffered from severe decline and low productivity. One of the factors which may have contributed to this decline is nutritional disorders. Nitrogen is one of the most important nutrients determining yield of citrus in the Sudan (Elkashif *et al.*, 2014; Elamin *et al.*, 2006; Ewida *et al.*, 2013). It is required by citrus in larger amounts than any other element (Ibrahim *et al.*, 2004). Soils of the arid tropics of the Sudan are generally deficient in nitrogen (SSAS, 1983). The use of nitrogenous fertilizers in citrus production is limited and farmers only apply random rates. General yellowing of old citrus leaves, which is an indication of nitrogen deficiency, can be observed in several Sudanese orchards (Osman, 2005).

Effective microorganisms (EM) is a mixture of a group of beneficial microorganisms that can coexist with each other. The main types of which are lactic acid bacteria, yeast, actinomycetes and photosynthetic bacteria. These microbes were blended in molasses or sugar medium maintained at a low pH (Higa and Parr, 1994).

Research work conducted on EM showed that the application of EM alone or in combination with other fertilizers enhanced the decomposition of organic matter and increased its efficiency, stimulated the activity of other microorganisms in the soil and hence increased plant growth and yield (Higa, 1991; Formoitz *et al.* 2007; Idris *et al.*, 2008).

Therefore, the objective of this study was to find out the effects of different nitrogen sources, effective microorganisms(EM) and their combinations on the vegetative growth of selected grapefruit cultivars.

MATERIALS AND METHODS

A field experiment was carried out at Umbarona nursery Wad Medani, Gezira State, Sudan, (latitude 14.38 N, longitude 33.48 E and altitude 409 masl) in June 2012. The objectives of this study were to find out the effects of different nitrogen sources, effective micro-organisms (EM) and their combinations on the vegetative growth of shrubs of selected grapefruit cultivars.

The soil of the experimental site was a silty soil, classified as Vertic, Ustifluvents, Fine loamy, mixed, isohyperthermic (SSAS, 1983). Treatments consisted of three grapefruit cultivars and seven fertilizer treatments.. Two years old shrubs of grapefruit cultivars Shamber, Red Blush and Marsh Seedless were used in this study and were budded on sour orange rootstock.

Fertilizer treatments were:

- 1. Control.
- 2. Urea (100%).
- 3. Chicken manure (100%).
- 4. Urea (50%) + chicken manure (50%).
- 5. Urea (100%) + effective micro-organisms (EM).
- 6. Chicken manure (100%) + EM.
- 7. Urea (50%) + chicken manure (50%) + EM.

Nitrogen sources were urea (46% N) and chicken manure (2.5%N) and their combinations. Fertilizers were applied at the rates of 0kgN/ha (control) and 43kgN/ha (Table 1).

Table 1. Nitrogen sources and corresponding amounts per hectare and per shrub, applied at the rate of 43 kg N/ha.

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-	Nitrogen source	Amount (kg/ha)	Amount (kg/ shrub)					
-	Control	Zero	Zero					
	Urea(100%)	94	0.6					
	Manure(100%)	1870	12					
_	Urea (50%)+manure (50%)	47 kg urea+935 kg manure	300g urea+6 kg manure					

Preparation of chicken manure compost

Chicken manure and dry leaves were placed in a pit one meter deep, watered weekly for 45 days, dried, dug up and analyzed. The result of the analysis of chicken manure compost showed that it contained 3.9% organic carbon and 2.4%N.

Effective micro-organisms solution was prepared by dissolving 1 ml of EM in 100ml of water and then applied at 50ml/seedling to the previously mentioned treatments.

Experimental design

Treatments were arranged in a split plot design with two replicates. Grapefruit cultivars were allotted to the main plots and nitrogen treatments to the sub-plots. Spacing between shrubs was $8m \times 8m$. Irrigation was applied at 7days interval except during the rainy season (July- October) where irrigation water was applied as needed. Weeds and insect pests were controlled as recommended.

Parameters studied

The following parameters were measured at monthly intervals:

- 1. Plant height (cm): It was measured at the soil surface to the tip of the shrubs.
- 2. Stem diameter (cm). It was measured at the soil surface.
- 3. Number of new branches.
- 4. Length of new branches (cm).
- 5. Branch diameter (cm).

Statistical analysis

Data were analysed using the standard analysis of variance procedure and means were separated according to Duncan's Multiple Range Test at 5% level of significance.

RESULTS AND DISCUSSION

Vegetative growth of grapefruit cultivars

There were significant differences in the vegetative growth parameters among grapefruit cultivars. There was a linear relationship between vegetative growth and time (months). This was most probably because temperatures were always above the optimum required for grapefruit growth and, hence, there was no period of cessation of growth. Marsh Seedless cultivar showed the highest values of plant height, stem diameter, number of new branches, length of new branches and diameter of new branches, compared to the other two cultivars which were comparable (Figs. 1-5).

These results are in agreement with those reported by Samson (1980) who showed that Marsh Seedless cultivar had the best vegetative growth and highest yield. He also found that fruits of Marsh Seedless cultivar had the highest juice content, which makes it a good choice for juice industry. Marsh Seedless is the parent grapefruit cultivar for all commercially seedless grapefruit cultivars. Pink Marsh and Red Blush appeared as bud mutations on Marsh Seedless. Ali Dinar and Osman. (1984) compared the performance of introduced grapefruit cultivars and found that Shamber cultivar had the least vegetative growth and lowest yield compared to other cultivars. They recommended a balanced fertilizer program in order to increase the productivity of citrus orchards in the Sudan. Sidahmed and Geneif (1984) reported that introduced grapefruit cultivars had more vigorous vegeta

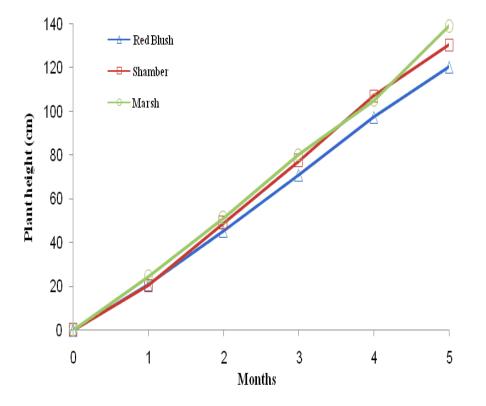


Fig. 1. Plant height of grapefruit cultivars.

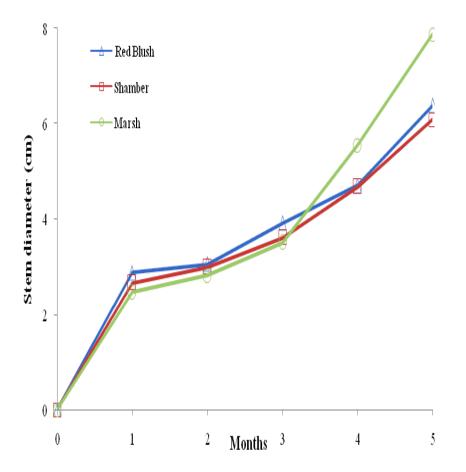
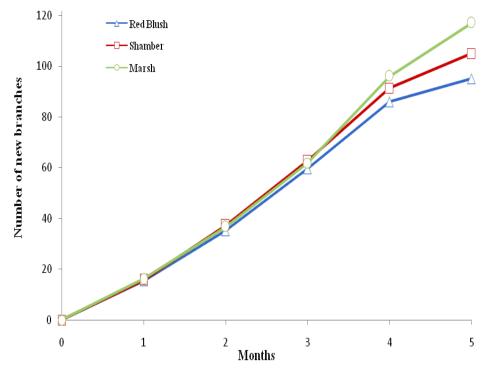
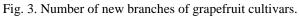


Fig. 2. Stem diameter of grapefruit cultivars.





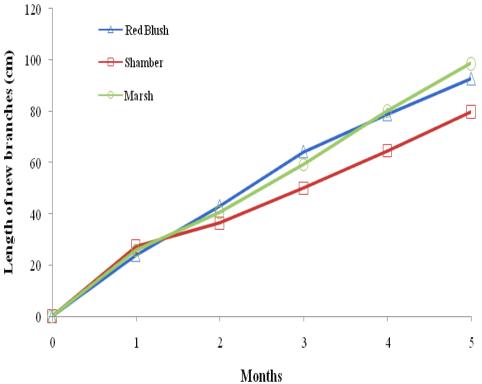
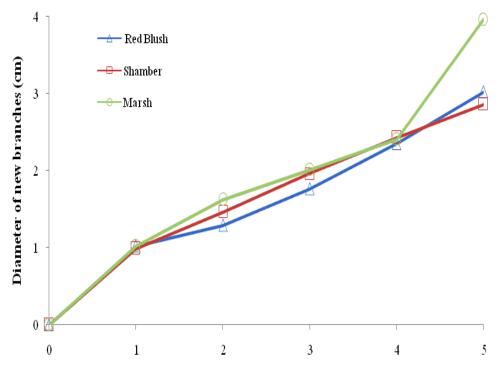


Fig. 4. Length of new branches of grapefruit cultivars



Months

Fig. 5. Diameter of new branches of grapefruit cultivars

Effects of fertilizer treatments on vegetative growth of grapefruit

The effects of fertilizer treatments on plant height, stem diameter, number of new branches, length of new branches and diameter of new branches of grapefruit cultivars were significant. The study clearly indicated the importance of nitrogenous fertilizers during the early growth of grapefruit shrubs. Results showed that the combination of urea, chicken manure and effective micro-organisms significantly gave the highest values, followed by chicken manure with effective micro-organisms, urea with chicken manure, urea with effective micro-organisms, urea alone, chicken manure alone and the least values of growth parameters were recorded for the control. (Figs.6-10).

These results clearly indicate that application of EM with chicken manure and/or urea increased their efficiency which was manifested in vigorous vegetative growth of grapefruit shrubs compared to treatments in which EM was excluded. Application of EM alone or in combination with other fertilizers has been reported to enhance the decomposition of organic matter, increased the efficiency of urea, stimulated the activity of other micro-organisms in the soil and, hence, increased plant growth (Higa, 1991; Kyan *et al.*, 1999). Idris *et al.* (2008) reported that application of EM (0.05%) to the soil coupled with chicken manure gave the best vegetative growth and highest yield of tomato.

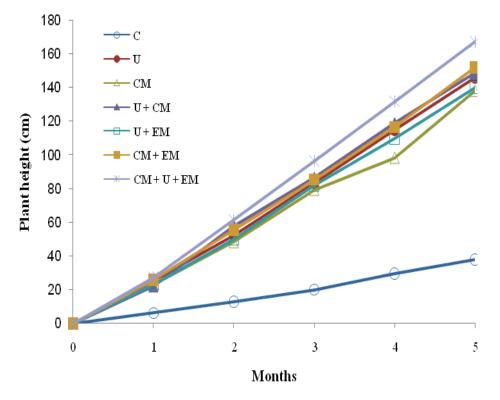


Fig. 6. Effects of fertilizer treatments on plant height.

C= control, U= urea, CM= chicken manure, U+CM = urea + chicken manure, U + EM = urea + effective microorganisms, CM+EM = chicken manure + effective microorganisms and U+CM+EM = urea + chicken manure + effective microorganisms.

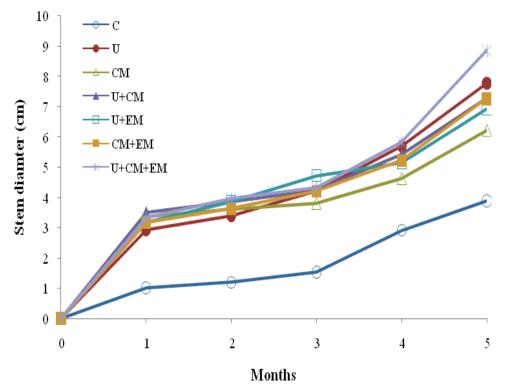
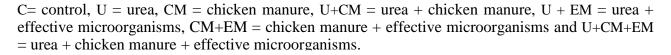
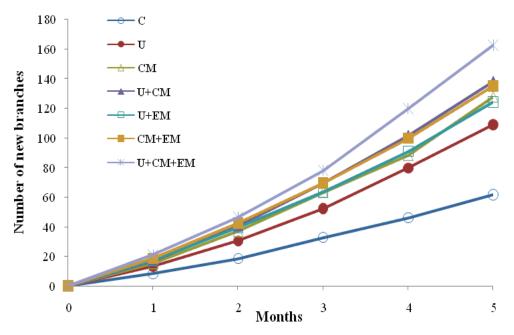
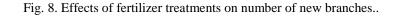


Fig. 7. Effects of fertilizer treatments on stem diameter.







C= control, U = urea, CM = chicken manure, U+CM = urea + chicken manure, U + EM = urea + effective microorganisms, CM+EM = chicken manure + effective microorganisms and U+CM+EM = urea + chicken manure + effective microorganisms.

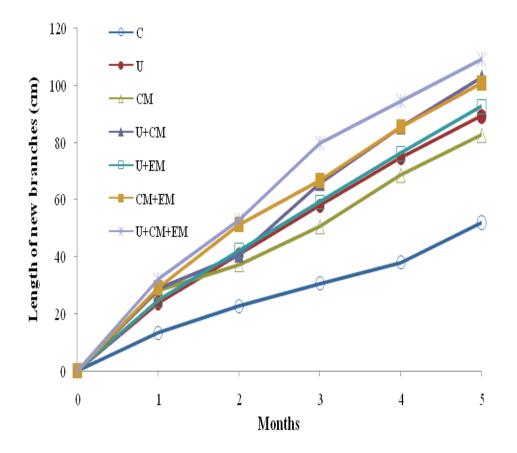


Fig. 9. Effects of fertilizer treatments on length of new branches.

C= control, U = urea, CM = chicken manure, U+CM = urea + chicken manure, U+ EM = urea + effective microorganisms, CM+EM = chicken manure + effective microorganisms and U+CM+EM = urea + chicken manure + effective microorganisms.

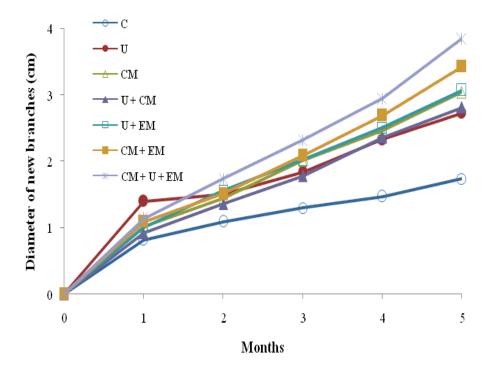


Fig. 10. Effects of fertilizer treatments on diameter of new branches

C= control, U = urea, CM = chicken manure, U+CM = urea + chicken manure, U+ EM = urea + effective microorganisms, CM+EM = chicken manure + effective microorganisms and U+CM+EM = urea + chicken manure + effective microorganisms.

Ibrahim *et al.* (2004) reported that application of chicken manure to muskmelons grown in heavy clay soils at the rate of 18t/ha significantly increased vegetative growth and resulted in the highest yield and the best fruit quality. Similarly, Elkashif *et al.* (2014) reported that application of chicken manure to banana cultivars grown in heavy clay soils improved the physical properties of the soil, increased the availability of micro nutrients and gave the highest yields. Ewida *et al.* (2013) showed that application of N fertilizer to mango trees in a combination of urea and cow manure at the rate of 86kg N/ha coupled with irrigation resulted the highest yields.

Interaction effects of fertilizer treatments and grapefruit cultivars on vegetative growth

The interaction effects of fertilizer treatments and grapefruit cultivars were significant on all vegetative growth parameters. For all grapefruit cultivars, the combination of urea, chicken manure and effective micro-organisms gave significantly the highest values, followed by chicken manure with effective micro-organisms, urea with chicken manure, urea with effective micro-organisms, urea alone, chicken manure alone and the least values of growth parameters were recorded for the control. (Table 2). These results indicated that the application of EM, which has been reported to enhance the decomposition of organic matter, increased the efficiency of chicken manure, stimulated the activity of soil micro-organisms and, hence, increased plant growth. Higa (1991) reported that application of EM in combination with chicken manure had positive effects on soil micro-organisms which

enhanced the decomposition of manure and made macro and micro nutrients available for plant uptake. Also, application of manure has been reported to improve soil physical and chemical properties, increased its porosity and water retention capacity (He *et al.*, 2003). Elkashif *et al.* (2014) noted that the new tend in agriculture in the Sudan should be directed towards the application of manures to increase both yield and quality of horticultural crops. The combination of EM with urea indicated that EM had beneficial effects on the efficiency of urea as evidenced by more vigorous vegetative growth compared to urea alone.

Results also showed that under all N fertilizer forms and combinations, the grapefruit cultivar Marsh Seedless resulted in the highest values of vegetative growth parameters which makes it a good choice for propagation and distribution to farmers for export purposes.

It could be concluded that the best vegetative growth was obtained when Marsh Seedless grapefruit cultivar was fertilized with a combination of urea, chicken manure and effective micro-organism

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Cultivars	Treatments	Н	SD	NNB	LNB	BD
Cultivars		(cm)	(cm)		(cm)	(cm)
RB	С	74.5 g	3.2 e	61.0 i	47.5 f	1.9 e
	U	136.5 e	7.1 c	109.5gh	92.5 c	3.0 b
	CM	129.5 f	5.9 d	129.5 de	89.5 c	3.2 b
	U+CM	139.0 e	7.2 c	144.5 c	123.5 a	2.8 bc
	U+EM	137.0 e	7.3 c	129.0 de	97.5 bc	3.1 b
	CM+EM	144.5 d	6.2 cd	119.5 fg	102.0 bc	3.0 b
	CM+U+EM	154.5 c	8.1 b	150.5 c	111.0 b	3.7 ab
	С	78.6 g	4.4 d	59.0 i	64.5 e	1.6 e
	U	148.5 d	6.8 cd	105.5 gh	80.0 d	2.4 cd
Sh	CM	148.0 d	5.9 de	123.5 ef	67.0 e	2.8 bc
	U+CM	169.5 b	6.7 cd	137.0 d	78.0 d	2.9 b
	U+EM	139.0 f	6.0 cd	120.0 fg	86.0 cd	3.1 b
	CM+EM	152.5 c	7.1 c	142.0 c	87.0 cd	3.6 ab
	CM+U+EM	166.5 b	8.0 b	161.0 b	96.5 c	3.8 a
	С	83.7 g	4.2 d	65.5 i	44.0 f	1.8 e
	U	153.0 c	9.6 a	112.5 gh	95.0 bc	2.8 bc
Μ	CM	137.5 e	6.9 cd	128.5 de	91.0 c	3.1 b
	U+CM	137.5 e	8.0 b	133.0 de	106.5 b	2.7 bc
	U+EM	144.5 d	7.5 c	123.5 ef	94.0 c	3.0 b
	CM+EM	160.0 b	8.6 b	144.0 c	113.0 ab	3.7 ab
	CM+U+EM	181.5 a	10.5 a	176.0 a	119.5 a	4.1 a
Sig.		**	**	**	**	**
SE <u>+</u>		3.4	0.28	2.99	3.6	0.06
CV (%)		6.21	10.00	5.98	9.92	3.05

Table 2. Interaction effects of grapefruit cultivars and fertilizer treatments on vegetative growth.

C= control, U= urea, CM = chicken manure, U+CM = urea+ chicken manure, U+ EM = urea + effective micro-organisms, CM+EM = chicken manure+ effective micro-organisms and U+CM+EM= urea+ chicken manure+ effective micro-organisms.

PH = Plant height, SD = Stem diameter, NNB = number of new branches, LNB = length of new branches, BD = branch diameter, RB=Red Blush, Sh=Shamber and M=Marsh Seedless.

Means followed by the same letters, in the same column are not significantly different according to Duncan's Multiple Range Test at 5% level.

** Significant at P=0.01.

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تأثير مصادر النتروجين والكائنات الحية الدقيقة الفعالة على النمو الخضري لشجيرات أصناف منتخبة من (*Citrus paradisi* Macf)

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الخلاصة

يعتبر القريب فروت من أهم ثمار الحمضيات في السودان ويمكن زراعته بنجاح في جميع أنحاء البلاد حيثما توفرت التربة المناسبة والمياه الكافية لنمو الأشجار. أجريت تجربة حقلية في ام بارونة، واد مدنى ولاية الجزيرة، السودان. تهدف هذه التجربة لمعرفة تأثير مصادر النتروجين المختلفة والكائنات الحية الدقيقة الفعالة ومزجها مع بعضها البعض على النمو الخضري لمعرفة تأثير مصادر النتروجين المختلفة والكائنات الحية الدقيقة الفعالة ومزجها مع بعضها البعض على السودان. تهدف هذه التجربة لشجيرات أصناف منتخبة من القريب فروت. قرر ولايش وشامبر وشامبر الشجير ات أصناف منتخبة من القريب فروت. تضمنت المعاملات ثلاثة أصناف من القريب فروت وهي مارش ورد بلش وشامبر وسيعة معاملات أسمدة وهي اليوريا، وزرق الدواجن، يوريا مع زرق الدواجن، يوريا مع زرق الدواجن، يوريا مع زرق الدواجن، يوريا مع الكائنات الحية الدقيقة الفعالة، زرق الدواجن مع الكائنات الحية الدقيقة الفعالة، يوري مع زرق الدواجن مع الكائنات الحية الدقيقة الفعالة، يوري مع الكائنات الحية الدقيقة الفعالة، يوريا مع زرق الدواجن مع الكائنات الحية الدقيقة الفعالة، يوريا مع زرق الدواجن مع الكل المعاملات توريا مع الكل المعاملات تلائة أصناف من القريب فرات أسامدة وسي معدل لالقوية الفعالة، يوريا مع زرق الدواجن مع الكل الحية الدوية العالة والشاهد. تمت إضافة الأسمدة الدواجن مع الكل المعاملات بعدل إلى المن المربي فروت هي المواجن مع الكل المعاملات بعدل 43 كجم نتروجين للهكتار. صممت التجارب بطريقة القطع المنشقة بحيث كانت أصناف القريب فروت هي القطع الرئيسة ومعاملات النتروجين هي القطع الثانوية. جمعت بيانات عن طول النبات (سم) وقطر الفرع (سم). وعدد الأفرع الحدية التابي أوضي عاصر أوض أعلى أعلى نمو خضري ويلا وشام وشام وشامر وسما وشام وشام أعلى منو خضري معار وشام وشام وشام وشامين روضا أوض وشعان وروض في في ماسودان ويلانات على أعلى مورف أوض وشام وشام وشامبر أوض التربة. ورق الدواجن والكائنات عن طول النبات (سم) وقلى أوض المام وروب ويلا فرن وروت ها ولكن المندة ولوت م ألفري وسمى أول النور وسما وروض أوش وربعة وطول الفرع (سم) وقط الفرع (سم). أوضحت النتائج أن الصنف مارش أعطى أعلى نمو خضري معار أوض بالمنو ورق الدواجن وروق الدواجن وروق الدواجن وروا الدوية الغالة أوطان أعطي أعلى مار وروق الدواجن ورو وروق الور الحيوب المويم أول وروى الوور