

ORIGINAL ARTICLES

Growth Analysis and Chemical Constituents of Garlic Plants in Relation to Morphological Growth Stages

Abou El-Magd, M.M., M.F. Zaki, Faten S. Abd El-Al and E.H. Abd El-Samad

Vegetables Crop Research Dept., National Research Centre, Dokki, Giza, Egypt

ABSTRACT

Field experiments were conducted for two years during winter season of 2009/2010 and 2010/2011 at a private farm in El-Khanater, Kalubia Governorate, Egypt, to analysis the vegetative growth and yield parameters, i.e. plant height, number of leaves per plant, neck and bulb diameter, bulbing ratio, fresh and dry weight of leaves, bulb and whole plant, total green yield and chemical properties, i.e. leaf pigment contents, chlorophyll (a, b and total) and carotenoids, bulb total carbohydrates and nutrients uptake (N, P and K) in both leaves and bulbs as well as growth attributes, i.e. leaf area, leaf area ratio, leaf weight ratio, specific leaf area, absolute growth rate, relative growth rate and net assimilation rate of garlic plants cv. Balady at different sampling dates, i.e. 90, 120, 150 and 180 days after sowing in relation to morphological growth stages using complete randomized block design with four replications. The obtained results pointed out that there were pronounced significant differences increase in plant height, number of leaves per plant, neck and bulb diameter, fresh and dry weight of leaves, bulb and whole plant and total green yield with the advancement of plant age (different sampling dates). Whereas bulbing ratio was steadily decreased with increasing plant age in both seasons. Concerning leaf growth attributes and growth attributes, leaf area, absolute growth rate, relative growth rate and net assimilation rate of garlic plants were increased gradually and achieved the highest values by the fourth sampling date (180 days after sowing). While leaf area ratio, leaf weight ratio and specific leaf area were decreased steadily until the attainment of a minimum values at 180 days after sowing. There were significant differences among different sampling dates in both season of study. Garlic leaf pigment contents, chlorophyll a, b and total were significantly increased and reached the highest values at 120 days after sowing. From that time, the chlorophyll content began to decrease sharply when the last sample was taken (150 days after sowing). Carotenoids in leaves and total carbohydrates in bulbs were significantly increased with the advancement of plant age. Carotenoids content were negatively correlated with chlorophyll. Nutrients uptake (N, P and K) in both leaves and bulbs were significantly increased over the time and achieved the highest values at 150 days after sowing. Nutrients uptake in garlic leaves and bulbs followed the same pattern in both seasons.

Key words: Garlic, Vegetative growth, Growth attributes, Leaf pigments, Total carbohydrate, N, P and K uptake.

Introduction

Garlic (*Allium sativum* L.) is the second most widely used cultivated bulb crops after onion in Egypt and it is also important foreign exchange earner. The centre of origin of garlic has been considered to be Central Asia, with secondary centers of diversification in China and the Mediterranean area (Etoh and Simon, 2002) and it was grown in Egypt in 2780 B.C. (Yamaguchi, 1983). It has been used for culinary and medicinal purposes by many cultures for centuries. It is still being employed in folk medicine all over the world for the treatment of a variety of diseases (Ali *et al.*, 2000). Garlic is a particularly rich source of organo-sulfur compounds, namely, allyl cysteine sulphoxides (ACSO) and their intermediate metabolites and derivatives (Randle and Lancaster, 2002 and Jabbes *et al.*, 2012), which are thought to be responsible for its flavor and aroma, as well as its potential health benefits (Augusti and Mathew, 1974; Block, 1985 and Tapiero *et al.*, 2004).

The term of plant growth analysis is refers to a useful set of quantitative methods that describe and interpret the performance of whole plants system. Plant growth analysis provides an explanatory, holistic and integrative approach to interpreting plant form and function. It uses simple primary data such as weights, areas, volumes and contents of plant or plant components to investigate processes within and involving the whole plant or crops (Hunt, 2003). Figuring out or characterizing of ontogenetic stages and measurement of plant growth parameters are important morpho-physiological basis for increasing garlic plant growth and productivity (Argüello *et al.*, 1997).

Like other crops the productivity of garlic depends on such growth attributes as leaf area index, crop growth rate, relative growth rate and net assimilation rate. In addition, growth analysis is a useful technique in the study of plants, because it bridges between the empirical and the mechanistic approaches to modeling growth and development, a necessary prerequisite to the understanding of the whole behavior of the plants (Brand *et al.*, 1987). The growth and developmental stages of *Allium sativum* include four stages, clove sprouting, shoot growth, bulb growth and bulb maturation as indicated by Del Pozo and González (2005) and Ledesma *et al.* (1997). Clove sprouting and emergence are controlled mainly by temperature (Takagi, 1990). Garlic plant growth and bulb formation depend on an increase in total soluble carbohydrates which depends on the photosynthetic activity of the leaves, a factor affected by light (Argüello *et al.*, 1997). It is well known that bulb initiation or bulb formation in garlic largely depend on the day length and temperature prevailing during growth period (Ledesma *et al.*, 1997 and Rahman *et al.*, 2004). Whereas, bulb development in garlic depends on the adequate translocation of photosynthetic substrates from the leaves to the bulb. Warm temperature has been identified to promote translocation of photosynthetic substrates from leaves to cloves (Kamenetsky *et al.*, 2004). A cooler temperature is needed to enhance shoot growth while a higher light intensity is needed to enhance bulb growth in garlic plant (Argüello *et al.*, 1997).

Net assimilation rate (NAR), is the measure of gaining in dry matter per unit of leaf area per unit of time and it is an important index of mean photosynthetic efficiency of a crop. The higher values of NAR at the latest growth stages might be related to the rapidly developing sinks, i.e. bulbs as in garlic and onion (Rahman *et al.*, 2004).

Ledesma *et al.* (1997) observed that the maximum values for leaf area index (LAI), net assimilation rate (NAR), relative growth rate (RGR) and total soluble carbohydrates (TSC) parameters were reached at garlic shoot growth stage. Translocation of photosynthetic substrates to the bulb was begun afterwards. They also added that during bulb growth stage, there were an increase in dry weight (DW) and TSC. Bulb filling took place 140 days after sowing date and continue until harvest stage. Harvesting stage is characterized by a minimum shoot DW, maximum bulb DW, minimum bulbing index (BI), maximum TSC and maximum soluble proteins. While, Muro *et al.* (2000) indicated that garlic leaf parameters (leaf number per plant, leaf area and leaf mass) reached their maximum values three weeks before harvesting stage. Leaf number per plant is an important factor for obtaining good results in crop production. Other factors, however, play important roles such as genotype and climate conditions. Leaf number per plant, as a rule, increases up to a certain point in plant development and then begins to decrease (Dalen, 1992 and Lewis *et al.*, 1995). In addition, Moravčević *et al.* (2011) stated that garlic plant develops 8 to 14 leaves and LAI is a very good indicator of a plant's photosynthetic capacity and is also very important in determining the levels of photosynthetic activity and yield. In the same respect, Haque *et al.* (2002) suggested that there was a positive correlation between crop growth rate and the leaf area index.

Argüello *et al.* (1997) reported that no significant changes in nutrient were observed during clove sprouting stage. Nutrient changes become important between the initiation of shoot growth (30 days after sowing date) and at the beginning of the bulb filling stage (140 days after sowing date). Maximum shoot growth and total soluble carbohydrates were reached at this stage. Also at this point, the sink-source relation between shoot and bulb is inverted, and the first signs of leaf senescence become evident.

The purpose of this work was to analysis of vegetative growth and yield parameters as well as growth attributes and some chemical properties of garlic plants at different sampling dates in relation to morphological growth stages.

Material and Methods

Field experiments were conducted for two years during winter season of 2009/2010 and 2010/2011 at a private farm in El-Khanater, Kalubia Governorate, Egypt to analysis the vegetative growth and yield parameters, i.e. plant height, number of leaves per plant, neck and bulb diameter, bulbing ratio, fresh and dry weight of leaves, bulb and whole plant, total green yield and chemical properties, i.e. leaf pigment contents, chlorophyll (a, b and total) and carotenoids, total carbohydrates and nutrient contents (N, P and K) in both leaves and bulbs as well as growth attributes, i.e. leaf area, leaf area ratio (LAR), leaf weight ratio (LWR), specific leaf area (SLA), absolute growth rate (AGR), relative growth rate (RGR) and net assimilation rate (NAR) of garlic plants cv. Balady at different sampling dates, i.e. 90, 120, 150 and 180 days after sowing in relation to morphological growth stages.

Soil samples were randomly collected from experimental site before planting and during land preparation from the top layer (0 - 30 cm) for soil physical and chemical analysis. Soil physical properties were analyzed using the procedures described by Black *et al.* (1981), while soil chemical analysis was determined according to the procedures described by Jackson (1973). Soil physical and chemical properties are presented in Table (1). While, chemical analysis of organic manure (poultry manure) added to the soil are presented in Table (2).

Table 1: Physical and chemical properties of the experimental soil.

A. Physical properties									
Sand %		Clay %			Silt %		Texture		
21.26		49.55			29.19		Clay loam		
B. Chemical properties									
E.C. mmhos /cm	pH	Meq/L							
		Cations				Anions			
		Ca ⁺⁺	Mg ⁺⁺	Na ⁺	K ⁺	CO ₃	HCO ₃	Cl	SO ₄
1.45	7.82	2.35	2.55	3.42	0.41	Nil	2.20	1.91	4.43

Table 2: Chemical analysis of used poultry manure during both seasons of 2009/2010 and 2010/2011.

Season	E.C.	pH	N %	P %	K %	Fe ppm	Mn ppm	Zn ppm	Cu ppm	Pb ppm
1 st	2.72	8.56	2.80	0.91	1.40	2400	342	140	97	1.01
2 nd	3.11	8.15	2.64	1.80	2.37	2300	266	180	36	1.20

The metrological data for the experimental area were also obtained from Central Laboratory for Agricultural Climate (CLAC), Agricultural Research Center (ARC), values were calculated and expressed as monthly interval means during the two growing seasons as shown in Table (3).

Uniform seed cloves obtained from outer ring cloves of bulb, free of bruises or infections with an average weight of 1.6 - 2.0g of locally available cultivar of garlic Balady were sown in the experimental field on the last week of September in both growing seasons of 2009/2010 and 2010/2011. Seed cloves were planted by hand on both sides of flood irrigated ridge of 70 cm width, 5.0 m length and 10 cm apart. Each experimental block included 3 ridges with a net area of 10.5 m².

Table 3: Monthly maximum and minimum air temperatures, relative humidity, and average daylight at the experimental area during both seasons of study.

Month	Average maximum temperature (°C)	Average minimum temperature (°C)	Relative humidity (%)	Average daylight hours/day
First season (2009/2010)				
September	30.9	18.8	44	12.4
October	29.3	18.1	40	11.5
November	25.8	16.3	41	10.9
December	21.2	12.4	45	10.0
January	18.8	9.7	41	10.4
February	19.7	9.4	40	11.3
March	27.5	12.7	32	12.0
April	29.5	13.0	34	13.0
Second season (2010/2011)				
September	29.7	17.9	46	12.4
October	28.4	17.4	44	11.5
November	23.7	15.8	41	10.9
December	21.0	11.3	36	10.0
January	17.5	10.3	31	10.4
February	18.7	8.7	37	11.3
March	25.2	11.2	33	12.0
April	26.6	13.4	34	13.0

The full amount of organic manure and phosphorus as well as one third dose of nitrogen fertilizer were applied at the time of final land preparation at rates of 10 m³ poultry manure, 150 kg/fed. calcium super phosphate (15.5% P₂O₅) and 80 kg/fed. ammonium nitrate (33.5% N), respectively. The remaining nitrogen amount (160 kg of ammonium nitrate) was split into two equal doses and side-dressed at 30 and 75 days after sowing date. Also potassium sulphate (48% K₂O) as a source of potassium was added at a rate of 100 kg/fed. This amount was divided into 2 equal portions and side-dressed at 90 and 120 days after sowing date. Standard agricultural practices for garlic crop production in the growing area, Delta region such as irrigation, top dressing, hoeing and protection against pests and diseases were carried out as recommended by Ministry of Agriculture.

Experimental design and statistical analysis:

The experimental design was a complete randomized block design with four replicates. Seed cloves were planted and plants were randomly sampled 90, 120, 150 and 180 days after sowing date. The obtained data were tabulated and subjected to statistical analysis and mean separation was done using the least significant differences (LSD) test at P<0.05 level of probability according to the method described by Snedecor and Cochran (1980).

*Data recorded:**A. Growth analysis:*

Ten plants were randomly uprooted from each experimental block every 30 days starting at 90 days after sowing date, i.e. 90, 120, 150 and 180 days to measure the following vegetative growth parameter:

- | | |
|--|----------------------------------|
| 1- Plant height (cm). | 2- Number of Leaves per plant. |
| 3- Leaf area (cm ²). | 4- Bulb diameter (cm). |
| 5- Neck diameter (cm). | 6- Bulbing ratio. |
| 7- Fresh weight of leaves, bulb and total plant (g). | 8- Total green yield (ton/fed.). |
| 9- Dry weight of leaves, bulb and total plant (g). | |

B. Growth attributes:

The following growth analysis attributes, i.e. leaf area ratio (LAR), leaf weight ratio (LWR), specific leaf area (SLA), absolute growth rate (AGR), relative growth rate (RGR) and net assimilation rate (NAR) were calculated using data set generated from different sampling dates of 90, 120, 150 and 180 days after sowing date according to Watson (1958) and Hunt (2003).

C. Chemical constituents:

1- Leaf pigment contents, chlorophyll (a, b and total) and total carotenoids in the fourth leaf of garlic plant were determined on each sampling date according to the method described by Devlin and Barker (1971).

2- Nutrient contents, on each sampling date, samples of garlic plant leaves and bulb were oven dried at 70°C for 72 hours and used to determine ion contents on a dry weight basis. Afterward, dried samples were powdered and wet digested as described by Wolf (1982). The percentages of nitrogen and phosphorus were determined using the modified micro Kjeldah method and colorimetric method using spectrophotometer (SPECTRONIC 20D, Milton Roy Co. Ltd., USA), respectively, according to the procedure described by Cottenie *et al.* (1982). While, potassium percentage was measured using flame photometer method (JENWAY, PFP-7, ELE Instrument Co. Ltd., UK) as described by Chapman and Pratt (1982).

3- Total carbohydrates content, in a dried sample of garlic bulb in each sampling date, total carbohydrates were measured according to procedure described by Dubois *et al.* (1965).

Results and Discussion*A. Growth analysis:**Plant height:*

There was a rapid statistically significant increase in plant height parameter among different sampling dates, i.e. 90, 120, 150 and 180 days after sowing expressed the different developmental stages of garlic plants in both seasons of 2009/2010 and 2010/2011 as presented in Table (4). The obtained values for plant height clearly interpreted that such parameter was characterized by a rapid gradually increment during sampling dates from 90 to 150 days after sowing then followed by a slow increment during the period from 150 till 180 days after sowing. At 90 days after sowing plant height value recorded 59.53 and 57.03 cm and reached 97.17 and 84.07 cm at 180 days after sowing in the first and second seasons, respectively.

Planting garlic plant during September gave the highest values of plant height because these plants received longer warm period and day light (30.9, 29.7°C and 18.8, 17.9°C and 12.4 hours/day, average maximum and minimum air temperatures and day light in the first and second seasons, respectively) which possibly enhanced meristematic elongation of plant and resulting in a maximum plant height.

These results are in good accordance with Abou El-Magd (1979); Soliman (1999) and Rahman *et al.* (2004) on garlic. In the same respect, warm temperature has been identified to promote photosynthetic activities and translocation of substrates within the plant (Kamenetsky *et al.*, 2004). Also a cooler temperature is needed to enhance garlic shoot growth (Argüello *et al.*, 1997).

Leaf number per plant:

Number of leaves per plant was significantly increased during different sampling dates except at the last sampling date 180 days after sowing. These results were true and similar in both seasons of study. The highest

number of leaves per plant was reached at 150 days after sowing, afterwards, leaves number per plant start to decrease at 180 days after sowing (Table 4).

The obtained results are in conformity with Dalen (1992) and Lewis *et al.* (1995), they stated that leaf number per plant, as a rule, increases up to a certain point in plant development and then begins to decrease. Other factors, however, play an important role such as genotype and climate conditions. Number of leaves per plant in garlic reached their maximum values three weeks before harvesting stage and then decreased. It is an important factor for obtaining good results in crop production (Muro *et al.*, 2000). In addition, Moravčević *et al.* (2011) stated that garlic plant develops 8 to 14 leaves.

Number of leaves per plant was adversely affected by low night temperature during the coldest months (December, January and February) prevailing during vegetative growth period (Nyengedzeni, 2010).

Nick and bulb diameter, and bulbing ratio:

Data presented in Table (4) clearly demonstrated that there was a gradual increased of neck and bulb diameter during the different developmental stages (sampling dates) in both seasons of 2009/2010 and 2010/2011. The significant increase in values of bulb diameter was superior than values of neck diameter. However, values of bulbing ratio were significantly decreased with increasing bulb diameter, this is due to bulbing ratio or bulbing index was calculated by dividing neck diameter over bulb diameter. Consequently, the value of bulbing ratio was decreased with sampling date from 90 to 180 days after sowing as a result of increasing bulb diameter. These findings were true in both seasons of study.

The maximum increase in bulb diameter and the maximum decrease in bulbing ratio were took place in the third sampling date (150 days after sowing), it reached 46.6, 46.5% and -68.8, -79.3% for bulb diameter and bulbing ratio in the first and second seasons, respectively.

The obtained results are in agreement with Ledesma *et al.* (1997) who pointed out that bulb filling took place 140 days after sowing date and continue until harvesting stage. Harvesting stage is characterized by a minimum bulb index.

Many investigators have been reported that bulb initiation or bulb formation in garlic largely depend on the day length and temperature prevailing during growth period, short day and lower temperatures stimulate plants to initiate or format bulb (Ledesma *et al.*, 1997; Rahman *et al.*, 2004; Kulkarni *et al.*, 2005 and Nyengedzeni, 2010). In addition, warm temperature has been identified to promote translocation of photosynthetic substrates from leaves to the bulb, resulted in enhancing of bulb growth (Kamenetsky *et al.*, 2004).

Fresh and dry weight:

The effect of different sampling dates, i.e. 90, 120, 150 and 180 days after sowing on the fresh and dry weight of garlic plant and its different organs (leaves and bulb) were shown in Table (4). Fresh and dry weight of leaves, bulb and whole plant of garlic were significantly increased with increasing plant age from 90 to 180 days. The highest values for garlic plant and its different organs fresh and dry weight were recorded at the fourth sampling date (180 days after sowing), while the lowest values were recorded at the first sampling date (90 days after sowing). The maximum percentage of increment in plant fresh and dry weight was found between the second and third sampling dates, it reached about 60.5, 53.9% and 57.1, 62.3% for total plant fresh and dry weight and 76.1, 69.7% and 80.5, 81.6% for bulb fresh and dry weight in the first and second seasons, respectively. While, the maximum percentage of increment for leaves fresh and dry weight was found between the third and fourth sampling dates in both seasons of study. Leaves fresh and dry weight increment percentage reached about 40.0, 40.6% and 53.2, 52.7% for leaves fresh and dry weight in the first and second season, respectively.

In general, it could be noticed that there was a great gain in fresh and dry weight of garlic plant during the last 30 days especially for bulb. This may be due to that at 180 days after sowing the climate conditions were as follow 27.5 and 25.2°C the average maximum temperature and 12.7 and 11.2°C the average minimum temperature and 12 hours/day the average daylight. Such climate conditions may be increased photosynthetic activity of the leaves, a factor affected by light and temperature and translocation of photosynthetic substrates to bulb which act as a sink. Moreover, the different between day and night temperatures was in favor of plant growth and development.

Bulb growth and development depends on the adequate translocation of photosynthetic substrates from leaves to the bulb. Warm temperature has been identified to promote translocation of photosynthetic substrates from leaves to bulb (Kamenetsky *et al.*, 2004). Also, a higher light intensity is needed to enhance bulb growth in garlic plant (Argüello *et al.*, 1997). Moreover, Ledesma *et al.* (1997) stated that the translocation of photosynthetic substrates to the bulb was begun after shoot growth stage. They also added that during bulb growth stage, there were an increase in bulb dry weight such increase reached to a maximum at harvesting stage.

Table 4: Effect of different sampling dates on vegetative growth parameters and total green yield of garlic plants during both seasons of 2009/2010 and 2010/2011.

Sampling dates (days)	Plant height (cm)	Leaves number/plant	Bulb diameter (cm)	Neck diameter (cm)	Bulbing ratio	Fresh weight (g/plant)			Dry weight (g/plant)			Total green yield (ton/fed.)
						Leaves	Bulb	Total	Leaves	Bulb	Total	
First season (2009/2010)												
90	59.53	6.57	1.53	0.87	0.57	21.05	5.15	26.20	2.68	0.78	3.46	0.686
120	70.70	7.00	2.07	1.12	0.54	24.26	9.17	33.43	5.24	1.28	6.53	0.802
150	87.00	7.80	3.88	1.24	0.32	46.32	38.33	84.66	8.64	6.58	15.22	2.032
180	97.17	7.77	5.37	1.36	0.25	77.21	64.37	141.58	18.46	16.58	35.03	3.398
L.S.D at 0.05	4.32	0.38	0.13	0.15	0.07	1.81	3.97	4.90	2.53	0.86	2.49	0.160
Second season (2010/2011)												
90	57.03	6.73	1.60	0.90	0.56	21.80	8.33	30.13	2.62	0.78	3.40	0.723
120	68.83	7.10	2.07	1.07	0.52	25.33	11.27	36.60	4.36	1.25	5.62	0.878
150	79.57	7.87	3.87	1.10	0.29	42.21	37.25	79.46	8.12	6.81	14.92	1.907
180	84.07	7.87	5.10	1.16	0.23	71.05	63.57	134.62	17.18	15.81	32.99	3.231
L.S.D at 0.05	5.68	0.45	0.18	0.12	0.04	1.54	2.59	3.16	0.84	0.91	1.28	0.076

Total green yield:

Total green yield of garlic plants was widely affected by plant ages in both seasons of 2009/2010 and 2010/2011 as shown in Table (4). Total green yield of garlic was increased significantly with increasing plant age. The highest value of total green yield (3.398 and 3.231 ton/fed.) was obtained with the fourth sampling date (180 days after sowing), while the lowest value (0.686 and 0.723 ton/fed.) was obtained with the first sampling date (90 days after sowing) for the first and second season, respectively.

The obtained results sharply demonstrated that the most increment in total green yield of garlic plant was occurred between the third and the fourth sampling date in both seasons of study. This may be due to a rapid development of bulb as a sink. Also, the sink-source relationship between shoot and bulb is inverted towards bulb growth and maturation as well as a fast translocation of photosynthetic substrates to bulb. In addition, it is a bulb filling stage. Similar results were found by many authors (Abou El-Magd, 1979 and Soliman, 1999) on garlic plants.

The translocation of photosynthetic substrates to the bulb was begun after the termination of shoot growth stage. During bulb growth stage, there was an increase in bulb dry weight and total soluble carbohydrates. Bulb filling took place 140 days after sowing date and continue until harvesting stage (Ledesma *et al.*, 1997).

B. Growth attributes:

Leaf growth attributes leaf area, leaf area ratio (LAR), leaf weight ratio (LWR) and specific leaf area (SLA):

Leaf area per plant:

It is highly evident from Fig. (1) that there was a significant increase in leaf area parameter during the different sampling dates in both seasons of 2009/2010 and 2010/2011. The total plant leaf area values reached 272.22 and 262.61 cm² at the first sampling date in both seasons, respectively, after that leaf area start to dramatically increase to reach 1249.68 and 1145.83 cm² at the fourth sampling dates in both seasons of study, respectively. The largest increase in leaf area was occurred between the third and fourth sampling dates, it almost increased two fold.

The obtained results may be due to that garlic plant produced new leaves until the third sampling date as previously shown in Table (4). Such new leaves continue to grow and expand. Also, Muro *et al.* (2000) indicated that garlic leaf number per plant and leaf area reached their maximum values three weeks before harvesting stage.

The obtained results are in agreement with the findings of Soliman (1999). In addition, Moravčević *et al.* (2011) stated that garlic plant develops 8 to 14 leaves and LAI is a very good indicator of a plant's photosynthetic capacity and it is also very important in determining the levels of photosynthetic activity and yield. In the same respect, Haque *et al.* (2002) suggested that there was a positive correlation between crop growth rate and the leaf area index.

Leaves are the main organ responsible for photosynthetic activities. Therefore, the amount of leaf dry matter increased as a result of increasing number of leaves per plant and total leaf area. Once garlic bulb initiated and started to develop after 60 days of plant growth, the photosynthetic efficiency of the leaves increased to meet their requirements as well as the growing bulbs (Rahman *et al.*, 2004).

Leaf area ratio (LAR), Leaf weight ratio (LWR) and Specific leaf area (SLA):

Data presented in Fig. (1) revealed that there were a considerable reduction in leaf area ratio, leaf weight ratio and specific leaf area parameters throughout different sampling dates during both seasons of study. Such reduction was gradually up to 120 days after sowing (the second sampling date), at that moment, the reduction start to be progressively until 150 and 180 days after sowing (the third and fourth sampling dates). The reduction recorded no significant differences between the first and second sampling dates. Also no significant differences were detected between the third and fourth sampling dates. While significant differences were noticed only between both of them. Similar results were obtained in both seasons of study.

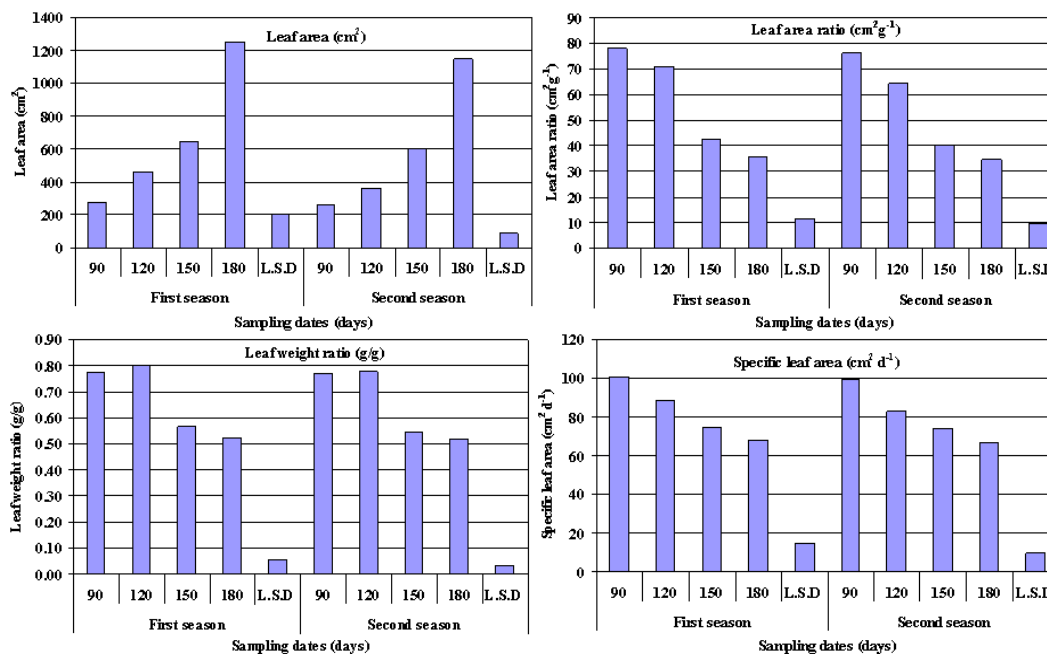


Fig. 1: Effect of different sampling dates on garlic leaf growth attributes (leaf area, leaf area ratio, leaf weight ratio and specific leaf area) during both seasons of 2009/2010 and 2010/2011.

The obtained results may be explained by defining the following terminology of leaf area ratio (LAR), it is the ratio, between total leaf area per plant and total dry weight per plant. Leaf weight ratio (LWR) it is the ratio between total leaf dry weight per plant and total dry weight per plant. Specific leaf area (SLA) it is the ratio between total leaf area per plant and total leaf dry weight per plant as demonstrated by Hunt (2003). All parameters used for calculating leaf growth attributes were gradually increased together over time. As a result the calculated values were decreased.

Muro *et al.* (2000) indicated that garlic leaf parameters (leaf number per plant, leaf area and leaf mass) reached their maximum values three weeks before harvesting stage. Leaf parameters are important factor for obtaining good results in crop production. While, Ledesma *et al.* (1997) observed that the maximum values for leaf area index (LAI) parameter was reached at the end of garlic shoot growth stage. Translocation of photosynthetic substrates to the bulb was begun afterwards.

In addition, Moravčević *et al.* (2011) stated that garlic plant develops 8 to 14 leaves and LAI is a very good indicator of a plant's photosynthetic capacity and it is also very important in determining the levels of photosynthetic activity and yield. In the same respect, Haque *et al.* (2002) suggested that there was a positive correlation between crop growth rate and leaf area index.

Absolute growth rate (AGR):

It is a simple index of plant growth, it is expressed as a rate of change in plant fresh or dry weight (mg) which is an increment in plant weight per a unit of time (day). Data generated for plant fresh weight were used to calculate absolute growth rate of garlic plant. It is calculated between every two sampling dates, 90-120, 120-150 and 150-180 days after sowing, respectively, as shown in Table (5). Absolute growth rate was significantly increased over the time to reach 660.44 and 602.22 mg/day by the third and fourth sampling date (150-180 days after sowing) in the first and second season, respectively.

At the initial stage of growth (90-120 days after sowing), absolute growth rate was slowly increased to the growth stage of 120-150 days after sowing. After that absolute growth rate was dramatically increased at the growth stage of 150-180 days after sowing. The same pattern was noticed in both seasons of study.

This is might be due to that, at the third sampling date garlic bulb begun to fill and acts as a sink. The maximum translocation of photosynthetic substrates from leaves to the bulb was evident. This led to a massive increase in plant fresh or dry weight as previously described in Table (4). In addition, climate conditions during this stage of growth were suitable for increasing the efficiency of photosynthetic and promote translocation of photosynthetic substrates from leaves to bulb.

These results are in agreement with the findings of Soliman (1999); Ledesma *et al.* (1997) and Kamenetsky *et al.* (2004).

Relative growth rate (RGR):

It is clear from Table (5) that there was a significant difference in relative growth rate during the different developmental stages in both seasons of 2009/2010 and 2010/2011. Relative growth rate was increased from 21.26 to 28.23 mg/g/day, in the first season and from 16.86 to 32.50 mg/g/day, in the second season for the first and second growth stages (90-120 and 120-150 days after sowing), respectively. While in the third growth stage (150-180 days after sowing), relative growth rate was decreased to reach 27.26 and 26.52 mg/g/day, in the first and second season, respectively.

This may be due to that the increment of plant fresh or dry weight in the first and second growth stages (90-120 and 120-150 days after sowing) was superior than the increment in the third growth stage (150-180 days after sowing). In addition, the lower value of relative growth rate recorded at the latest growth stage might be also related to the leaf senescence.

The obtained results are in concurrence with the findings of Soliman (1999) and Ledesma *et al.* (1997). They showed that the maximum values for relative growth rate (RGR) parameter was reached at the end of garlic shoot growth stage. Translocation of photosynthetic substrates to the bulb was begun afterwards. They also added that during bulb growth stage, there were an increase in dry weight. Bulb filling took place 140 days after sowing date and continue until harvesting stage.

Net assimilation rate (NAR):

Net assimilation rate, is the measure of gaining or accumulating in dry matter per unit leaf area per unit time and it is an important index of mean photosynthetic efficiency of a crop. Data in Table (5) clearly indicated that the net assimilation rate was significantly increased with increasing growth stage from 90-120 to 150-180 days after sowing. This was true in both seasons of study. The highest value of net assimilation rate was obtained during the third growth stage (150-180 days after sowing). Whereas, the lowest value was recorded by the first growth stage (90-120 days after sowing) in both seasons of 2009/2010 and 2010/2011.

The higher values of net assimilation rate at the latest growth stage, 150-180 days after sowing might be related to the rapidly developing of bulb and it is acts as sink as reported by Haloï and Baldev (1986) and Rahman *et al.* (2004).

The obtained results are in good agreement with the findings of Soliman (1999) and Ledesma *et al.* (1997) they observed that the maximum value for net assimilation rate (NAR) was reached at the end of garlic shoot growth stage. They also added that during bulb growth stage, there were an increase in bulb dry weight. Bulb filling took place 140 days after sowing date and continue until harvesting stage. Harvesting stage is characterized by a maximum bulb dry weight.

Table 5: Effect of different sampling dates on growth attributes (absolute growth rate, relative growth rate and net assimilation rate) of garlic plant during both seasons of 2009/2010 and 2010/2011.

Sampling dates (days)	Absolute growth rate (mg/day)	Relative growth rate (mg/g/day)	Net assimilation rate (mg/cm ² /day)
First season (2009/2010)			
90 - 120	102.22	21.26	0.30
120 - 150	289.78	28.23	0.53
150 - 180	660.44	27.76	0.73
L.S.D at 0.05	74.66	2.41	0.13
Second season (2010/2011)			
90 - 120	73.89	16.86	0.24
120 - 150	310.22	32.50	0.65
150 - 180	602.22	26.52	0.72
L.S.D at 0.05	63.19	5.18	0.11

C. Chemical constituents:

Garlic leaf pigment contents:

Data obtained on the changes of chlorophyll a, b, total chlorophyll and total carotenoids in garlic leaves during its developmental stages in seasons of 2009/2010 and 2010/2011 are presented in Table (6). Results indicated that there were significant increases in garlic leaf pigments (chlorophyll a, b and total) and it reached to maximum values at the second sampling date (120 days after sowing) and then tend to decrease at the third sampling date (150 days after sowing). This trend was similar in both seasons of study. This is may be due to the incidence of leaf senescence which accompanied with chlorophyll degradation.

Table 6: Effect of different sampling dates on garlic leaf pigments, bulb total carbohydrates and nutrients uptake in leaves and bulb of garlic plants during both seasons of 2009/2010 and 2010/2011.

Sampling dates (days)	Chlorophyll (mg/100g FW)			Total Carotenoids (mg/100g)	Total carbohydrates rates (mg/100g)	Nutrient uptake (mg/plant)					
	a	b	Total			Nitrogen		Phosphorus		Potassium	
						Leaves	Bulb	Leaves	Bulb	Leaves	Bulb
First season (2009/2010)											
90	21.27	18.64	39.91	10.43	10.04	7.93	5.20	1.92	0.35	5.02	1.15
120	28.44	25.13	53.57	15.51	18.46	20.95	26.61	3.81	0.49	20.68	1.70
150	21.61	22.69	44.30	26.54	32.77	28.30	35.42	7.37	4.64	28.63	9.27
L.S.D at 0.05	1.76	1.40	5.10	1.91	0.82	2.53	0.89	0.63	0.75	4.94	0.11
Second season (2010/2011)											
90	20.57	19.51	40.07	11.20	10.67	8.54	6.50	2.18	0.45	6.62	1.22
120	26.16	25.67	51.83	16.23	19.63	19.46	19.46	4.20	0.67	18.89	1.54
150	24.14	21.10	45.24	29.40	33.40	28.47	29.30	7.37	4.74	26.16	10.07
L.S.D at 0.05	1.65	1.81	3.13	1.10	0.47	3.64	0.73	0.51	0.24	6.18	1.51

Carotenoids content in garlic leaf were found to increase significantly and reached to the highest values (26.54 and 29.40 mg/100g) with the third sampling date (150 days after sowing) in the first and second seasons, respectively. Whereas, the lowest values were recorded by the first sampling date (90 days after sowing) in both seasons.

Carotenoids content were negatively correlated with chlorophyll a, b, and total (Sadegh *et al.*, 2011). There is a positive correlation between carotenoids and chlorophyll a and b synthesis. It seems that chlorophyll isn't formed until sugar reduction and carotenoids seem to appear or increased when chlorophyll begun to degrade (Ebrahinzadeh, 2000). In the same respect, Mashayekhi and Neumann (2006) reported a direct correlation between chlorophyll formation and secondary metabolites specially anthocyanins in carrot.

Total carbohydrates:

It is clear from Table (6) that total carbohydrates in bulb of garlic were widely and significantly affected by the progress of age towards maturation. Total carbohydrates increased in a linear form. The lowest values for total carbohydrates in bulb of garlic were recorded at 90 days after sowing. The highest values were obtained at 150 days after sowing. These results were similar and true in the two seasons of the experiment.

The obtained results are in harmony with Ledesma *et al.* (1997) who summarized that the maximum values for total soluble carbohydrates (TSC) was reached at the end of garlic shoot growth stage. Translocation of photosynthetic substrates to the bulb was begun afterwards. They also added that during bulb growth stage, there were an increase in total soluble carbohydrates. At harvesting stage garlic bulbs are characterized by a maximum total soluble carbohydrates and soluble proteins.

Nutrients uptake (N, P and K):

The results of the changes in the N, P and K uptake of garlic leaves and bulb during plant development in the two seasons of 2009/2010 and 2010/2011 are shown in Table (6). Data showed that there was a significant increase in N, P and K uptake in leaves and bulb of garlic plants from 90 to 150 days after sowing. In general, it could be confirmed that nutrient contents were significantly affected by the progress of age towards maturation.

The highest values of uptake N, P and K were observed at 150 days after sowing (the third sampling date) and the lowest values were recorded at the age of 90 days after sowing (the first sampling date). Uptake N, P and K in garlic plant leaves and bulb followed the same pattern in the two seasons of 2009/2010 and 2010/2011. These results are in agreement with the findings of Soliman (1999) on garlic. In addition, Argüello *et al.* (1997) confirmed that no significant changes in nutrient were observed during clove sprouting stage. Nutrient changes

become important between the initiation of shoot growth (30 days after sowing date) and at the beginning of the bulb filling stage (140 days after sowing date).

References

- Abou El-Magd, M.M., 1979. Effect of level of soil moisture and nitrogen fertilizer on the growth, nitrogen metabolism and yield of garlic. Ph.D. Thesis, Plant Production Dept., Fac. Agric., Zagazig Univ., Egypt.
- Ali, M., M. Thomson and M. Afzal, 2000. Garlic and onions: their effect on eicosanoid metabolism and its clinical relevance. *Prostaglandins Leukotrienes and Essential Fatty Acids*, 62: 55-73.
- Argüello, J.A., S.B. Núñez and A. Ledesma, 1997. Bulbing physiology in garlic (*Allium sativum* L.) cv. "Rosado Paraguay" III. Nutrient content in garlic plants: its relation to growth dynamics and bulb morphogenesis. *Acta Hort.*, 433: 417-426.
- Augusti, K.T. and P.T. Mathew, 1974. Lipid lowering effect of allicin (diallyl disulfide oxide) on long-term feeding in normal rats. *Experientia*, 30: 468-470.
- Black, C.A., D.D. Evans, L.E. Ensminger, G.L. White and F.E. Clark, 1981. *Methods of soil analysis. Part 2*, Agron. Inc. Madison, Wisconsin, USA.
- Block, E., 1985. The chemistry of garlic and onions. *Sci. Amer.*, 252: 114-119.
- Brand, D.G., G.F. Weetman and P. Rehster, 1987. Growth analysis of perennial plants. The relative production rate and its yield components. *Ann. Bot.*, 59: 45-53.
- Chapman, H.D. and P.F. Pratt, 1982. *Methods of Plant Analysis, I. Methods of Analysis for Soil, Plant and Water*. Chapman Publishers, Riverside, California, USA.
- Cottenie, A., M. Verloo, L. Kickens, G. Velghe and R. Camerlynck, 1982. *Chemical analysis of plants and soils. Laboratory of Analytical and Agrochemistry*. State University, Ghent, Belgium.
- Dalen, E.J., 1992. How Do Leaves Grow? *Bioscience*, 42: 423-432.
- Del Pozo, A. and M.I. Gonzalez, 2005. Developmental responses of garlic to temperature and photoperiod. *Agric. Tech.*, 65: 119-126.
- Devlin, R.M. and A.V. Barker, 1971. *Photosynthesis*. Van Nostrand Reinhold, Univ. of Minnesota, USA.
- Dubois, M., K.A. Giles, J.K. Hamilton, P.A. Rebers and F. Smith, 1965. Colorimetric method for determination of sugars and related substances. *Anal. Chem.*, 28: 350-356.
- Ebrahimzadeh, H., 2000. *Plant Physiology, Photosynthesis*, Tehran University Press, Iran.
- Etoh, T. and P.W. Simon, 2002. Diversity, fertility and seed production of garlic. In: Rabinowitch, H.D., Currah, L. (Eds.), *Allium Crop Science: Recent Advances*. CAB Int., Wallingford, pp: 101-117.
- Haq, S., A. Sattar and H.R. Pramanik, 2002. Dry matter accumulation and partitioning and growth of garlic as influenced by land configuration and cultivars. *Pak. J. Biol. Sci.*, 5: 1028-1031.
- Haloi, B. and B. Baldev, 1986. Effect of irrigation on growth attributes in chickpea when grown under different dates of sowing and population pressure. *Indian J. Plant Physiol.*, 29: 14-27.
- Hunt, R., 2003. Growth and development: Growth analysis, individual plants. *Encyclopedia of Applied Plant Sciences*, 579-588.
- Jabbes, N., I. Arnault, J. Auger, B.A. Dridi and C. Hannachi, 2012. Agro-morphological markers and organo-sulphur compounds to assess diversity in Tunisian garlic landraces. *Sci. Horti.*, 148: 47-54.
- Jackson, M.L., 1973. *Soil Analysis*. Constable Co. Ltd., London, UK.
- Kamenetsky, R., I.L. Shafir, H. Zemah, A. Barzilay and H.D. Rabinowitch, 2004. Environmental control of garlic growth and florogenesis. *J. Amer. Soc. Hort. Sci.*, 129: 143-146.
- Kulkarni, M.G., S.G. Sparg and J. Van Staden, 2005. Influence of temperature and watering frequencies on seed germination and seedling growth of *Omithogalum longibracteatum* and *Tulbaghia violacea*. *Sci. Horti.*, 107: 103-109.
- Ledesma, A., S.B. Núñez and J.A. Argüello, 1997. Bulbing physiology in garlic (*Allium sativum* L.) cv. "Rosado Paraguay" II. Characterization of ontogenic stages by shoot growth dynamics and its relation to bulbing. *Acta Hort.*, 433: 405-416.
- Lewis, L.A., D.L. Ojeda, M.O. Salazar and R.J. Campbell, 1995. Effect of population density on growth, development and yield of garlic (*Allium sativum* L.) cv. Vietnamita. *Proc. Interamerican Soc. Trop. Horticult.*, 39: 23-26.
- Mashayekhi, K. and K.H. Neumann, 2006. Effects of boron on somatic embryogenesis of *Daucus carota*. *Plant Cell Tissue and Organ Culture*, 84: 279-283.
- Moravčević, D., V. Bjelić, D. Savić, J.G. Varga, D. Beatović, S. Jelačić and V. Zarić, 2011. Effect of plant density on the characteristics of photosynthetic apparatus of garlic (*Allium sativum* var. *vulgare* L.). *African J. Biotech.*, 10: 15861-15868.
- Muro, J., I. Irigoyen, C. Lamsfus and M. Fernandez, 2000. Effect of defoliation on garlic yield. *Sci. Horti.*, 86: 161-167.

- Nyengedzeni, M., 2010. Yield and quality responses of Egyptian white garlic (*Allium sativum* L.) and wild garlic (*Tulbaghia violacea* harv.) to nitrogen nutrition. M.Sc. Thesis, Plant Production and Soil Science Dept., Faculty of Natural and Agricultural Sciences, Pretoria University, South Africa.
- Rahman, S., A. Islam, S. Haque and A. Karim, 2004. Effect of planting date and gibberellic acid on the growth and yield of garlic (*Allium sativum* L.). *Asian J. Plant Sci.*, 3: 344-352.
- Randle, W.M. and J.E. Lancaster, 2002. Sulphur compounds in alliums in relation to flavour quality. In: Rabinowitch, H.D., Currah, L. (Eds.), *Allium Crop Science: Recent Advances*. CAB Int., Wallingford, pp: 329-356.
- Sadegh, A., A. Vahid, M. Kambiz and J.M. Seyyed, 2011. Garlic physiological characteristics from harvest to sprouting in response to low temperature. *J. Stored Products Postharvest Res.*, 2(15): 285-291.
- Snedecor, G.W. and W.G. Cochran, 1980. *Statistical Methods*. 12th ed., Iowa State Univ. Press, Ames, Iowa, USA.
- Soliman, M.F.Z., 1999. Studies on intercropping some vegetable crops on garlic plants. M.Sc. Thesis, Vegetable Dept., Fac. Agric., Cairo Univ., Egypt.
- Takagi, H., 1990. Garlic (*Allium sativum* L.). In *Onions and Allied Crops*, Brewster J.L. and H.D. Rabinowitch (Eds), CRC Press Inc., Boca Raton, Florida, USA.
- Tapiero, H., D.M. Townsend and K.D. Tew, 2004. Organosulfur compounds from alliaceae in the prevention of human pathologies. *Biomedicine and Pharmacotherapy*, 58: 183-193.
- Watson, D.J., 1958. The dependence of net assimilation rate on leaf area index. *Ann. Bot.*, 22: 37-54.
- Wolf, B., 1982. A comprehensive system of leaf analysis and its use for diagnosing crop nutrients status. *Comm. Soil Sci. Plant Anal.*, 13: 1035-1059.
- Yamaguchi, M., 1983. *World Vegetables: Principles, Production and Nutritive Values*. AVI, Westport, CT.