REPUBLIC OF TURKEY
ERCIYES UNIVERSITY
GRADUATE SCHOOL OF NATURAL AND APPLIED SCIENCES
DEPARTMENT OF AGRICULTURAL SCIENCES AND TECHNOLOGIES

GENOTYPIC DIFFERENCES IN NITROGEN EFFICIENCY AND ROOTSTOCK POTENTIAL OF SOME LOCAL TOMATO VARIETIES OF GHANA AND TURKEY

Prepared By
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Prof. Dr. Halit YETİŞİR

MSc. Thesis

August 2018
KAYSERI

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KAYSERI

COMPLIANCE WITH SCIENTIFIC RESEARCH ETHICS

I, **Mohammed Bello ADAM**, hereby declare that this project work is the results of my research and for that matter has never being submitted any were for any degree.

Mohammed Bello ADAM

.....

COMPLIANCE WITH GUIDE

The write-up for this master's thesis topic **"Genotypic Differences in Nitrogen Efficiency and Rootstock Potential of Some Local Tomato Varieties of Ghana and Turkey"** was prepared base on the directives of Erciyes University's Graduate Thesis write-up guide.

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ACCEPTANCE AND APPROVAL

With keen supervision of **Asst. Prof. Dr. Abdullah ULAŞ** and **Prof. Dr. Halit YETİŞİR,** the thesis write-up with regards to the topic **"Genotypic Differences in Nitrogen Efficiency and Rootstock Potential of Some Local Tomato Varieties of Ghana and Turkey"** was prepared by **Mohammed Bello ADAM** was accepted master's degree thesis by the departments Agriculture Science and Technology, Graduate School of Natural and Applied Sciences, Erciyes University.

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Erciyes University, Graduate School of Natural and Applied Sciences
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ABSTRACT

As a result of rising practices of applying overrate nitrogen (N) fertilizers by growers with intention of increasing yield has cause negative effect on plants ecosystems, animals and bacteria in our environment. On the other side of the coin, the effect of inefficient accessibility of fertilizer causes reduction of soil fertility which is mostly attributed to loss of organic matter. Furthermore, limit to access of N fertilizer cause hikes in prices, which lead reduction in agriculture production. However, plant breeders are finding alternative approach to improve uptake and utilization of N in plants. The aim of this study was to initially determine the genotypic differences in N efficiency of some local and hybrid tomato varieties and further identify rootstock potential of some selected N-efficient tomato varieties via reciprocal grafting. Two nutrient solution experiments were conducted at plant physiology laboratory of Erciyes University Agriculture Faculty. The first experiments were dated 12th September 2017 whilst second experiments were dated 2nd February 2018. Both experiments were conducted in a controlled growth chamber in 6 weeks with different N rates (low-N: 0.3 mM and high-N 3.0 mM) which was supplied in complete randomized block design with three replication. In the experiments, 4 hybrids and 10 local Turkish varieties and 4 local Ghanaian tomato cultivars were used. There were significant genotypic differences in N efficiency among tomato varieties in the Experiment I and Experiment II. Results of Experiment I confirmed that *HelenaF1* and *ALT* can be characterized as 'N-efficient' while *P005* and *Karahidir* as 'N-inefficient' tomato varieties. The results of Experiment II confirmed that grafted plants are 'N-efficient' than non-grafted plants. The graft combinations of *Karahidir/ALT* (S1/R2) and *Karahidir*/*Helena* (S1/R1) showed best performance in N-efficiency as compared to other combinations. All these clearly indicated that N-efficient rootstocks could be used to improve the N efficiency of tomato plants by grafting.

Keywords: N uptake, N utilization, tomato, biomass, grafting, vigorous rootstock.

BAZI TÜRK VE GANA YEREL DOMATES ÇEŞITLERI ARASINDAKI GENOTIPIK AZOT ETKINLIK FAKLILIKLARIN VE ANAÇLIK POTANSIYELLERIN BELIRLENMESI

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ÖZET

Yetiştiricilerin daha fazla ürün elde etmek amaçıyla aşırı dozda azotlu (N) gübre uygulamaları, çevremizde bulunan bitki ekosistemini, hayvanları ve bakterileri olumsuz yönde etkilemektedir. Madalyonun diğer yüzüne bakıldığında, gübre alamamalarından dolayı yetersiz gübre uygulamaları neticesinde de organik madde kaybına bağlı olarak toprak verimliliği azalmaktadır. Ayrıca yüksek fiyatlardan dolayı azotlu gübre kullanımın azalması tarımsal üretimde düşüşlere yol açmaktadır. Fakat bitki ıslahçıları bitkideki azot alım ve kullanım miktarlarını arttırmaya yönelik alternatif yaklaşımlar ve çalışmalar yürütmektedirler. Bu çalışmanın amacı; bazı yerel ve hibrit domates çeşitleri arasındaki azot etkinlik bakımından genotipik farklılıkları belirlemek ve daha sonra azot etkin olarak belirlenmiş olan bazı domates çeşitlerinireciprocal aşılayarak onların anaçlık potansiyellerinin belirlenmesidir. Bu çalışmada Erciyes Üniversitesi Ziraat Fakültesi'ne ait Bitki Fizyolojisi Laboratuvarında kontrollü iklim koşulları altında iki adet su kültürü besin çözeltisi denemesi kurulmuştur. Birinci deneme 12 Eylül 2017 de, ikinci deneme ise 2 Şubat 2018 tarihinde kurulmuştur. Her iki deneme, kontrollü iklim odalarında tesadüfi deneme parselleri deneme desenine göre 3 tekerrürlü olarak düzenlemiş olup, 2 farklı azot dozunda (düşük-N: 0.3 mM and yüksek-N 3.0 mM) yürütülmüştür. Bu denemede 6 adet Türk ticari hibrit ve 8 adet Türk yerel domates çeşidi ve 4 adet Gana yerel domates çeşidi kullanılmıştır. Azot etkinliği bakımından tomates çeşitleri arasında istatistiki olarak genotipik farklılıklar I. ve II. denemede tespit edilmiştir. I. Deneme sonuçlarına göre *HelenaF1*ve *ALT* 'N-etkin' çeşitler iken, *P005*ve *Karahıdır* 'N-etkinsiz'çeşitler olarak belirlenmiştir. II. Deneme sonuçlarına göre ise azot etkinliği en yüksek olan aşı kombinasyonu *Karahidir/ALT*(S1/R2) ve *Karahidir/Helena* (S1/R1) olarak tespit edilmiştir. Tüm bulgulara göre, azot-etkinsiz bir tomates çeşidinin azot-etkin olan güçlü bir anaç üzerine aşılanması sonucunda domates bitkisinin azot etkinliğinin artığı tespit edilmiştir.

Anahtar Kelimeler: N alımı, N kullanımı, domates, biyokütle, aşılama, güçlü anaç.

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LIST OF ACRONYMS

°C: degrees Celsius

$ZnSO_4$: Zinc sulphate

EC: Electrical conductivity

$(NH_4)_2SO_4$: Ammonium sulphate

R-CH-(NH2)-COOH: General formula of an amino acid

$Ca(NO_3)_2$: Calcium nitrate

kLUX: Kilo lux

$CaCO_3$: Calcium carbonate

NR: Nitrate reductase

$CaSO_4$: Calcium sulphate

NUE: Nitrogen use efficiency

CO_2: Carbon dioxide

SPAD: single-photon avalanche diodes

O_2: Oxygen (molecule)

UV: Ultra violet

N_2: Nitrogen (molecule)

MoFA: Ministry of Food and Agriculture Ghana

NH_4^+: Ammonium

S 1: Scion 1

NO_2: Nitrite

S 2: Scion 2

NO_3^- : Nitrate

R 1: Rootstock 1

$CuSO_4$: Cupper sulphate

R 2: Rootstock 2

H_2O: Water

SSA: Sub Saharan Africa (SSA)

H_2SO_4: Sulphuric acid

FAO: Food and agriculture organization

H_3BO_3: Boric acid

FAOSTAT: Food and agriculture organization statistical databases

HCl: Hydrochloric acid

cm: centimeters

K_2SO_4: Potassium sulphate

mm: millimeters

KH_2PO_4: Monopotassium phosphate

mg: milligram

KNO_3: Potassium nitrate

USDA: United States Department of Agriculture

$MgSO_4$: Magnesium sulphate

NPK: Nitrogen, Phosphorous, and Potassium

$MoNa_2O_4$: Sodium molybdate

Fe-EDDHA: Ethylene diamine-N, N bis (2-hydroxyphenylacetic acid)

NaCl: Sodium chloride

NaOH: Sodium hydroxide

NEED: N-(1-naphthyl)-ethylenediamine hydrochloride

T_0: Time 0

T_{60}: Time 60

WL: Wavelength

Total-chlo: Total chlorophyll

SAS: Statistical Analysis Software

TC: Total carotenoids

ICP-AES: Inductively Coupled Plasma

Atomic Emission Spectroscopy

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CHAPTER ONE

INTRODUCTION

The stability and growth of every country in the world rely mainly on economic and social factors. One attributes to these factors is agricultural production (DeGraft *et al.*, 2014). As the population of humans on earth keeps on rising day by day, there is the need for farming activities to be intensified in order to provide more food for consumers (Christiaensen *et al.*, 2011). Nevertheless in sub-Saharan Africa food insecurity is a problem, which causes hunger and poverty and this is to the fact that there is less agriculture production which mostly comes from rural areas. People from these areas engage in agriculture for leaving but do not have financial power to increase agriculture production (Barrett *et al.*, 2006). For a crop to have appropriate nutrient for growth, the nutrients in the soil should be sufficient and accurately balanced. However, nutrient depletion in the soil occurs due to erosion, leaching, removal of harvest, volatilization which causes loss of nitrogen due to burning at high temperature and fixation which also occurs in phosphorous. Soil fertility can be improved by application of solid matter deposits, decomposition of organic matter, nitrogen fixation, weathering and chemical fertilizer (Van Schöll *et al.*, 1998). Among all these modes of fertilization, chemical fertilizers are more convenient to growers due to it merits such as being readily available to plants and containing high mineral nutrients which are necessary for plant growth and yield production. Moreover, chemical fertilizers have usually immediate positive effects on the plant growth, occupying fewer places for storage, easily transportable and applicable to the field and therefore much preferred than organic fertilizers. However they have some demerits when overrate application is made. Some of these are destruction of microorganisms, pollution of water bodies, leaching, acidification or alkalization of the soil and degrading of soil structure due to decomposition of organic matter in the soil (Chen, 2006). Some of the chemical

fertilizer commonly used come in the form of urea, potassium sulphate, potassium chloride, potassium magnesium sulphate, natural rock phosphate, tri super phosphate, super phosphate, mono ammonia phosphate, di-ammonia phosphate, calcium ammonia nitrate and ammonia sulphate (Van Schöll *et al.*,1998). Among all these types of chemical fertilizers, nitrogen (N) fertilizers has being widely used and accepted by growers for the past four decades due to its contribution to doubling of global food production. As a result of rising practices of applying overrate nitrogen fertilizers by growers with the intention of increasing yield has caused negative effect on plants ecosystems, animals and bacteria in our environment. To give an illustrate situation is damage of water bodies by eutrophication (Giles, 2005) and also overrate of nitrogen fertilizers causes leaching of arable fields (Tilmam, 1999). On the other side of the coin, the effect of inefficient accessibility of fertilizer causes reduction of soil fertility which is mostly attributed to loss of organic matter. Scarcity of fertilizer mostly affects the Sub-Saharan Africa (SSA). The farmers from this part of Africa are small-scale farmers so scarcity of fertilizers causes hikes in fertilizers price which they cannot afford and surely cause reduction in agriculture production, poverty and economic instability (Greenland, 1998; Syers 1997). In addition, the impact of nitrogen fertilizer on crop production is not efficient since crops can absorb less than 50% of applied nitrogen from the soil for utilization (Mosier *et al.,* 2004). The rest of the applied nitrogen fertilizer on the arable land run into water bodies, which is harmful to users and escape of nitrous gas into the atmosphere also cause global warming. Because of this situation at hand, plant breeders are targeting to come up with crop genotypes which utilize nitrogen very efficiently (Hirel *et al.*, 2007). In this critical situation, utilizing nitrogen efficiently is very vital to reduce losses concerning nitrogen fertilizer application (Good *et al.*, 2004). According to Moll *et al.* (1982) nitrogen use efficiency can be explained as yield made out of grain according to unit of nitrogen available in the soil. Nitrogen use efficiency can be further categorized into two stages, which are; uptake efficiency and utilization efficiency. Uptake efficiency which is explained as the mechanism in which plant is able to access nitrate and ammonium ions from the soil. Also utilization efficiency is explained as the mechanism of producing yield out of grain by utilizing nitrogen (Zhao-Liang, 2000; Raun and Johnson, 1999).With regard to significant of nitrogen use efficiency, plant breeders are targeting alternative physiological procedures for nitrogen extraction from the surface of the soil for translocation, assimilation and

redistribution in plants (Foulkes, *et al.,* 2009; Masclaux-Daubresse *et al.,* 2010). Selecting and planting genotypes which could result into good yield with respect to low nitrogen consideration is paramount for effective result (Barraclough *et al.,* 2010).

Tomato (*Lycopersicum esculentum* L.,) is being categorized as a nightshade crop from the *solanaceae* family. In the world, tomato is a well-known vegetable eaten most by people from different lifestyles (Grandillo *et al.,* 1999). It is mostly eaten in diverse way that is notable in our foods, drinks, and sauces and eaten in a raw state (Alam *et al.,* 2007). Tomato is of great important to the health of humans because it is embedded with a lot of nutrient (Arah *et al.,* 2015). Tomato is also full of dietary fibers, sugar, essential amino acid, vitamins and minerals. In addition, tomato contains phosphorous, iron, vitamin B and C. Moreover, cultivation of tomato is now a business hub across the world and it is also used mostly in industries (Ayandiji *et al.,* 2011). According to (Aksoy and Kaymak, 2016) the production of tomato stands at 162 million tons using an estimated field of 4.8 million hectares in the world. The country with highest yield of tomato is China with about 50 million tons of tomato followed by India producing also about 17.5 million tons. However in Turkey the total amount of tomato produced annually stands at 11-12 million tons. Most of the tomato production in Turkey comes from Manisa, Mersin, Izmir, Bursa and Antalya (Aksoy and Kaymak, 2016). In dish of many Ghanaian, tomato is a key ingredient. It is mostly used in cooking local delicacies and delicious dishes like stews and soups (Ellis *et al*., 1998). However tomato production in Ghana is not encouraging because it is in record that 510.000 metric tons of tomato is produced annually with importation of 27.000 metric tons of processed tomato from Europe (Robinson and Kolavalli, 2010). In addition, cultivation of tomato is on the rise by Ghanaian farmers but this has not reached the high request of consumers which therefore compels importation of tomato from Burkina-Faso. This issue of low agriculture productivity in Ghana and other African countries is due to decline in soil organic matter and soil fertility, which is caused by lack of good farming practices (Stoorvogel, *et al.,* 1993). Lost soil fertility can not only be replaced by compost from animal and plant residue. In these situation mineral fertilizers is vital to increase soil fertility that is at the end result increasing agriculture production. Also another problem of African farmers is the lack of enough funds to purchase these mineral fertilizers as most of these farmers are poor and also they face challenges with

accessibility of the mineral fertilizer for utilization (Sanchez *et al.,* 1997). In order to increase agriculture productivity and retain soil fertility, it is important to explore nutrient and crop management system in order to maximize nutrients from soil reserves, organic manures, fertilizers and biological nitrogen fixation (Palm, 1997).

Currently across the world, the act of grafting is becoming well known with some crops in *Solanaceae* and *Cucurbitaceae* family (Lee, 2010). The origin of grafting initially started in Japan and Korea in 1920 where watermelon plant was used as scion which was grafted on squash plant which was used as rootstock, the intention of this was to overcome problem of soil pathogens mainly *Fusarium oxysporum* (Yamakawa, 1983). After which this horticultural technology widely spread in Japan, Korea, Europe and Asia in intensive cultivation of crops. Use of grafting gives a lot of merits to growers because; (1) causes reduction in bacterial and fungal contamination, (2) able to regulate wilting in plant (3), (4) enhance better development and growth of plant, (5) prevent salinity and drought stress and (6) improve absorption of mineral and nutrient from the rootstock to the shoot (Revero, 2003). Researcher has found out that better water and nutrient uptake which contributes to yield increment is due to genotypes which have vigorous root system. Grafting is found to contribute to better absorption of some macronutrient like nitrogen and phosphorous (Ruiz and Remero, 1999; Leonardo and Giuffrida, 2006).

Several factors highlight the significance of this research. The most prominent of which are increasing N fertilizer prices, high rates of rural poverty, increasing rules limiting fertilizer usage, in attempts to minimize environmental pollution, and the continues cultivation of marginal land due to population growth and pollution. A main approach to solve these problems is breeding and cultivation of new tomato cultivars or grafting with appropriate tomato rootstocks, which efficiently use the available nitrogen. Unfortunately, no information is available on the mechanisms of the growth effects of grafting on the morphological and physiological plant traits related to nitrogen efficiency in tomato crop.

Therefore the aims and objectives of this research were; (1) to determine genotypic differences in N efficiency and the importance of N uptake and N utilization efficiency for genetic variation among various tomato cultivars (2) to determine rootstock potential of some selected N-efficient tomato varieties via grafting, (3) to identify morphological

and physiological growth effects of grafting and their interactions between N-efficient tomato rootstocks and N-inefficient scion varieties under two contrasting N levels.

CHAPTER TWO

LITERATURE REVIEW

2.1 Origin and Domestication of Tomato

Tomato is from the family *Solanaceae* and it is in the same family with eggplant, potato, pepper and Tobacco (Van Dam *et al.,* 2005). Origination of tomato (*Lycopersicum esculentum* L.,) began in regions of Andean and areas of Peru, Ecuador, Chile, Bolivia and Columbia (Peralta and Spooner, 2006). Domestication of Tomato began somewhere in the 16th century in Mexico (Paran and Van der Knaap, 2007). Spanish traders during the 16th century migrated tomato from Mexico to Europe and after which it then spread to the African continent, Middle East, Southern and eastern Asia. Tomato is a perennial crop, which is mostly grown as annually and it has a consumable fruit which is red in nature but has short shelf life (Brown *et al.,* 2005). With regards to ranking of vegetable mostly grown in the world, Tomato is the second on the list (Antunes, 2017).The total land size for cultivation of tomato in the world is about 4.8 million hectares with production of tomato estimated at 162 million tons (Aksoy and Kaymak, 2016). The country with the highest production of tomato in the world is China followed by India, United States of America and Turkey respectively (Ordookani and Zare, 2011). The annual increase in consumption of tomato is at 4.5%, this figure was observed from the year 2004 (Aherne *et al.,* 2009). Also, in Ghana tomato is one of the most consumed vegetable (Aher *et al.,* 2009). Production of tomato in Ghana also provides job opportunity for Ghanaians within the Bolga, Ada and Ofinso north districts (Norman, 1992).

2.2 Botany of Tomato

The height of tomato plant meeting the normal growth requirement range from two-meter-tall or higher than that and tomato is cultivated all year round (perennial plant). Tomato plant comes in three different types which are; indeterminate type (tall), determinate type (bush) and semi-indeterminate type (semi-bush). The stem of tomato is mostly glandular, hairy, coarse and solid. The stem can be categorized into two growth systems, which are prostrate and erect. Also the length of growth of root beneath the soil is 50 cm or above this figure. It has tap root system, which is vigorous and also lateral and adventitious root is produced from the main root. On the adaxial surface of leaf is full of glandular hairs and the arrangement of the leaves are spirally in nature. Tomato leaf is 10-30 cm wide and 15-50 cm long and the shape of leaf is ovate to oblong. It has clustered flowering structure and produce about 6-12 flowers. The length of it petiole is about 3-6 cm. Its flower is bisexual in nature with a diameter of about 1.5 - 2.0 cm. The two flowers are opposite in arrangement. The petal when fully mature reflects a yellow colour and per flower there are 6 petals with a length of 1.0 cm. The anther of tomato plant which is yellow in colour is comprises of 6 stamens. The ovary has 2-9 partitions. Pollination of tomato plant is more of self-pollination than cross-pollination. With regards to pollination of tomato plant, bumblebees and bees are very vital (Van Dam *et al.,* 2005). At the immature states of tomato, fruit display a green colour and also hairy in nature but when fully matured it display red, orange or yellow colour. The shape of tomato fruit is globular to oblate in nature and it is also describe as flesh berry (Relf, McDaniel and Morse, 2009). Seeds of tomato are numerous with pear or kidney shape. Tomato seeds are brown in nature with a width of 2-4 mm and length of 3-5 mm. Within the endosperm lies the embryo, which is coiled up. For every 1000 seeds of tomato it weight are estimated to be 2.5-3.5 grams (Van Dam *et al.,* 2005).

Classification and Categorization of Tomato Fruit

Determination of fruit quality and standards of tomato is paramount in classifying fruit for utilization. They are classified based on their size and they are categorized as; the giant type which are (>100 mm), the large type (100 mm≤ to ≥ 80 mm), medium type (85 mm≤ to 65 ≥ mm) and small type (65 mm ≤ to ≥ 50 mm) (Luengo *et al.,* 1999). Classification of tomato is also based on the shape of the fruit, some of which are quick cultivar with flat and ripped fruit, late cultivar which has large fruit, cultivar that has oblong fruit shape and lastly cultivars of cherry tomato (Van Dam *et al.,* 2005). Also According to Norman (1992) tomato can be categorized into different varieties based on features such as determinate and indeterminate, colour of the fruit, shape, disease resistance and time of maturity. In different regions of Ghana there are different tomato varieties grown by farmers according to suitability of the variety to different conditions. The commonly grown tomato varieties in Ghana includes; Wosowoso, Laurona 70, Rio Grande, Tropimech, Pectomech VF Cac J and Roma VF as stated by Ministry of Food and Agriculture of Ghana (Godia, 2014). Among all these varieties the most ideal variety that consumers like is the Pectomech, possessing better market value than the others (Robinson and Kolavalli, 2010). Also in Turkey some marketable tomato cultivars commonly grown include Beril, Yeni Talya, Swanson, Beaufort and Arnold (Oda, 1995). Most Tomato production comes from five cities in Turkey, which are; Manisa, Izmir, Bursa, Mersin and Antalya (Aksoy and Kaymak, 2016).

2.3 Uses and Nutritional Value of Tomato

Consumption of tomato is very vital to the human body as it provides a lot of nutrients needed for growth and healthy development (Arah *et al.,* 2015).Tomato is consumed for different ways such as recipe for salads, use in drinks and also use for cooking in different kind of foods (Alam *et al.,* 2007). Tomato egg sandwich, grilled chicken-tomato, tomato stew, tomato-watermelon sorbet and tomato chili soup are some type of foods where tomato is a key ingredient (Arah *et al.,* 2015). Tomato is rich in nutrients such as phosphorous, minerals, essential amino acids, iron, fiber, vitamins and sugar (Ayandiji *et al.,* 2011). Tomato also has form of carotenoid called lycopene, which is abundant in tomato and it has anti-oxidant properties responsible for fighting against cardiovascular malfunctions and cancer. Vitamin C and A, which are found in tomato

are responsible for better eyesight. Its fiber content helps in good digestion and weight reduction in human body (Arah*et al.,* 2015). About 123 milligrams of nutrient can be obtained from matured tomato (Table 2.1).

Table 2.1. Nutrient Content of Tomato

Nutrient	Amount
Calcium	1.2mg
Carbohydrate	4.7g
Copper	0.073mg
Dietary fiber	1.5g
Fat	0.2g
Iron	0.33mg
Magnesium	1.4mg
Niacin	0.731mg
Pantothenic acid	0.109mg
Phosphorus	3mg
Potassium	292mg
Protein	1.0g
Thiamin	0.046g
Total sugars	3.23g
Vitamin C	16.9mg

Source: The USDA National Nutrient Database (2010)

2.4 Economic Importance of Tomato

Among all vegetable crops in world, tomato (*Lycopersicum esculentum* L.,) is one of the most preferred vegetables by consumers and it is eaten by many people around the globe (Grandillo, 1999). After potato, tomato is the crop that provides revenue to growers and it is mostly cultivated globally and also it is key vegetable crop in many agricultural industries (Godia *et al.,* 2014) As reported by USDA-NASS (2010) in many countries including United State of America, production of tomato contributes significantly to agricultural economy. As in 2010 in Virginia 51 million dollars was made from fresh market tomato. To peasant farmers in Ghana cultivation of tomato is a source of income for them. Most cultivation of tomato and other crops are carried within savanna forest and transitional zones of Ghana (Norman, 1992). Also in Turkey process tomato such as tomato paste create many job opportunities for its inhabitants and the country is known to earn a lot of money from foreign exchange (Tezel, 2003).

2.5 Growth Requirement of Tomato

Tomato (*Lycopersicum esculentum* L.,) is known to be a warm season crop. For maximum yield and quality tomato fruit, approximately dry and cool weather is needed. Tropical and temperate to hot conditions are suitable for the growth of tomato plant (Van Dam *et al.,* 2005). For ideal growth of tomato, temperature between 21 and 24 °C is required. Tomato is sensitive to temperature below 10 °C and above 38 °C, this may be harmful to the plant tissue (Rice *et al.,* 1990). The morphological appearance and yield of the crop is influenced by light intensity. Excessive drought cause splitting of fruit and also shedding of flowers and buds. Nevertheless, excess humidity causes mold infection and fruit rot (Van Dam *et al.,* 2005). Cultivation of tomato can be done on different types of soil but for best of growth, plant needs deep well drained, fertile, loamy or sandy loamy soil (Kelley *et al.,* 2010). Tomato plant needs soil with sufficient amount of nutrient and pH ranging from 5.5-6.8 for growth. For optimum growth of plant, addition of organic matter is needed but excess of organic matter causes increase water holding capacity and result in deficiency of nutrient in the soil (Van Dam *et al.,* 2005). Tomato is a heavy feeder crop that responds positively to major macronutrient, specifically nitrogen, phosphorous, and potassium (NPK) (Detweiler *et al.,* 2014). Among all the macro nutrient needed for growth of tomato plant, nitrogen is relatively required in high quantity with 140-280 kg per hectare followed by phosphorous needed in a quantity of 20-60 kg per hectare and lastly potassium is also needed in quantity of 180 kg per hectare. However overrate of nitrogen in the soil leads to increments of vegetative growth at the expense of fruit setting, decrease in fruit quality and maturity (Kelley *et al.,* 2010).With regards to irrigation of tomato plant 20 mm of water is needed every week when the weather is cool however hot and dry weather calls for more irrigation of 70 mm of water every week (Van Dam *et al.,* 2005).

2.6 Challenges of Tomato Production

One of the most preferred and eaten vegetable around the globe is tomato, which is eaten in different forms, this could be in a raw state or processed form (Heuvelink, 2005). However, in this twenty first century the request for consumption of tomato has increased in the world (Asgedom *et al.,* 2011). This has led to importation of tomato by countries such as United States of America (21%), Russia Federation (11.7%), Germany (10.1%), France (7.6%), United Kingdom (5.7%), Pakistan (3.6%), Netherlands (3.4%),

Canada (2.7%) and United Arab Emirates (2.2%) as of 2013 (Aksoy & Kaymak, 2016). Tomato is known to be source of income to many peasant farmers in Africa but of recent time tomato production in Africa is confronted with many constrains. In Nigeria for instant, most farmers depend on rainfall for production of tomato and this problem is based on the fact that these farmers do not have the financial capability to purchase irrigation system so there for limiting tomato production to rainy seasons (Adenuga *et al.*, 2013). In Ghana the story is similar in terms of limited production of tomato and this result from hikes in cost of hiring worker right from land preparation, transplanting and harvest which surmount to half of entire budget of production (Clottey *et al.*, 2009). Other challenges facing tomato sector in African countries including Ghana are inadequate and low standard production leading to importation of tomato from neighboring countries and issues of diseases and pest (Robinson and Kolavalli, 2010). In addition, other challenges include hikes in price of conveying the produce from production unit to the market centers, increase in price of inputs such as inorganic fertilizer which is always expensive to the peasant farmers and lastly lack of access to processing and storage amenities (Robinson and Kolavalli, 2010). Until these challenges are tackled with all serious tomato production in Africa will still be hampering.

2.7 History of Grafting

The request for food by humans has led to domestication and farming causing relocation in settlement and modernization. For many years, man has being struggling to find new ways to increase yield of crop cultivation (Masterson, 2013). A fast and alternative way to this problem is grafting. Grafting is a method which was used many years ago in woody plant to increase its production by joining two woody plants which are similar in species by means of vascular system. Nevertheless, historically the origin of grafting has not been identified. As a result of grafting two plants of similar species, there has being an increase in volume, yield and improvement in quality of woody plants (Masterson, 2013). As in 1500 BC grafting of fruit trees was common by the Chinese horticulturists and this is believed to be where grafting started so therefore Chinese horticulturists are acknowledged for that (Hartman, *et al.*, 2002). Aristotle's (384-322 BC) and his colleague Theophrastus (371-287 BC) wrote on compatibility in grafting which was a prevailing problem for those who were currently in the horticulture sector

(Masterson, 2013). The act of wood grafting was usually done among the Roman Empire. The act of grafting used in tree fruit prevail in dark ages with the territories of botanical garden of Islamic faith, Christian convents and European continent (Mudge *et al.*,2009).Gradually grafting of trees become well known across the globe (Ofori, 2015). In the vegetable sector, the use of grafting started in Japan and Korea during the late 1920s where watermelon plants were used as scion and was grafted on the rootstock of squash plant. The technique of vegetable grafting was used in melons to minimize the occurrence of pathogens (mainly *Fusarium oxysporum)* present in the soil (Lee, 1994). After which grafting was mostly practiced in areas where land was constantly under use and also in areas where cultivation of agriculture was scarce (Hartman and Kester, 2002). Currently across the globe grafting of vegetable is an act practiced in many crops. As of 2008 in Korea and Japan most cultivation of vegetables was produced by grafting with a 700 million seedlings per country (Lee *et al.,* 2010). Grafting has rapidly grown thought-out Europe. Spain is ranked the first in grafted transplant production (129.8 million annually), followed by Italy (47.1 million annually) and France (28 million annually) respectively (Lee *et al.,* 2010; Morra and Bilotto, 2009).Vegetable grafting gives a lot of merit to commercial crops such as tomato (*Lycopersicum esculentum* L.,), grafting helps the growers to make money when used effectively in production of tomato (Masterson, 2013).

2.8 Significant of Grafting

Environmental stress is one major factor hindering the growth and yield of crops. Nutrients, water, presence of oxygen, temperature, light, pathogen, metal ion concentration are basic components that lead to environmental stress. Planting of crops that are tolerant or resistant to environment stress is a way out to deal with this problem (Sánchez *et al.,* 2010). Many breeding programs and methods have being used to deal with this issue of environmental stress in crops but it turn to be unproductive and slow. Grafting cultivar which is thoroughly scrutinized onto a strong and promising rootstock is the best and alternative way to deal with environmental stress (Lee and Oda, 2010). The use of grafting *in Cucurbitaceae and Solanaceae* plants is on the rise in this modern era (Lee *et al.,* 2010). Inculcating the practice of grafting into Ghana's fruit vegetable sector will be vital to yield increments since it has many advantages (Ofori, 2015).

Disease Tolerant and Resistance

Grafting is known to combat notable diseases such as bacteria, nematodes, viruses and fungi in plants. Also further research has proven that grafting can be used to control soil borne disease and also limit over application of pesticide with the aim of minimizing diseases (Bletsos, 2005, Pavlou *et al*., 2002; Giannakou and Karpouzas 2003; Besri, 2001). Utilization of grafting in tomato production has aid in minimizing corky rot triggered by *Pyrenochaeta lycopersici,* influencing proper root development and good nutrient uptake (Bradley, 1968). Mostly in agribusiness for cultivation of watermelon, tomato and melons under open field or green house condition, grafting is used to prevent infection of root knot nematodes and pathogens found in the soil. Grafting has proven to be important in eliminating bacteria wilt with regard to *solanaceous* crops such as eggplant and tomato in Asia (Oda, 1999).

Increase in Crop Production

To many growers the technique of grafting has help in yield increment leading to high profit margin. On commercial scale talking about yield of some horticultural crop like tomato, enlargement in size of fruit has influence on fruit yield (Pogonyi *et al.,* 2005). Also grafting in tomato is known to cause better yield increments and harvest which is as a result of vigorous root system it possess (Upstone, 1968). Also as stated by Yetisir and Sari (2007) grafting contribute to yield increment in watermelon. To explain further there has being significant yield increments for grafting eggplants with wild solanum rootstock as matched with non-grafted plant (Ibrahim *et al*., 2001; Rahman *et al*., 2002). Research carried by Lee and Oda (2010) shows that most grafted plants show a better absorption of nutrients and water and this is to the fact that rootstock used for grafting is a vigorous type. Better nutrient and water absorption from a vigorous rootstock result into high yield in crops (Ruiz and Romero, 1999; Leonardi and Giuffrida, 2006).

Better Nutrient Absorption

The technique of grafting is widely use in the horticultural sectors (including ornamentals, fruits and vegetables) and it aid in better absorption of nutrients and proper utilization of nutrients among different type of plants species (Nawaz *et al*., 2016). Also it has being proven in vegetable crops that grafted plants which has a compatible

rootstock show better absorption of nutrient and water and also utilize nutrients very effectively (Gregory *et al.,* 2013; Albacete *et al.,* 2015). Many researches carried out showed that grafting enhances better uptake of macronutrients like potassium, nitrogen, phosphorous, calcium, magnesium and iron and also in addition of other micronutrients than non-grafted plants (Pulgar *et al.,* 2000; Rivero *et al.,* 2005; Rouphael *et al.,* 2008; Salehi *et al.,* 2010). Research carried by Borgognone *et al.,* (2013) showed that when tomato (*Lycopersicum esculentum* L.,) was grafted onto Maxifort rootstock (*S. lycopersicum* L. x *Solanum habrochaites*) there was better absorption of macro and micro elements and also the growth of scion increase by 11%. Ruiz *et al*. (1997) also find out that there was efficient absorption of nitrogen when melon cultivars 'Yuma' and 'Gallicum' were grafted on three hybrid rootstock of cucurbit which are 'Shintoza', 'RS-841' and 'Kamel. Better absorption and effective use of nitrogen by grafted plant help to minimize low yield in crops mostly caused by insufficient soil fertility (Simonne *et al.,* 2010). In addition grafting has shown to increase absorption and use utilization of phosphorous. Also in another research shows that the presence of phosphorous in grafted plants is much higher than plants which are not grafted (Flores *et al.,* 2010). There is better absorption of potassium in grafted eggplants than eggplants which is not grafted as stated by Leonardi and Giuffrida (2006) from their findings.

Drought Stress

Currently in the world, drought is now a major issue hampering crop cultivation and it is gradually turning into bigger constrains for many growers (Passioura, 1996). Drought is a major challenge causing severe reduction in agricultural production (Isendahl and Schmidt, 2006). Severity of drought has negative impact on growth and development of plants which results in reduction in yield and gigantic financial loss (Pervez *et al.,* 2009). The fast and rapid solution to this issue of drought stress in crop production specifically in fruit vegetables can be solved by grafting onto rootstocks which will be tolerant to drought stress (Garcia-Sanchez *et al.,* 2007; Satisha *et al.,* 2007). Grafting research carried by Clearwater *et al.,* (2004) on drought using fruit from kiwi and grape showed that drought tolerant root stock was important in increasing quality of fruits, growth and production of fruits with regards to drought stress (Clearwater *et al.,* 2004). Nevertheless there is limited research with regards to grafting of fruit vegetables. Comparably eggplant is known to have efficient water absorption than tomato so

therefore it will be more promising to find out the effect of grafting in these crops with regards to water stress (Abadelhafeez *et al.,* 1975). Finding from Schwarz *et al.,* (2010) shows that most grafting done on rootstock which are resistant to drought turn up to increase fruit quality and production. There is also improvement in fruit quality and yield as cherry tomatoes were grafted onto rootstocks which are tolerant to drought (Sanchez-Rodriguez *et al.,* 2011).

High and Low Temperatures

One environmental factor causing wilting, death of plant, delaying of fruit ripening is temperature. This has consequently resulted in low agricultural production and financial deficit (Ahn *et al.,* 1999). Cultivation of vegetables in cold climate condition mostly in winter, spring and autumn expose the crops to chilling temperature, which are problematic to growers in these regions. Most fruit vegetables like cucumber, tomato eggplant, melon, and pepper are sensitive to chilling effect. When the temperature is between 8–12 ˚C these crops can adapt to this condition but the extend of adaptability varies among crops (Hansen *et al.,* 1994; Criddle *et al.,* 1997). Temperature below this figure is detrimental to many horticultural crops orienting from the tropical and sub-tropical regions as this leads to malfunction, necrotic of cell and lastly death of plant. (Allen and Ort, 2001; Enema *et al.,* 2005). Some researchers have shown that grafting watermelon onto root of squash (‘Shintosa’ a hybrid variety) enhances endurance during cold conditions (Davis *et al.,* 2008). Ofori (2015) also reported that, there was better adaptability to cold condition when cucumber was grafted on the rootstock of squash. On the contrary temperature at high rate also has negative effect on growth of plants and crop production and this is as result of physiological, morphological, biochemical and molecular disorder (Schwarz *et al*., 2010). Abdelmageed and Gruda (2009) find out that there were massive improvements in growth and yield when plants were grafted on rootstocks which are tolerant to heat. Eggplant is known to survive under high soil temperature therefore using it as rootstock by grafting it with tomato plant will probably be a positive one during high temperature.

Salinity

The presence of salinity in water bodies and soil is another environmental stress factor, which greatly influence crop yield (Santa Cruz *et al.,* 2002). Also with regards to

irrigation of crop excess application of fertilizer and desertification has led to many salinity problems on arable land across the world and also negatively affecting crop production (Rivero *et al*., 2003). However findings from Colla *et al.* (2006) shows high yield when watermelon plants were grafted on salt-tolerant rootstocks. Plants tolerant to salinity are also obtained by grafting tomato plants on to rootstock genotype which can endure salinity problem (Fernandez-Garcia *et al.,* 2004).

2.9 Grafting Process

Grafting is the act of connecting two living plants parts (root stock and scion) by process of tissue redevelopment, where by connection of plants parts lead to development of a single living plant (Janick, 1986). Before selecting a preferable grafting method certain consideration must be taken into account, which are; type of plant, maturity of the plant, quantity of plants to be grafted, control of the environment and structure of healing (Sakata *et al.*, 2005). There are four procedure in grafting which are; (1) selecting root stock and scion varieties, (2) By a physical process conduct a graft union, (3) the union of the plant parts must heal, (4) grafted plants should be adaptable onto field of cultivation (Lee and Oda, 2010; Lee *et al*., 2010). Most growers perform grafting by hand but in recent time, grafting is done by machine, robots and semi-automatic machines which save time and cost (Lee *et al*., 2010).

Hole Insertion Grafting

Hole insertion grafting is mostly suitable for watermelon due to smaller size of its seedling than the rootstock of squash or bottle gourd. With regards to this method of grafting seeds grown for rootstock are planted 7-8 or 3-4 days earlier for squash and bottle gourd compared to seeds of watermelon. After 7-8 days of planting seeds of watermelon, grafting is than conducted. With regards to aubergine and tomato, seeds grown for rootstock are planted 5-10 days before planting seed for scion. Grafting is then done 20-25 days after planting seeds meant for scion (Lee *et al*., 2010). Prior to grafting, seedling use for grafting should be vigorous, uniform and healthy (Bie *et al*., 2017).

Splice Grafting

Another name given to this type of grafting is one-cotyledon splice grafting or tube grafting. This method is extensively practice by farmers and people who deal in grafting on large scale for sell. Using this method in vegetables is either with a robots/machine or by hand. The shoot meristem and a single leaf of cotyledon are cut at an angle of 35-45°. Splice grafting is commonly used for s*olanaceous* crops (example tomato, eggplant, pepper and potato). Grafting pins, tubes and clips are used for holding the grafting point together (Bie *et al*., 2017).

Cleft Grafting

Another name for cleft grafting is called wedge or apical grafting. With this technique of grafting the seedling meant for rootstock is truncated and a vertical cut is conducted at the middle of stem laterally to axis of the stem. At least 1-3 leaves is taken off the scion and at the portion closer to end of stem, a slant cut is made to produce a wedge. The wedged scion is then put into split portion of the rootstock after which the grafted portion is grip by a clip until the time the grafted portion heals. Cleft grafting is easy to perform in tree plants than vegetables. Grafting of *Solanaceae c*rops such as chilies and aubergine is done by this method but scarcely practice in cucurbits (Lee *et al*., 2010; Johnson *et al*., 2011).

2.10 Significance of Nitrogen for Crop Production and the Environmental Effects

One of the important elements needed for crop production is nitrogen, as plants need it for growth and developments. Also nitrogen is very vital in animals as it helps in production of egg, milk, wool, energy needed for various activities, reproduction, growth and maintenance (Van der Hoek, 2001). In recent time mechanism put in place to increase crop production and retaining of soil fertility involve application of synthetic fertilizer, a typical example is nitrogen. Organic C and N stock in the soil was mainly increase by application of organic manures, leguminous plants and practice of land following. However this ancient practice is replaced by invention of synthetic nitrogen fertilizer which is more efficient and also helps in eradicating elemental constraints (Hirel *et al.,* 2011). Utilization of nitrogen fertilizer over fifty years ago has led to significant increase in fiber, feed and food production across the world (Dobermann and

Cassman, 2004). However dissemination of nitrogen fertilizer is not even across the world leading to limited access in Sub Sahara Africa and this has consequent effect on production of local food which does not meet the demand of growing human population (Greenland, 1998; Syers, 1997). Nevertheless in developed countries mostly in Europe excess use of nitrogen fertilizer have negative impact on environments, to cite some examples are; greenhouse effect, formation of acidic rain and eutrophication (Gastal and Lemaire, 2002; Zhaohui *et al.*,2002). Excess application of nitrogen fertilizer increases the amount of nitrate in water bodies and crops which has harmful impacts on human health as these are sources of food and water for many (Ikemoto *et al.,* 2002). In recent time, there is limitation on accessibility of land for crop cultivation and also the importance of reducing contamination of the atmosphere and water bodies call for effective utilization of nitrogen fertilizer in other to sustain quality of land and increase food production for the rising human population (Cassman *et al.,* 2002).

Source of Nitrogen

The major source of N is N_2 gas, which is made of 78% of earth's atmosphere. Before plants can get access to N, inert N_2 has to be process by biological or by the industrial methods (Goatley and Hensler, 2011). Biological methods convert N_2 gas into ammonium (NH_4^+) and nitrate (NO_3^-) for easy uptake by plants. This method is commonly referred to as biological nitrogen fixation process. It can further by classify as biological and non-biological process. *Rhizobium* bacteria presence in the nodules of legumes is able to transform N_2 gas into readily accessible nitrogen for plants uptake (Symbiotic nitrogen Fixation). Also blue green algae and free living bacteria presence in the soil engage in N_2 fixation process (non-symbiotic nitrogen fixation) (Goatley and Hensler, 2011). Another source of nitrogen common in the soil is derived from decomposition of organic matter. This happens when decomposed organic matter releases ammonia, which reacts with water to form ammonium. Ammonium formed is then converted into inorganic N form (Nitrate) by decomposers (bacteria and fungi) through a method called mineralization. Both ammonium and nitrate can be accessed by plant for growth. With respect to ecosystem, animal feed on plants which end up in producing animal manure and these bye products of animal with plant residue ends up in the soil as source of nitrogen (Zhang and Raun, 2006). In recent time, utilization of nitrogen fertilizer in crop cultivation is a common act because nitrogen fertilizer aid in

improving soil fertility and crop yield. It also helps in better absorption and assimilation of N by plants (Hirel *et al.,* 2011). Nitrogen fertilizer is produce by Haber-Bosch process whereby hydrogen gas reacts with nitrogen gas to produce ammonia. Nitrogen present in the soil is either in organic or inorganic states. Amino acids and amino sugars are organic sources of nitrogen whilst ammonium, nitrate, nitrite nitrous oxide, nitric oxide and elemental nitrogen are inorganic source of nitrogen in the soil (Goatley and Hensler, 2011).

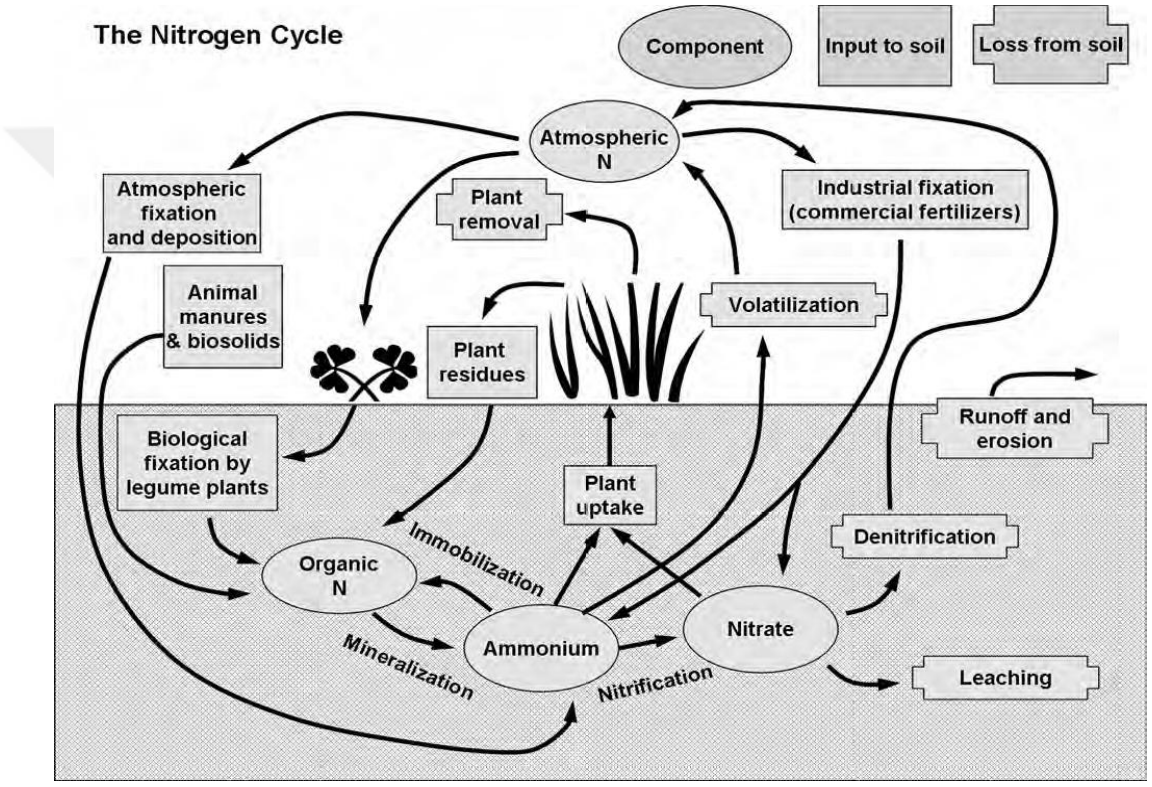


Figure 2.1. Nitrogen cycle (Goatle and Hensler, 2011).

Role of nitrogen in plant growth

Next to carbon, nitrogen is the element needed in large by plants for growth and developments. Nitrogen is basic component of protein, nucleic acid, chlorophyll, co-enzymes, phytohormones and secondary metabolites (Hawkesford *et al.*, 2012). Maximum amount of nitrogen leads increase in photosynthetic activities, leaf area and total amount of assimilation (Ahmad *et al.*, 2009). In plant, total leaf biomass and maximum leaf area are the parameters influencing yield increment in crops (Rafiq, *et*

al., 2010). Therefore, increase in yield among various crops across the world is attributed to high amount of nitrogen with better management practices (Smil, 2001). In addition, nitrogen enhances better root growth, improvement in quality of fruits and regulates growth and development of plants in general (Bloom, 2015; Hemerly, 2016). However in tomato plants nitrogen deficiency causes abnormal growth, yellowing of leaves, reduction in fruit production, decrease in fruit size and also leads to poor quality of tomato fruits. Nevertheless excess nitrogen supply to tomato plant cause reduction in fruit production, increase in vegetative growth and plant lodging (Sainju *et al.*, 2003).

2.11 Nitrogen Use Efficiency in Plants

Nitrogen use efficiency (NUE) in plants can simply be explain as yield gain with regards unit of nitrogen present in the soil (Hirel *et al.,* 2007). Also the explanation from Moll *et al. (*1982) NUE is the grain produce with respect to unit of nitrogen present in the soil. NUE can be categorized into uptake efficiency (quantity of nitrogen taken by plant from the soil/amount of nitrogen present in the soil) and utilization efficiency (yield/nitrogen taken by plant), (Hirel, *et al.,* 2007). For about forty years now application of nitrogen fertilizer in crop production has risen to 7.4 folds whilst total yield gain was 2.4 folds which tells us that NUE has being low for all this years (Tilman, *et al.,*2002). Approximately 50 to 75% of N fertilizer applied on arable land where continuous cultivation of crop is carried out, does not efficiently utilize N fertilizer (Raun, and Johnson 1999; Asghari and Cavagnaro, 2011). Non utilized N fertilizer volatize into the atmosphere, causes surface runoff, leach into water bodies, and also causes denitrification by microbes which are all harmful to the environment (Vitousek, *et al.,* 1997). However nitrogen losses can be decreased by carrying out continuous agronomic practices like crop rotation, utilization of N fertilizer in a rationalize way, imbibing crop residue in the soil and also setting up of crop cover on the field (Malézieux *et al.,* 2009). Furthermore NUE in plant can be enhanced by using genetic modification or breeding varieties that can have very good nitrogen absorption from the soil and effective utilization (Hirel *et al.,* 2007; Good, Shrawat and Muench, 2004). In recent time, many studies are carried out in the fruit and vegetable sector on how to generate new farming methods and practices which will enhance maximum N uptake by plants and at the end reduces waste of N fertilizer. In addition, the negative effect of N fertilizer on the environment can be minimized by choosing varieties that

can maximize nitrogen use efficiency and increase crop production (Lynch, 1998). However, reduction in crop production which is mainly caused by less NUE in high yielding varieties can be overcome by grafting them onto rootstocks that has the ability to enhance NUE in scion. Nevertheless NUE in plants is influenced by factors from its surroundings, some of which are temperature, radiation, rainfall and it effect on insects, diseases, root microbes and allelophathy (Baligar *et al.*, 2001).

2.12 Genotypic Variation in Nitrogen use Efficiency

In recent time, the rising need of food by the growing human population across the world calls for farming practices which are safe to the environment and also prevent prospect problems on climate challenges due to excess application of N fertilizer by farmers in cultivation. These challenges could be solved by enhancing nitrogen use efficiency in plants (Zhang *et al.,* 2015). A genotype is considered to be N efficient either when it has ability to transform N taken from the soil into yield which is relatively effective than other genotype (Sattelmacher *et al.,* 1994) or when it is able to produce yield higher than regular quantity with respect to low rate of N (Gueye and Becker, 2011). Differences in genotype arise due to heritable traits of specific crop specie or variety, which differs in growth or production in relation with other species or similar species in a bearable or unbearable condition for cultivation (Fageria, 1989). Functional and physical traits (root, leaves etc) of a genotype influences N absorption and utilization (Schenk, 2004; Thorup-Kristensen and Sørensen, 1999; Thorup-Kristensen and van den Boogaard 1999). Nevertheless similar genotype receiving varied rates of N exhibit different level of N efficiency (Burns, 2004). Also absorption of N after flowering in plant is a strong evident to identify genetic variation in crop yield at higher or low rate received (Berry *et al.,* 2010, Ulas *et al.,* 2013). In crops, maize, alfalfa and rice are known to exhibit genotypic variation with respect to N efficiency (Harrison *et al.,* 2004, Lawlor, 2002; Machado and Fernandes, 2001). Also research carried by Ruiz and Romero (1998) shows that, genetic variation in tomato occurs as result of absorption and use of foliar nitrate. Acquiring much information on characteristics which related to NUE is very vital in identifying cultivars which are N efficient (Ji, 2014). Researchers have constantly tried for about 30 year to breed and come out with cultivars which are nutrient efficient and also eager to know what causes nutrient efficiency in plants but the possibility of developing cultivars which are

nutrient efficiency is inadequate (Fageria*et al.,* 2008). Furthermore to get cultivars that are nutrient efficient, better knowledge on concept of genetics in N uptake and N utilization at various stages of plant growth is needed (Loudet *et al.,* 2003).

2.13 Plant Characteristics Influencing Nitrogen Efficiency

Environmental issues and limited access to resources for extensive agricultural cultivation calls for the search of plants which are more nutrient efficient. Also effort of breeding nutrient efficient plants is hindered by the reliable screening criteria. Hence, there is the need to know metabolic activities and characteristics which are important to nutrient efficiency (Sauerbeck and Helal, 1990). In plants NUE is as results of uptake efficiency (where N is taken from the soil or N fertilizer) and utilization efficiency (extracted N is converted into yield) (Hirel *et al.,* 2007). Therefore recognizing mechanisms in relation to absorption, translocation, utilization and mobilization in crop plants should be intensive in order to improve efficient utilization of N relations (Sauerbeck and Helal, 1990). The root functions as a system, which absorb water and mineral nutrient including N from the soil (Wu *et al.,* 2005). Nutrient efficiency in plant is enhanced when the plant has more root hairs per unit of root length, more root density and longevity and also maximum rate of root penetration (Wastermann, 2005). Soil pH, soil texture and density, diseases, pests, water log and drought are some factors influences rooting depth with respect to N availability and absorption (Vos, 1997). Nevertheless N absorbed by root and leaves is converted into amino acid (Hirel and Lea, 2001). The amino acid formed is later use in production of enzymes and protein, which turn to be the building block of plant structure and photosynthetic system (Mae *et al.,* 1983). In plants, remobilization of stored N starts at post flowering stage. The shoot/and or root at this stage engage in translocation of N in form of amino acid to storage and reproductive organs (example seeds, trunk or bulb) and this occurs as result of hydrolysis of protein (Masclaux *et al.,* 2001).

CHAPTER THREE

MATERIALS AND METHODS

Two hydroponic experiments (Exp.1 and Exp. 2) were conducted by using a non-flow nutrient film technique (NFT) in a controlled growth chamber situated in the Plant Physiology Laboratory of Erciyes University, Faculty of Agriculture Kayseri Turkey. The average day/night temperatures was 25/22 °C, the relative humidity was 65-70% and about 350 μmol m^{-2} S^{-1} photon flux was supplied in a photoperiod of 16/8 h of light/dark regimes in the controlled growth chamber. Two hydroponic experiments were arranged in a completely randomized block design with four replications and two plants in each pot (replication). In both experiments, all nutrients were supplied to the plants by using a non-flow nutrient film technique (NFT) experienced in 8 L plastic pots. The nutrient solution was prepared by using distilled water contained analytical grade (99% pure) chemicals according to modified Hoagland and Arnon formulation. Nutrient solution in each 8 L pot was continuously aerated with using an air pump. Both experiments were monitored daily and data collected on specific intervals. Data on plant height, leaf number, leaf chlorophyll (SPAD) value, photosynthetic rate, and leaf senescence were collected on weekly basis, whilst data on shoot and root biomass, leaf area index were measured at the end of the experiment. Other parameters that were measured at the end of the experiment were; *Nitrate reductase* (NR) enzyme activity, leaf chlorophyll, carotenoid contents and root morphology (root length, root volume and average root diameter).

3.1 Plant Material

The plant materials used in these experiments were 6 commercial hybrid tomato cultivars (*M1103f1 Maydof1, Perfectf1, Helenaf1, Falcon, Riogrande*) and 7 local tomato cultivars obtained from Aegean Agricultural Research Institute (*SN48, SN159,*

SN160, SN456, MSC50, Ege50, Mass1001), and 1 local tomato cultivar (*Karahidir*) from Kayseri in Turkey and also 4 local tomato cultivars (*ATS02, ATS06, ALT, P005*) obtained from crop research institute, Kumasi-Ghana.

3.2 Treatments and Experimental Design

With regards to both experiment, two different N levels (low-N: 0.3 mM and high-N: 3.0 mM) were used as treatments for the solution cultural experiments. In the first experiment, treatments were laid in a Completely Randomized Block Design with three replications. Tomato seeds were pre-germinated in petri dishes and hardened in peat media for about 7-10 days. Two seedlings of each variety were transplanted into 8 liter containers containing a modified Hoagland nutrient solution which was grown for 5 weeks. Half rate of nutrient solution was given in the first week of transferring the tomato seedling into the nutrient solution as a precaution against any injury to the seedlings. All other factors of humidity, light intensity among others were maintained constantly. In the second experiments, two cultivars which were N-efficient were selected to be used as rootstock whilst another two N-inefficient cultivars were selected to be used as scion. Treatments were again laid in a Completely Randomized Block Design with three replications. Selected tomato seeds were pre-germinated in petri dishes and hardened in peat media for about 7-10 days. Tomato seedling were than grafted by using tube grafting method, after which grafted tomato seedling was place in transparent plastic container which was covered. The tomato seedling was left in the plastic contain for 7 days in order to allow healing of the grafted parts. Two seedling of the grafted tomato seedlings were transplanted into an 8-liter containers containing a modified Hoagland nutrient solution which was grown for 5 weeks. Half rate of nutrient solution was again given in the first week of transferring the grafted tomato seedlings into the nutrient solution as a precaution against any injury to the grafted tomato seedlings. All other factors of humidity, light intensity among others will were again maintained constantly.



Figure 3.1. Setup of the first experiment (screening for efficient N variety)



Figure 3.2. Setup of the second experiment (determining root potential of the varieties via grafting)

3.3 The Nutrient Solution

The nutrient solution used for both experiments was prepared following the procedures of Hoagland's plant nutrient solution formula. Into each of the 8L container, we gave: 750 μM K_2SO_4; 500 μM KH_2PO_4; 650 μM $MgSO_4$; 50 μM NaCl; 10 μM H_3BO_3; 0.5 μM for $MnSO_4$ and $ZnSO_4$; 0.4 μM for $CuSO_4$ and $MoNa_2O_4$. Also given was 80 μM Fe-EDDHA. For the high N treatment, 750 μM and 2250 μM of $(NH_4)_2SO_4$ and $Ca(NO_3)_2$ were given respectively. Whilst 75 μM $(NH_4)_2SO_4$ and 225 μM $Ca(NO_3)_2$ were given for the low N treatment. To avoid damage to root cells from the use of distilled water, 1000 μM per liter of $CaSO_4$ was added to the solution. Electric conductivity (EC) of 1.0 was maintained whereas a teaspoon full of $CaCO_3$ was applied to each container as a buffer to keep a 7.0 pH. The nutrient solution was monitored on daily basis in order to correct any elemental deficiencies. The solution was constantly replaced at a 7-day interval.

3.4 Measurements

3.4.1 Leaf Single-Photon Avalanche Diode (SPAD)

With respect to the first and second experiments, the leaf chlorophyll was measure after the second week of the start of each experiment using chlorophyll meter SPAD 502 Plus. This was constantly done on a weekly basis to the end of these experiments.



Figure 3.3. Chlorophyll meter

3.4.2 Photosynthesis

In both experiments, photosynthesis rate was measured with Portable Photosynthesis System LI-COR (LI-6400XTP) on a weekly basis from the second week to fourth week. Well-developed leaf was randomly selected from one plant in each replication for measuring the photosynthetic rate.



Figure 3.4. Portable Photosynthesis System LI-COR (LI-6400XTP)

3.4.3 Leaf Senescence

Deficiency of nitrogen plant system is also known to cause leaf senescence, this happens when leaves experience tip burn and yellowing of leaves. This normally occurs in old leaves as N is remobilize from old leaves to young leaves which is as result of N deficiency (Činčerová, 1990). In both experiments, the leaves of the plants were monitored keenly and from the onset of leaf senescence daily record were taken.

3.4.4 Leaf Area

Total leaf area (LA) of the tomato plants was measured in the first and second experiments with leaf area device LI-COR (LI 3100, model C). The measurement was taken at time of harvest and also measurement was recorded in centimeters square (cm^2).



Figure 3.5. Leaf area meter

3.4.5 Shoot and Root Biomass

The fresh weight of both shoot and root were measure in gram (g) within time of harvest. After which the measured samples were dried using an oven dryer at a

temperature of 75°C. The drying lasted for 10 days to retain a moisture content of 5% in both experiments. Also after drying the samples, the dry weight of shoot and root were measured accordingly.

3.4.6 Plant Height

The height of the plants in both experiments was measured on a weekly basis with a measuring tape. The measurement was taken from the base of the stem to the shoot apical meristem.

3.4.7 Nitrate reductase enzyme (NRA) activity

The nitrate reductase enzyme analysis was done using procedure suggested by Harley (1993). The analysis was done during the harvesting period in both experiments. The steps involves as follows:

- Fresh sample of plant was taken and weighed at 2 grams per samples after which it was chopped in pieces
- Chopped sample was placed into falcon tubes labeled time-0 (T_0) and time-60 (T_{60}).
- To prevent exposing the sample to light rays, the tubes were covered with aluminum foil.
- 10 ml of assay buffer solution, [100 mM phosphate buffer, pH 7.5; 30 mM KNO_3; 5 % (v/v) propanol] was added to each sample of T_0 and T_{60}.
- T_0 container was quickly put into water bath containing boiling water for 5 minutes after which it was removed and cooled at room temperature. On the other hand T_{60} was kept at room temperature for 60 minutes, after which the samples were put into the water bath also containing boiling water for 5 minutes and later cooled at room temperature.
- Colour development reagent was added to the samples to help detect nitrite. This was done by adding 5 ml of sulphanilamide (1% sulphanilamide in 25 % HCl) and 0.02% N-(1-naphthyl)-ethylenediamine hydrochloride (NEED) solutions and then shake energetically and allow colour to develop within 20 minute interval.

- For each sample the optical density of each standard tube was measured at a wave length of 540 nm by using a spectrometer. The nitrate reductase (NR) was calculated as *"micromoles of nitrite produced per hour per gram of fresh weight"* using the formulae below.

$$\mu\text{moles } NO_2^- \, h^{-1} \, g^{-1} = \frac{\mu\text{moles } NO_2^- (T_{60}) - \mu\text{moles } NO_2^- (T_0)}{\text{Weight of plant sample}}$$



Figure 3.6. Nitrate Reductase Enzyme measurement with UV spectrometer

3.4.8 Leaf Total Chlorophyll and Carotenoid Content

During the time of harvest 0.1 gram (g) of fresh leaf samples were taken from each replication of both treatments in order to determine the leaf total chlorophyll and carotenoid contents by using the T80+ UV-VIS Spectrometer (PG Instruments Limited, UK).Taken samples were placed in capped plastic tubes and 10 ml of ethylene alcohol (95% concentration) was given to each sample. After which it was placed in darkness at room temperature for a night, this was done to aid in extraction of leaf pigment. The samples were measured in the first and second experiment respectively at wavelength of 470 nm, 648.6 nm and 664.2 nm. From the measurement of the spectrometer, Total chlorophyll (Total-chlo) and total carotenoids (TC) was calculated by using Lichtenthaler's (1989) formulae.

$$\text{Total-Chlo} = \frac{(5.24\ WL664.2 - 22.24\ WL648.6) \times 8.1}{\text{Weight of plant sample (g)}} \text{ [mg/g plant sample]}$$

$$TC = \frac{[(4.785\ WL470 + 3.657\ WL664.2) - 12.76\ WL648.6] \times 8.1}{\text{Weight of plant sample (g)}} \text{ [mg/g plant sample]}$$

Note: *WL470, WL648.6 and WL664.2 refer to spectrometric readings at wavelength 470 nm, 648.6 nm and 664.2 nm respectively.*



Figure 3.7. UV Spectrometer (measurement of total Chlorophyll and Carotenoid contents)

3.4.9 Root Length, Area and Volume

The total root length (cm), area (cm^2) and volume (cm^3) of both experiments were measured with WinRhizo (Regular LA2400 of Regent Instruments Inc.) image analysis program in combination with Epson Expression 11000XL scanner. From each sample fresh harvest roots of 5 grams was taken for analysis. The root samples were put into the scanning tray after which water was added and with the help of plastic forceps the roots were uniformly spread across the tray. It was then scanned with WinRhizo system's installed on a computer which was connected to the scanner. Total plant root length, area and volume were measured by ratio of root weight of sample to the total root weight.



Figure 3.8. Epson scanner used for root scanning

3.4.10 Analysis of Protein/Nitrogen Determination

Plant growth system is controlled and enhanced by N being a major component in many enzymatic proteins (Sinfield*et al.,* 2010). Kjelddahl in 1883 proposed a procedure for measuring amount of nitrogen, which is the basis for protein analysis and this procedure has being used by many scientists for over 130 years (Sáez-Plaza*et al.,* 2013). This method (Kjeldahl nitrogen determination method) was used in measuring the protein/nitrogen content of the various tomato genotypes (for first and second experiments) under the high and low rate of nitrogen. With respect to each leaf sample;

1. 0.2 grams (g) leaf sample from each replication were weighed and poured into a digested tube and one Kjedahl reaction tablet was added to each sample.
2. 10 ml of H_2SO_4 (98% concentration) was added and heated for four hours at 400ºC beginning from 100ºC for 30 minutes and later increased by 50ºC at every 30 minutes interval until the 400ºC mark was reached.
3. The digested samples were allowed to cool and 30 ml of distilled water was added to the samples to dilute it.
4. The digested samples were put into a distilling unit and distilled with 50 ml 0f 35% (w/v) NaOH for a period of two minutes. Distilled samples were taken for quantity of 10 ml (2% w/v) with a container, which also contained combination of boric acid and indicator solution.
5. Titration was than perform by adding 0.05 normality of H_2SO_4 to the content. Titration was performed with a digital titrator. The N% content was calculated using the formula stated below.

$$\text{Nitrogen (\%)} = \frac{[(\text{Sample titre value} - \text{Blank titre value}) \times (\text{N of acid} \times 1.4007)]}{\text{Weight of plant sample (g)}}$$



Figure 3.9. Total Nitrogen determination

Note:

Combination of boric acid and indicator solution was prepared by using this procedure; Methyl red and Bromocresol green was weighed at 0.1 gram and poured into two different transparent containers which was then dissolved with 100ml of ethylene alcohol. Simultaneously 1.0 litre of boric acid with 2% concentration was prepared by dissolving 20 grams(g) of boric acid powder into distil water. 7 ml and 10ml of Methyl red and Bromocresol green was added accordingly to the prepared boric acid and mixed. At the end the pH was checked and controlled to 5.0.

3.4.11 Element Analysis

At the end of the experiment first and second, element analysis was done to determine the quantity of minerals accumulated by the shoot and the root of the various tomato genotypes. This was done by ashing 0.5g of dry samples in the oven at 450 °C for four hours. 5 ml of HCl with 20% concentration was added to the ashed samples and filtered (Figure 3.11). The filtered solutions were then diluted with distilled water to a volume 50 ml. Out of which 10 ml was taken for Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES) analysis. The ICP-AES results were converted into percentages (%) and parts per million (ppm).



Figure 3.10. Preparation of samples for element analysis

3.5 Data Analysis

Statistical analysis of the both nutrient solution experiments data was performed using SAS Statistical Software (SAS University Edition, Institute Inc., Cary, NC, USA). A two-factorial analysis of variance was performed to study the effects of genotype (Experiment I) or grafting combination (Experiment II) and nitrogen and their interactions on the plants. Levels of significance are represented by *P ≤ 0.05, **P ≤ 0.01, ***P ≤ 0.001, and ns means not significant. Differences between the treatments were analyzed using Duncan's multiple test (P ≤ 0.05).

CHAPTER FOUR

RESULTS

4.1 Results of Experiment I

4.1.1. Shoot Biomass Production

From the data collected at the end of the experiments indicate that fresh and dry shoot weight of plants under high N rate was higher than that of low N rate (Fig. 4.1 and Fig. 4.2). The mean fresh shoot weight under high N rate was 355.69 g with *ALT* (499g), *Maydo* (471.67g), *PerfectF1* (435.33g), *Mass1001* (428g), *HelenaF1* (426g) and *MSC50* (422g) respectively producing the highest fresh shoot weight whilst *P005* (248.67g), *Riogrande* (269g) and *Falcon* (273.5) producing the lowest fresh shoot weight under high N rate. Also under low N rate 105.35 g of mean fresh shoot weight was produced, the highest fresh shoot biomass was recorded in *SN159* (160g), *Maydo* (133.33), *ATS06* (126g) *Helen*a (116g) , *M1103F1* (116g) and *ALT* (110g) whilst the lowest fresh shoot weight was recorded in *Karahidir* (78g), *SN456* (84g), *P005* (88g), *SN48* (88g) and *PerfectF1*(88g). The mean of shoot dry weight under high N rate was 30.85 g, *ALT* (46.96g), *Maydo* (40.93g), *HelenaF1* (39.86g), and *PerfectF1* (38.71g) produced the highest shoot dry weight whilst *ATS02* (12.75g), *Falcon* (17.40g) and *ATS06* (21.38g) produced the lowest shoot dry weight. Also with respect to the mean of dry shoot weight under low N rate 11.14 g was produced, where *MaydoF1* (14.77g), *HelenaF1* (14g), *MSC50* (12.83g) produced the highest biomass and *Karahidir* (6.65g), *ATS06* (7.83g) and *P005* (9.27g) produced the lowest biomass. From the analysis of variance of fresh and dry shoot weight indicates that, there were significant differences ($P < 0.001$) among variety. Also nitrogen rates significantly ($P < 0.001$) affect fresh and dry shoot weight of various tomato varieties. In addition, interaction between nitrogen supply and the varieties shows to be significant ($P < 0.001$).

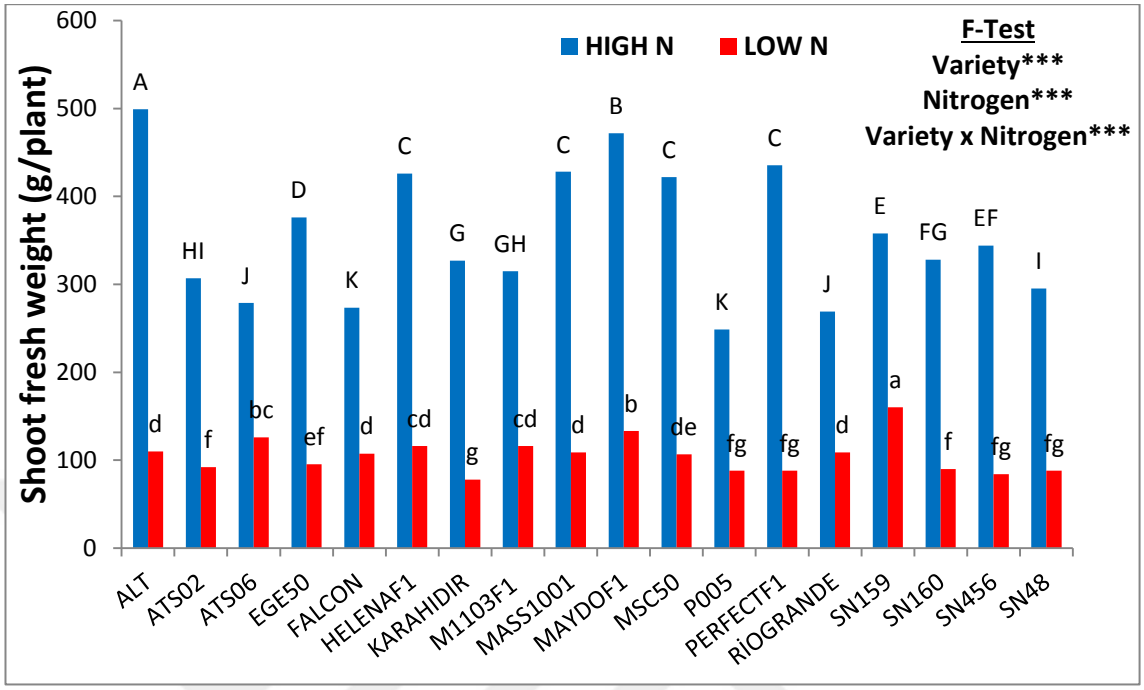


Figure 4.1. Shoot fresh weight of different tomato varieties under high and low N rates.

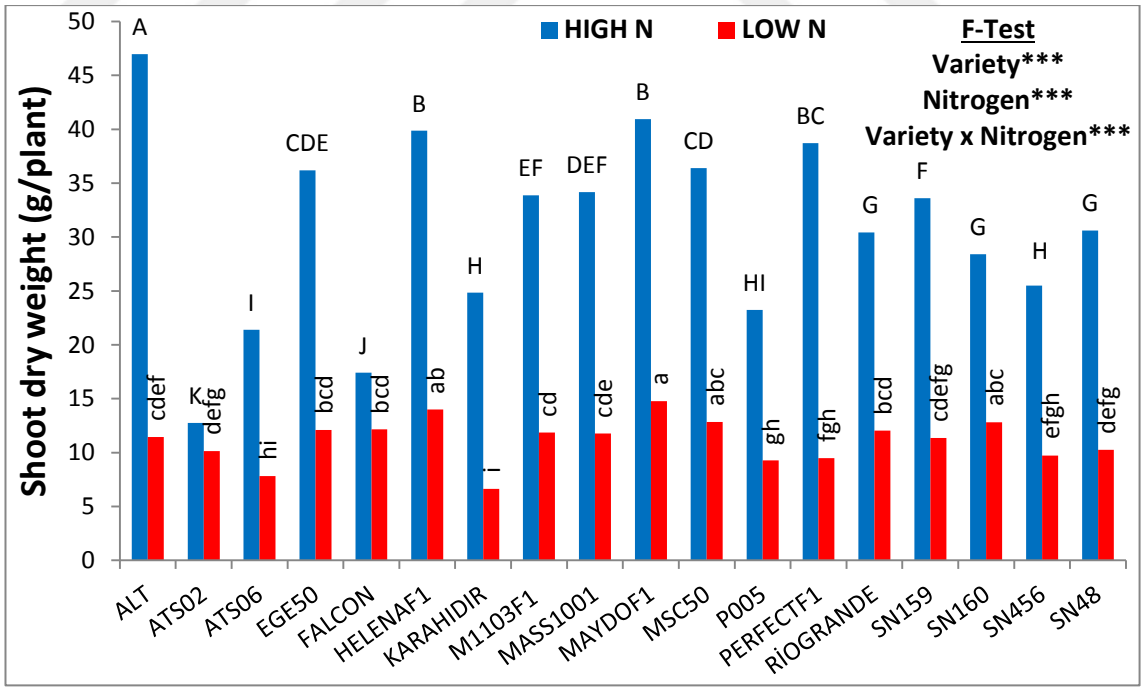


Figure 4.2. Shoot dry weight of different tomato varieties under high and low N rates.

4.1.2. Root Biomass Production

Similarly as the shoot biomass, root biomass of the plants under high N rate was greater than low N rate (Fig. 4.3 and Fig. 4.4).Results from the fresh root weight indicates that under high N rate, a mean of 59.31 g was produced where *Riogrande* (87.66 g), *SN456* (83.1g), *M1103F1* (81.17g), *MSC50* (74.75g), ALT (74.06g) and *HelenaF1* (69.85g) produced the highest root biomass whilst *ATS06* (35.8g), *P005* (37.56g), *Falcon* (40.13g) and *EGE50* (46.8g) produced the lowest root biomass. Also under low N rate of fresh root weight, a mean of 44.18 g was produced where *ALT06* (55.03g), *Riogrande (*54.5g), *ATS06* (52.6g) and *PerfectF1* (49.35g) produced the highest fresh root biomass whilst *Karahidir* (29.49g), *EGE50* (33.2g) and *MSC50* (35.87g) produced the lowest fresh root biomass. In Addition with respect to root dry weight under high N rate, a mean of 3.13 g was produced where *M1103F1* (4.65g), *PerfectF1* (4.46g), *ALT* (4.43g) and *SN456* (4.23g) produced the highest dry root biomass whilst *ATS06* (1.35g), *P005* (2.03g), *Falcon* (2.09g) and *Karahidir* (2.22g) produced the lowest dry root biomass. Also root dry weight under low N rate produced a mean of 2.28g where *ATS06* (2.82g) and *Riogrande* (2.76g) produced the highest dry root biomass whilst *Karahidir* (1.25g) and *EGE50* produced the lowest dry root biomass. With regards to fresh and dry root weight there was significant differences ($P < 0.001$) among varieties and also there was significant difference ($P < 0.001$) among N rate as well as interaction between variety and nitrogen supply. A keen observation from our results of shoots and root biomass indicates *ALT* and *HelenaF1* performed better than all the varieties whilst *P005* and *Karahidir* had the worst performance among the varieties.

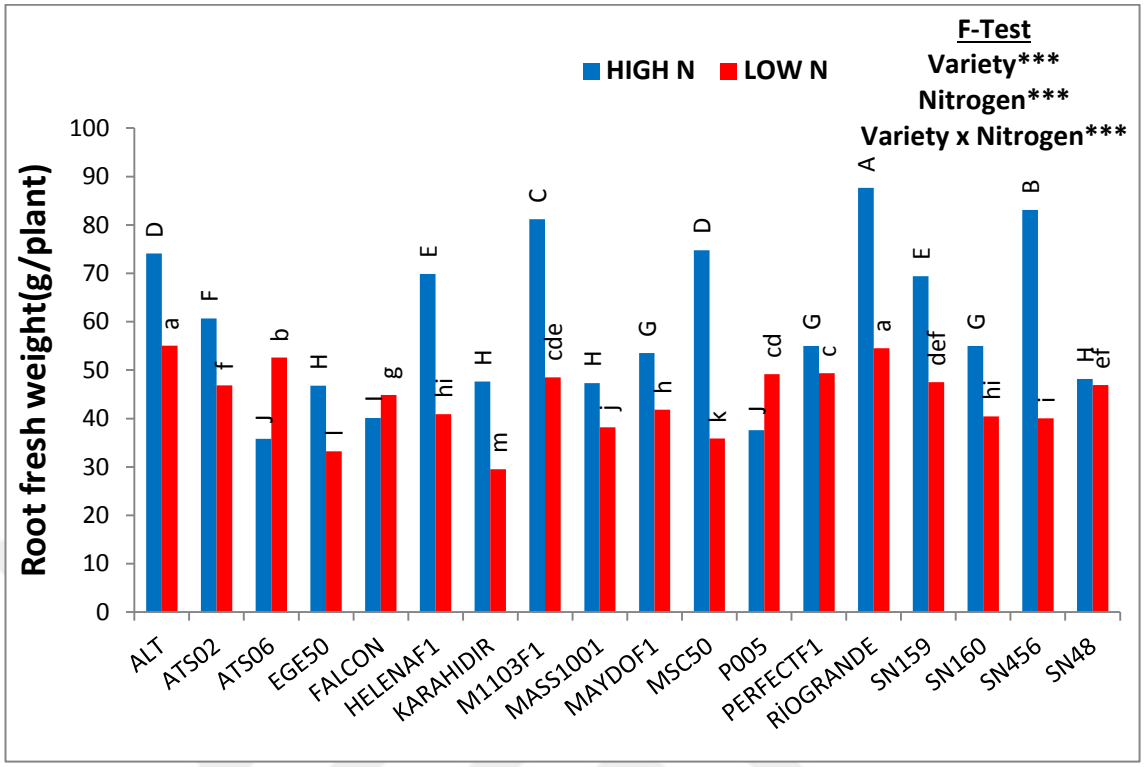


Figure 4.3. Root fresh weight of different tomato varieties under high and low N rates.

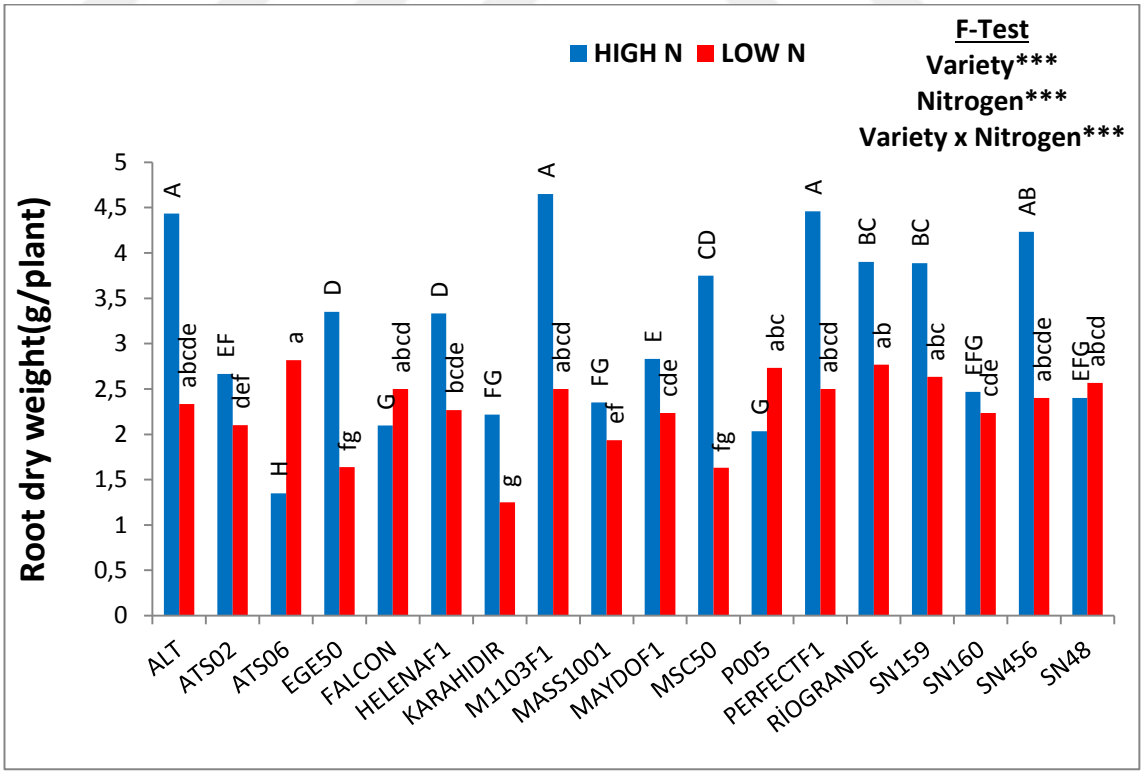


Figure 4.4. Root dry weight of different tomato varieties under high and low N rates

4.1.3. Leaf area

Results for the Leaf area analysis indicate that there was significant difference among varieties ($P < 0.001$); also the effect of N rate on leaf area among various varieties was significantly different ($P < 0.001$). In addition, the interaction between N rate and varieties with respect to leaf area was significant ($P < 0.001$) (Fig. 4.5). As indicated beneath *ALT, EGE50, Mass1001* and *HelenaF1* recorded the highest leaf area respectively under high N rate whilst *ATS02, SN48 and P005* also had lowest leaf area respectively under the same high N rate. Under low N rate *MaydoF1, PerfectF1,ATS06* and *ALT* recorded highest leaf area respectively whilst *P005, SN456* and *Mass1001* recorded the lowest leaf area respectively.

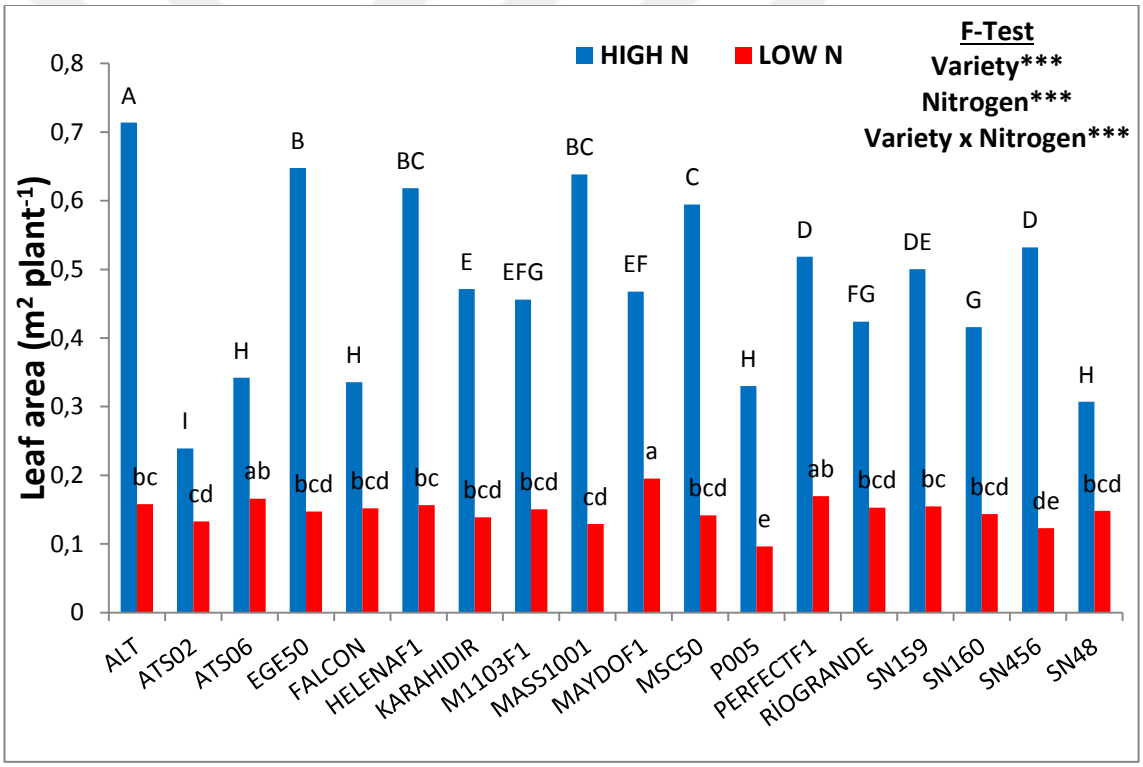


Figure 4.5. The effect of N levels on total leaf area.

4.1.4. Root length, volume and diameter

From the experiment conducted it was observed that the root length of plants under high N rate was greater than those under low N rate except *ATS02* and *ATS06* (Fig. 4.6). Also with exception of *ATS02*, *ATS06* and *Falcon* plants under high N rate produced root volume which is greater than that under low N rate (Fig. 4.7). In addition, plants under high N rate produced higher root diameter than those under low N rate with exception

of *ATS02*, *ATS06*, *Falcon* and *P005* (Fig. 4.8). Root length, volume and diameter varied significantly ($P < 0.001$) among varieties. Also the effect of N rate on various root parameters (length, volume and diameter) was significant. In addition, there was significant difference ($P < 0.001$) among variety and nitrogen supply interactions. Under high N rate the mean total root length, volume and diameter were 1656.97 m, 101.02 cm^3 and 16.25 mm respectively. *ALT*, *SN456* and *M1103F1* recorded the highest root length whilst *ATS02*, *ATS06* and *Falcon* recorded the lowest root length respectively under high N rate. *M1103F1*, *MSC50* and *ALT* had the highest root volume whilst *ATS06*, *ATS02*, *Falcon* and *P005* recorded the lowest root volume respectively under high N rate. *ATS06* and *P005* produced the lowest root diameter whilst *Riogrande*, *SN456* and *M1103F1* produce the highest root diameter under N rate. Also under low N rate the mean total root length, volume and diameter were 1030.67 m, 65.74 cm^3 and 12.91mm respectively. *ATS06*, *ALT*, *ATS06* and *HelenaF1* produced the highest root length whilst *karahidir*, *MSC50* and *SN48* produced the lowest root length. Also *ALT*, *ATS06* and *Riogrande* produced the highest root volume whilst *EGE50*, *MSC50*, *Karahidir* produce the lowest root volume. Lastly *Riogrande*, *SN159* and *ATS02* produce the highest root diameter whilst *EGE50*, *Karahidir* and *MSC50* produced the lowest root diameter.

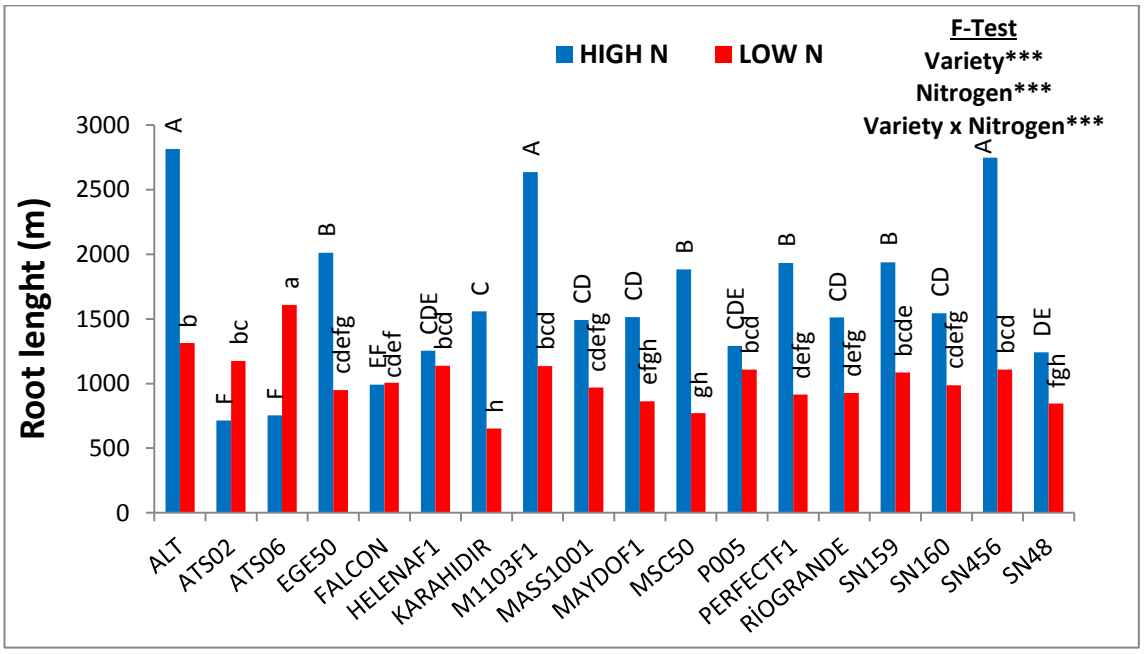


Figure 4.6. Root length. Means with same letter are not significantly different

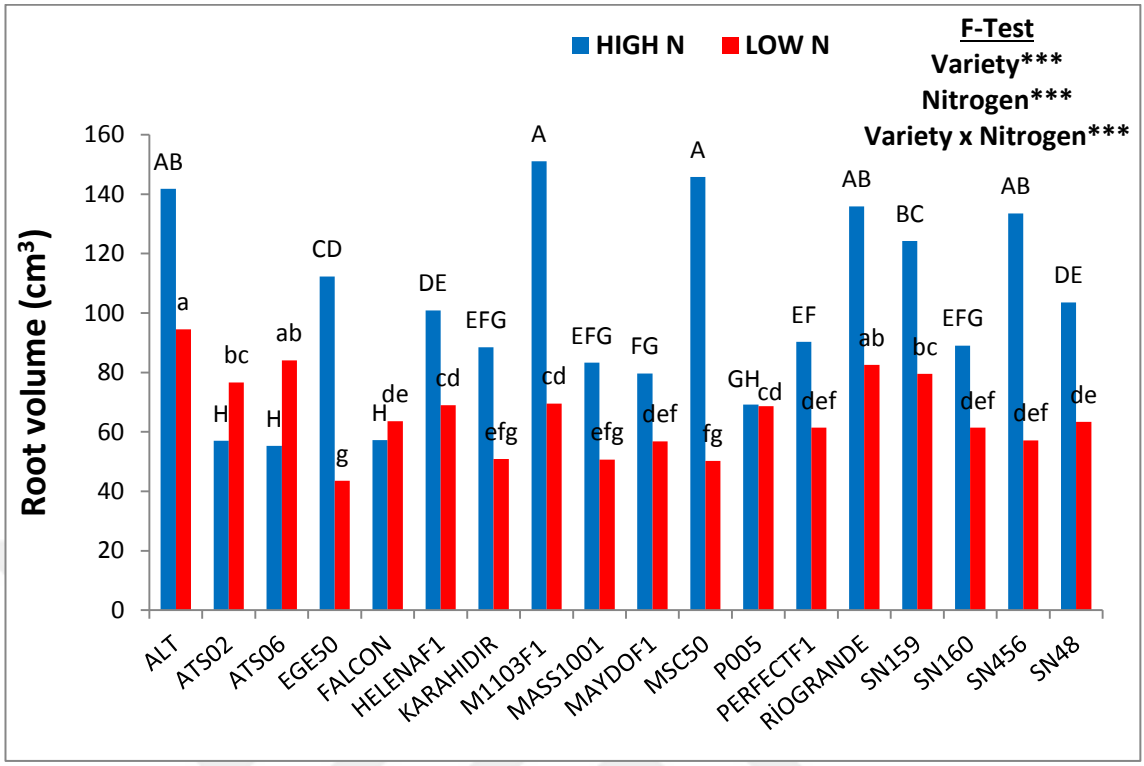


Figure 4.7. Root volume. Means with same letter are not statistically different.

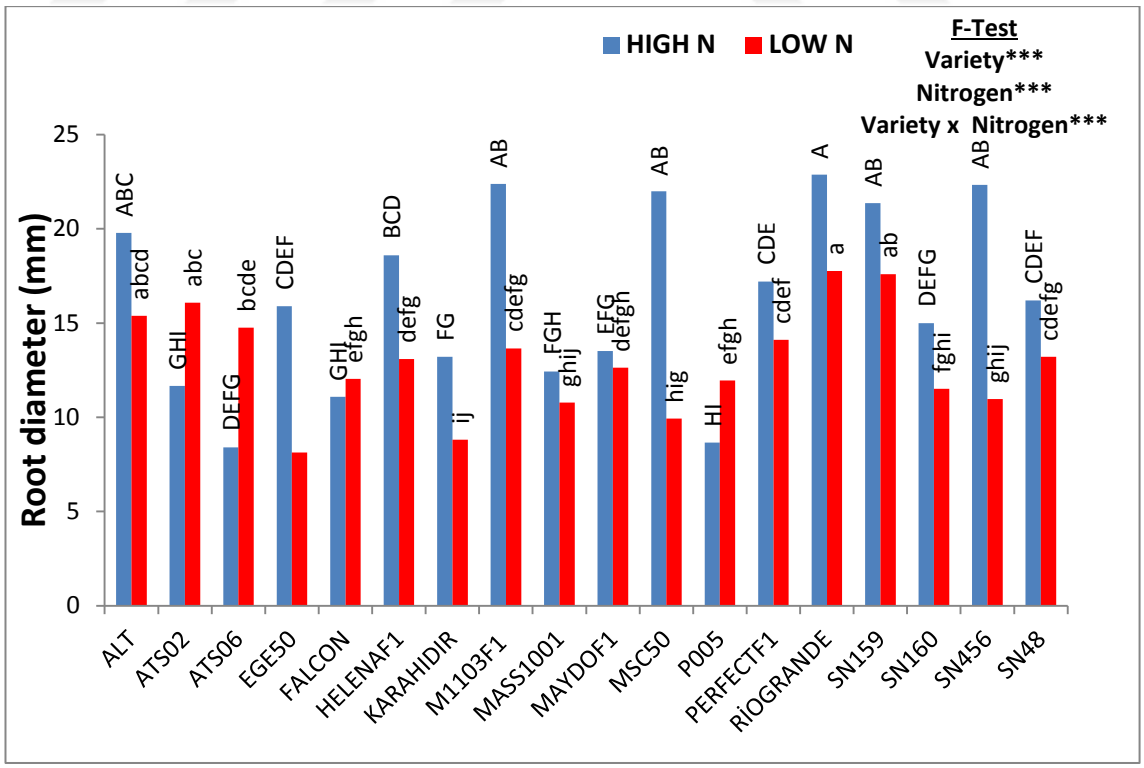


Figure 4.8. Root diameter. Means with same letter are not significantly different.

4.1.5. Nitrate Reductase Activity

One key factor of nitrogen use efficiency considered in measurement of crop is analyzing of nitrate reductase (NR) activity (Fig. 4.9). Analyzing the NR activity of the tomato varieties used for this research shows significant genotypic differences ($P < 0.001$) in NR activity. Nitrogen rates also significantly ($P < 0.001$) affected the NR activity. There was also significant interaction ($P < 0.001$) between nitrogen supply and varieties. The NR activity present in the leaf was greater in high N rate than low N rate of plants except *Falcon* and Mass*1001*. *Riogrande*, *ATS06* and *EGE50* were the highest under high N rate whilst *SN456*, *FALCON and MSC50* were the lowest under high N rate. The rest of the varieties did not vary significantly from one another and were within extreme. Also under low N rate *MASS1001*, *Riogrande* and *EGE50* where the highest whilst *SN456*, *P005* and *MSC50* where the lowest.

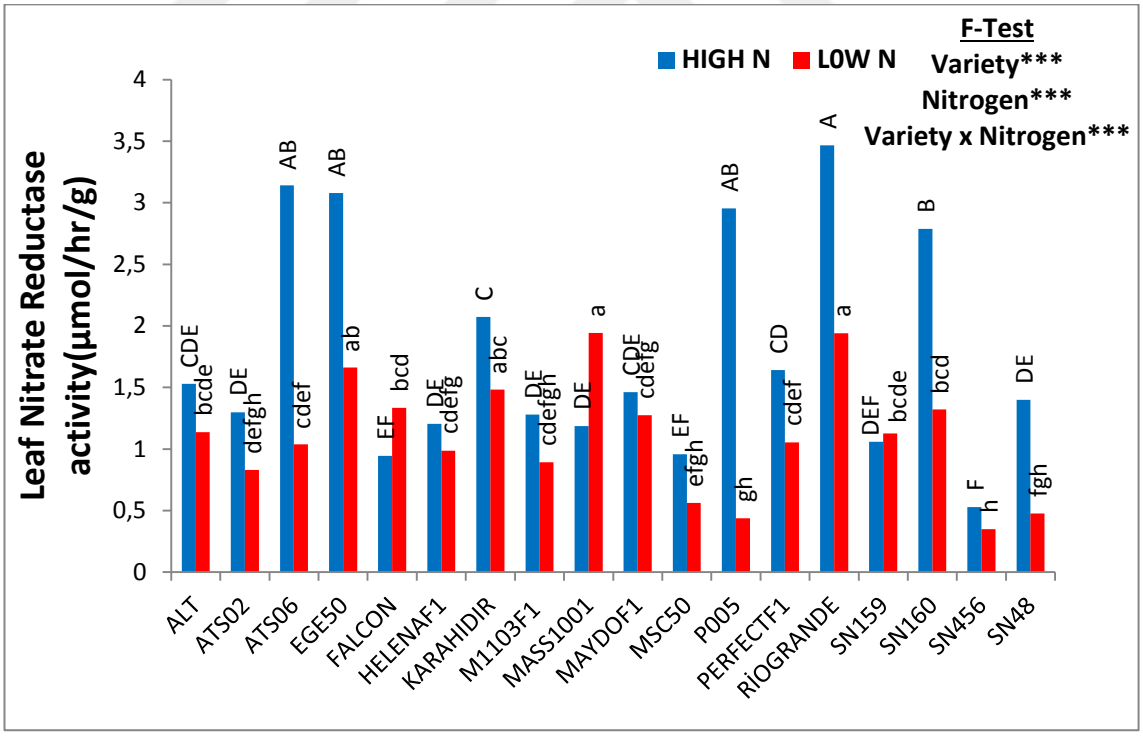


Figure 4.9. Leaf nitrate reductase activity rate. Means with the same letter are not significantly different.

4.1.6. Shoot and root N concentration

From the experiments it was observed that *Riogrande, SN456, HelenaF1* recorded the highest shoot N concentration respectively under high N rate whilst *P005, MSC50, MaydoF1* and *SN48* recorded the lowest shoot N concentration respectively under high N rate (Fig. 4.10). However under low N rate *M1103F1, Karahidir, PerfectF1, SN48* and *Riogrande* recorded the high shoot N concentration respectively whilst *HelenaF1, MASS1001, P005, SN159* recorded the lowest shoot N rate respectively under low N rate. Results of root N concentration also indicates that *MaydoF1, Riogrande, ATS06* recorded the highest root N concentration respectively under high N rate whilst *EGE50, SN456, MASS1001, ALT, SN159* and *P005* recorded the lowest root N concentration respectively under high N rate (Fig. 4.11). However under low N rate *Falcon, MASS1001, and ALT* recorded highest N root concentration respectively under low N rate whilst *ATS06* and *Riogrande* recorded the lowest root N concentration under low N rate. The shoot and root N concentration saw significantly ($P < 0.001$) among the varieties. Again shoot and root N concentration of various plants were significantly ($P < 0.001$) affected by N rates. Significant difference ($P < 0.001$) also existed between nitrogen supply and variety interaction of both shoot and root of various plants.

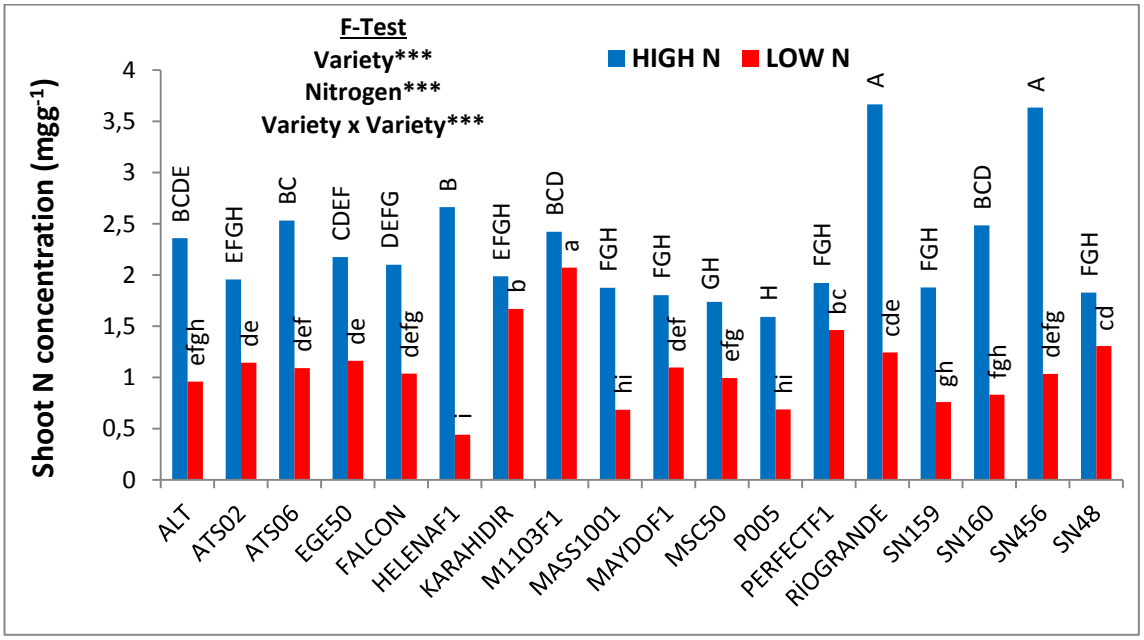


Figure 4.10. Shoot N concentration

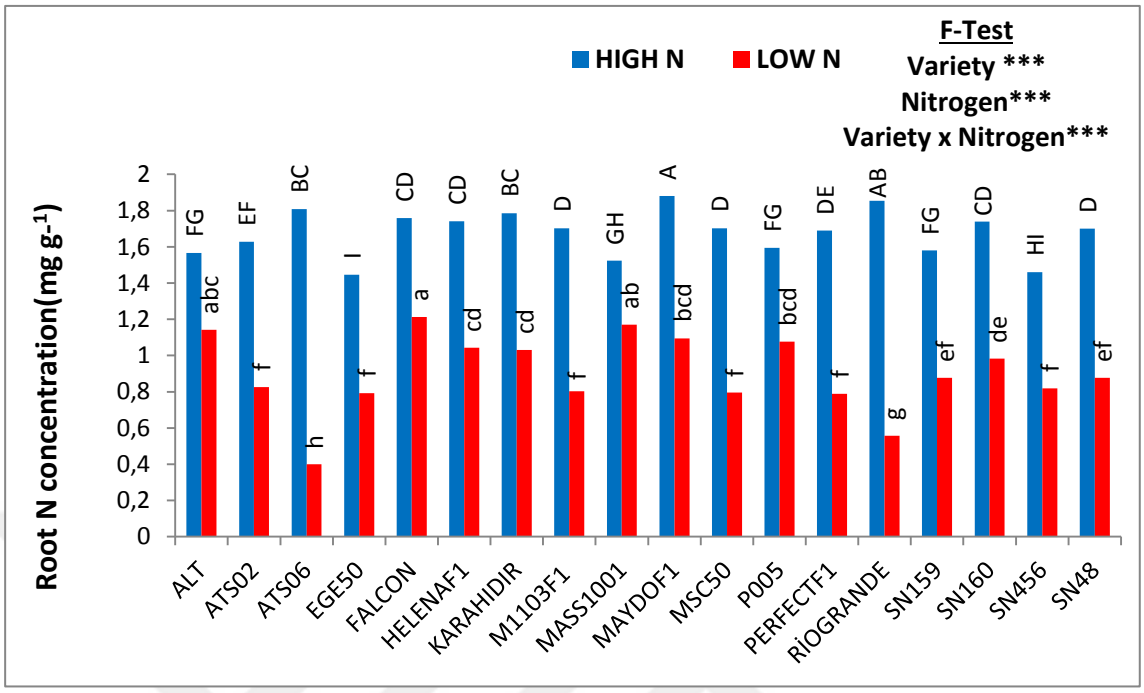


Figure 4.11. .Root N concentration

4.1.7. Shoot and Root total N content

From figure 4.14 and 4.15 it can be seen that shoot and root N uptake (mg g^{-1}) varied significantly ($P < 0.001$) among the varieties (Fig. 4.12 and Fig. 4.13). Also N uptake by shoot and root of various plants were significantly ($P < 0.001$) affected by N rates used for the experiment. Also there exists significant interaction ($P < 0.001$) between nitrogen supply and the varieties. Generally the shoot N uptake was higher than the root N uptake. Under high N rate *Riogrande, ALT* and *HelenaF1* recorded the highest shoot N uptake whilst *ATS02, Falcon, P005* and *Karahidir* recorded the lowest shoot N uptake. Under low N rate *M1103F1, MaydoF1* and *Riogrande* recorded the highest shoot N uptake whilst *HelenaF1, P005* and *Mass1001* recorded the lowest shoot N uptake. *M1103F1, PerfectF1, Riogrande* and *ALT* recorded the highest root N uptake under high N rate whilst *ATS06, P005, MASS1001, Falcon* and *Karahidir* recorded the lowest N uptake under High N rate. Whilst under low N rate *Falcon, P005, ALT, MaydoF1* and *HelenF1* recorded the highest root N uptake whilst *ATS06, Karahidir, MSC50* and *EGE50* recorded the lowest root N uptake under Low N rate.

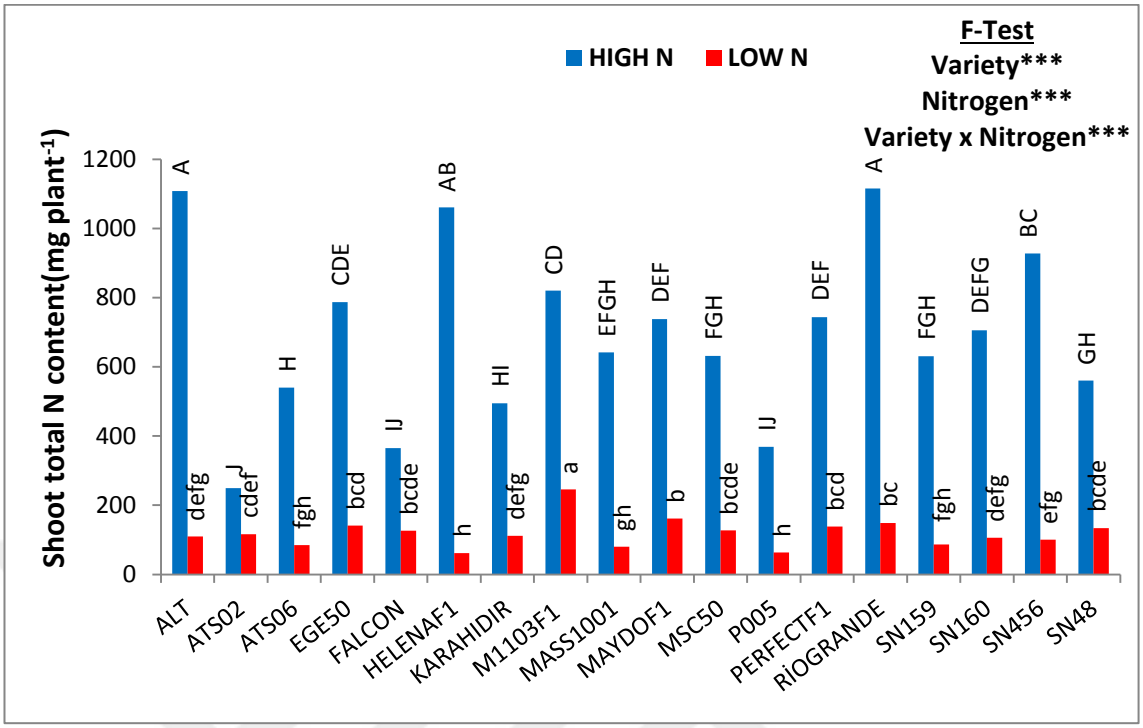


Figure 4.12. Shoot total N content

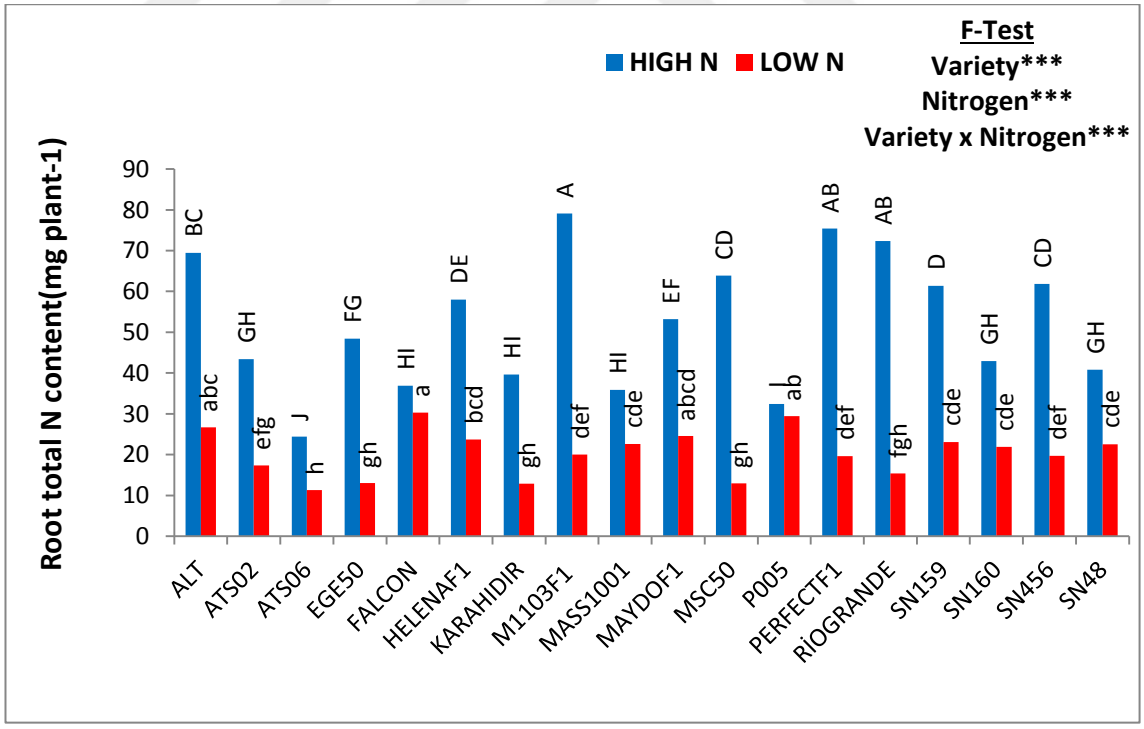


Figure 4.13. Root total N content

4.1.8. Leaf Chlorophyll content (SPAD)

In leaf, the green pigment called chlorophyll has the ability to convert light energy into chemical energy by method of photosynthesis (Fig. 4.14). The output of photosynthesis rate is greatly influence by activities of leaf chlorophyll content and light intensity. With respect to 18 genotypes used for the experiment shows, that SPAD values were significantly different ($P < 0.001$) among these varieties. N rates also significantly ($P < 0.001$) influenced leaf SPAD. There was also a significant difference ($P < 0.001$) between varieties and nitrogen rates. Under high N rate *MaydoF1, Karahidir, HelenaF1* and *SN48* recorded the highest SPAD value whilst *P005, ALT, SN456 a*nd *Riogrande* recorded the lowest SPAD value. Under low N rate *Karahidir, ATS02, HelenaF1* and *SN48* recorded the highest SPAD value whilst *P005, ALT, Riogrande* and *EGE50* recorded the lowest SPAD value. Generally out of 18 varieties used for the experiments, the SPAD values of 13 varieties under high N rate was greater than that of low N rate except *ALT, ATS02, Karahidir, ATS06, SN159* and *SN456* where the low N rate was greater than high N rate.

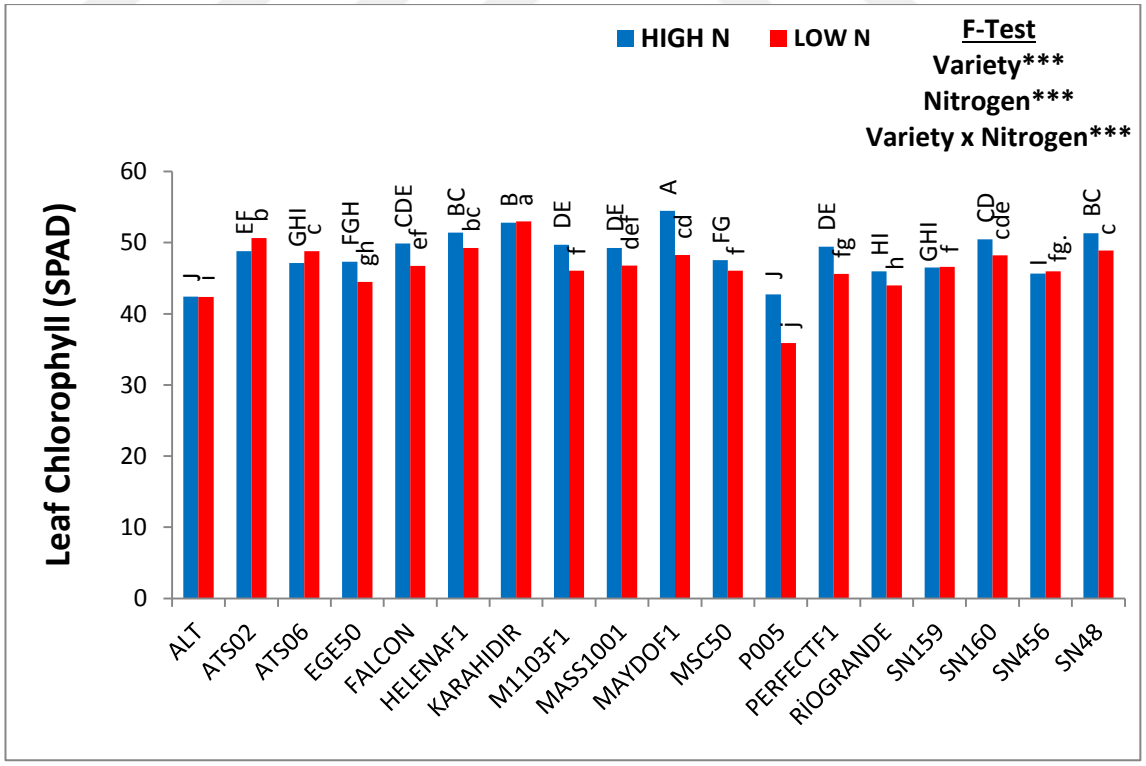


Figure 4.14. Leaf chlorophyll (SPAD). Means with the same letter are not significantly different

4.1.9. Leaf Total Chlorophyll and Carotenoid Content (spectrometric readings)

From the spectrometer reading and calculations of leaf total chlorophyll and carotenoid content (mg/g) revealed that there is a significant different ($P < 0.001$) among the varieties. N rates also significantly ($P < 0.001$) affected total chlorophyll and carotenoid content. Significant interaction ($P < 0.001$) also existed between nitrogen rates and the varieties. As seen in Figure 4.17 and 4.18 respectively below, all the varieties under high N rate both in total chlorophyll and carotenoid content was greater than that of low N rate except *karahidir, Mass1001, SN160* in total chlorophyll and *SN48, SN456, perfectF1* and *HelenaF1* in total carotenoid. Under high N rate *ATS02, MaydoF1, Falcon* and *HelenaF1* recorded the highest leaf total chlorophyll whilst *ALT, SN456 MSC50* and *P005* recorded lowest. Under low N rate *ATS02, Karahidir, EGE50* and *Mass1001* recorded the highest total chlorophyll content whilst *ALT, SN456, P005* and *MSC50* recorded the lowest total chlorophyll content. With respect to total carotenoid content *Riogrande, M1103F1, Karahidir* and *MSC50* recorded the highest under high N whilst *Falcon, ALT, SN48* and *SN456* recorded the lowest under high N rate. Under low N rate *SN48, Riogrande, PerfectF1* and *SN456* recorded highest leaf carotenoid content respectively *whilst P005, ALT, ATS06* and Falcon recorded the lowest leaf carotenoid content.

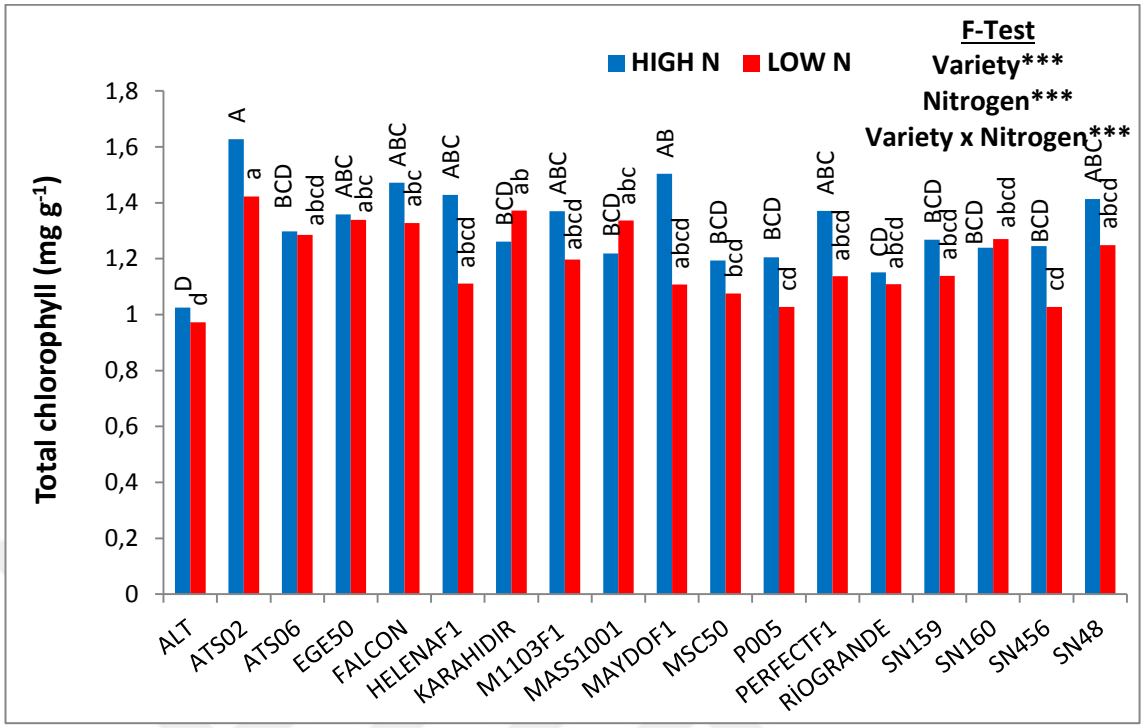


Figure 4.15. Leaf total chlorophyll content. Means with the same letter are not significantly different

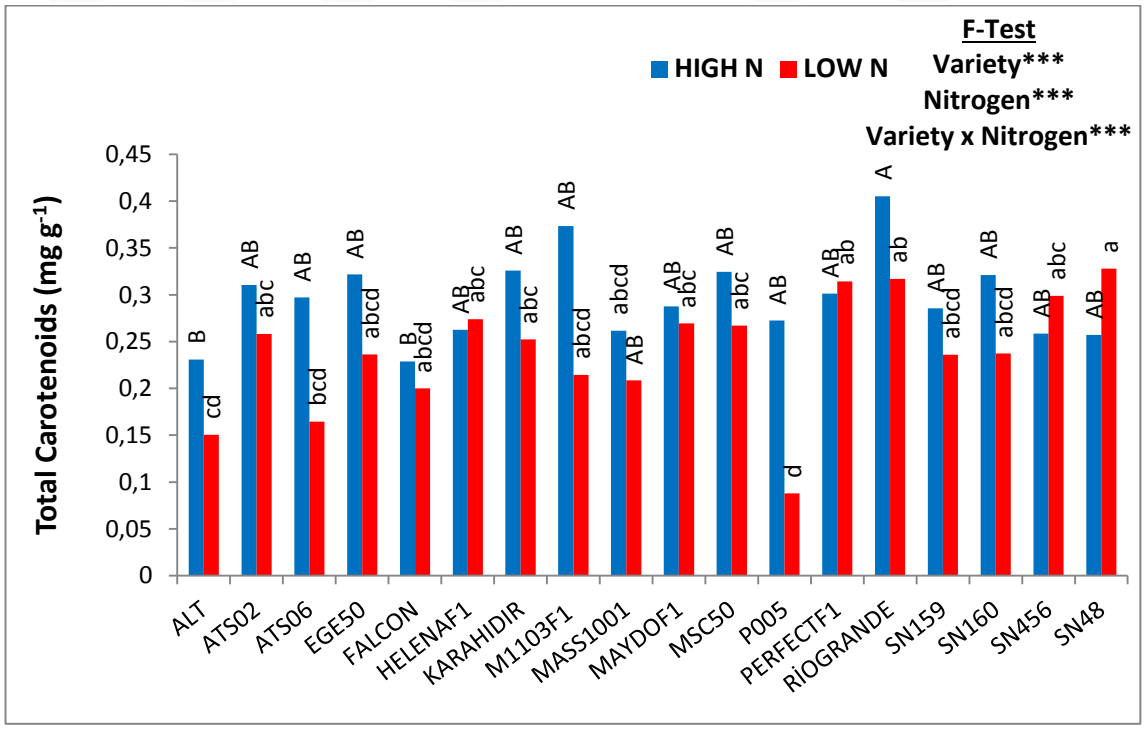


Figure 4.16. Total carotenoid content. Means with the same letter are not significantly different.

4.1.10. Photosynthesis

Photosynthesis rate was significantly different ($P < 0.001$) among varieties and nitrogen rates (Fig. 4.17). There was also significant interaction ($P < 0.001$) between Nitrogen rates and the varieties. Under high N *rate HelenaF1, Riogrande, ATS02* and *SN160* recorded the highest photosynthetic activities whilst *P005, PerfectF1, MSC50* and *Mass1001* recorded the lowest photosynthetic activities. However under low N rate *HelenaF1, Riogrande, Karahidir* and *ATS06* recorded the highest photosynthetic activities whilst *P005, ALT, SN456* and *MSC50* recorded the lowest photosynthetic activities

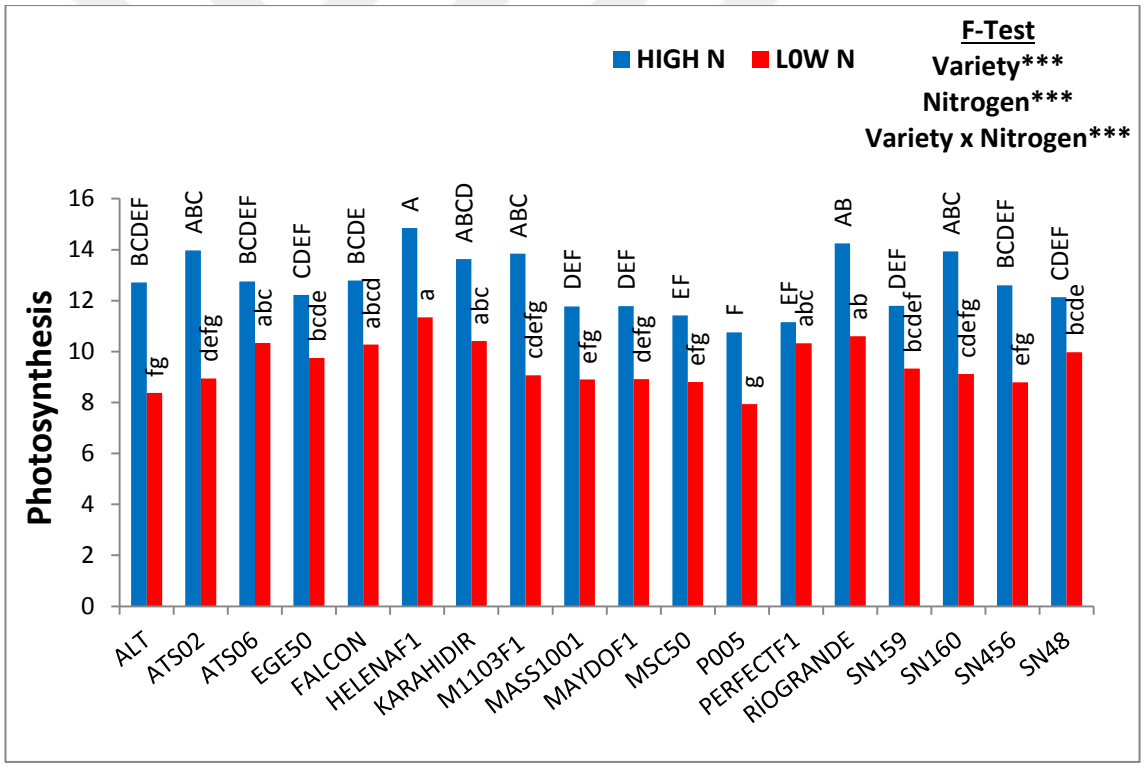


Figure 4.17. Photosynthetic activity. Means with the same letter are not significantly different.

4.1.11. Plant Height

Stunted growth is known to be one symptoms of N deficiency in plants (Fig. 4.18). Due to this fact plants height measured saw significant differences ($P < 0.001$) among varieties and nitrogen rates. There was also significant interaction ($P < 0.001$) between nitrogen rates and the varieties. Under high N rate *MaydoF1, HelenaF1, P005* and *ALT* recorded the tallest plant height whilst *SN48, EGE50, ATS02* and *SN160* recorded the shortest plant height. However under low N rate *MaydoF1, HelenaF1, P005, Mass1001* and *ALT* recorded the tallest plant height whilst *SN160, SN48, SN456* and *EGE50* recorded the shortest plant height.

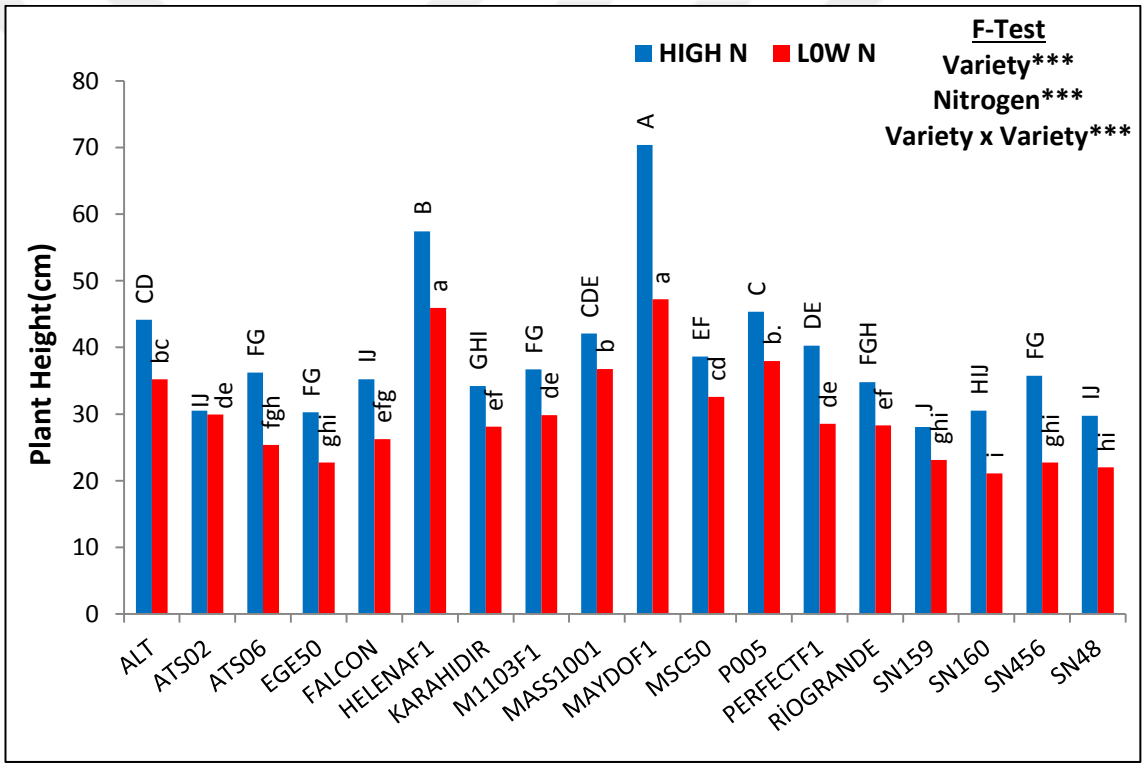


Figure 4.18. Plant height. Means with the same letter are not significantly different.

4.2 Results of Experiment II

4.2.1. Shoot Biomass Production

Results of fresh and dry shoot weight indicate that, there were significant differences ($P < 0.001$) among grafted combinations (Fig. 4.19 and Fig. 4.20). Also nitrogen rates significantly ($P < 0.001$) affected fresh and dry shoot weight of various grafted combinations. In addition, interaction between nitrogen supply and the grafted combinations shows to be significant ($P < 0.001$). From figure 4.19 indicates that *karahidir* and *ALT* grafted combination (S1/R2), *P005* and Helena combination (S2/R1), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT c*ombination (S2/R2) recorded the highest fresh shoot weight respectively under high N rate whilst *P005* (S2) and *Karahidir* (S1) also recorded the lowest fresh shoot weight respectively under high N rate. However under low N rate *P005* and *Helena* (S2/R1), *karahidir* and *ALT* combination (S1/R2), *P005* and *ALT* combination (S2/R2), *Karahidir* and *Helena* combination (S1/R1) respectively recorded the highest fresh shoot weight respectively whilst *P005* (S2) and *Karahidir* (S1) recorded the lowest fresh shoot weight under low N rate. Results of dry shoot weight also shows that *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *P005* and *ALT* combination (S2/R2) recorded the highest dry shoot weight respectively under high N rate whilst *P005* (S2) and *Karahidir* (S1) recorded the lowest dry shoot weight respectively under high N rate. However under low N rate *P005* and *Helena* combination (S2/R1), *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2) recorded the highest dry shoot weight respectively under low N rate whilst *P005* (S2) and *Karahidir* (S1) recorded the lowest dry shoot weight respectively under low N rate.

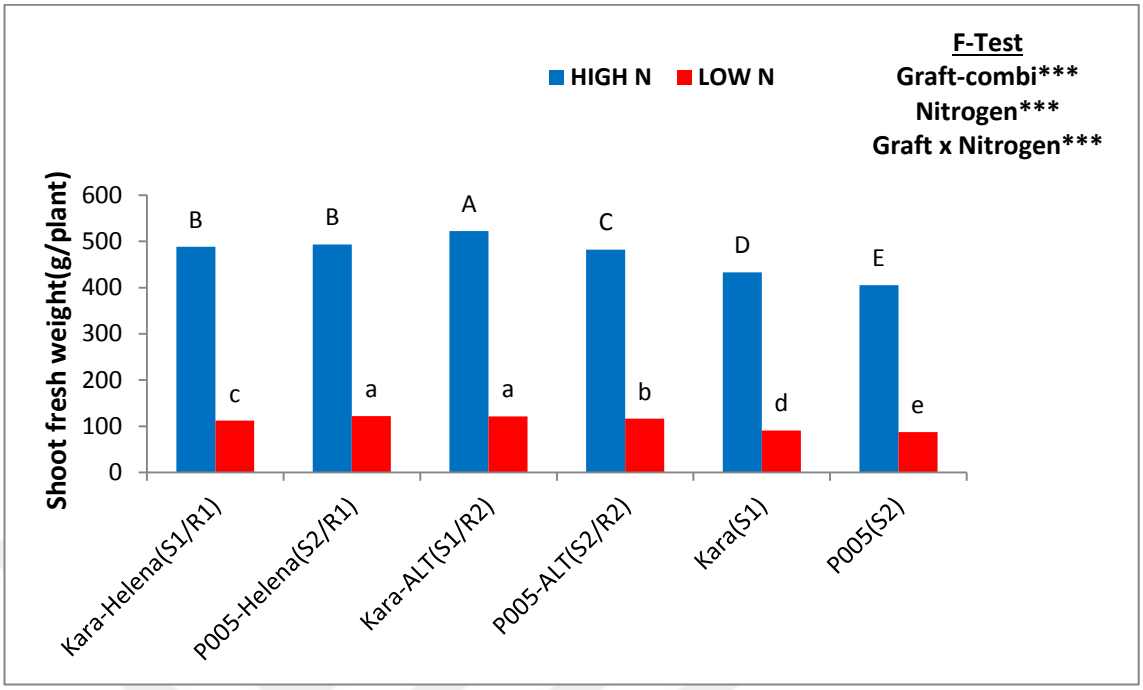


Figure 4.19. Shoot fresh weight of grafted and non-grafted tomato varieties under high and low N rates.

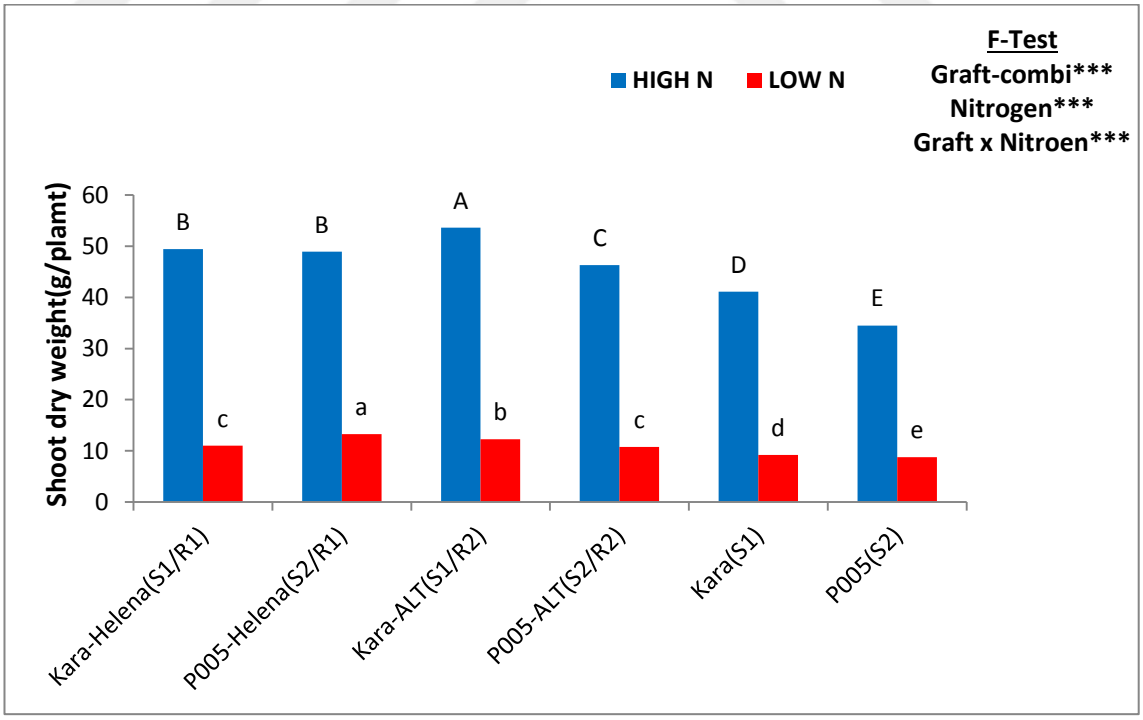


Figure 4.20. Shoot dry weight of grafted and non-grafted varieties under high and low N rates.

4.2.2. Root Biomass Production

Results of fresh root weight also indicates that *P005* and *ALT* grafting combination (S2/R2), *Karahidir* and *ALT* combination (S1/R2), *P005* and *Helena* (S2/R1), *Karahidir* and *Helena* combination (S1/R1) produced the highest fresh root biomass respectively under high N rate whilst *Karahidir* (S1) and *P005* (S2) also recorded the lowest fresh root biomass respectively under high N rate (Fig. 4.21). Nevertheless under low N rate *P005* and *ALT* combination (S2/R2), *Karahidir* and *Helena* combination (S1/R1), *karahidir* and *ALT* combination (S1/R2), *P005* and *Helena* (S2/R1) recorded the highest fresh root biomass whilst *P005* (S2) and *Karahidir* (S1) recorded the lowest fresh root weight under low N rate.

In addition, *P005* and *ALT* combination (S2/R2), *karahidir* and *ALT* combination (S1/R2), *P005* and *Helena* (S2/R1), *Karahidir* and *Helena* combination (S1/R1) recorded the highest dry root weight respectively under high N rate whilst *Karahidir* (S1) and *P005* (S2) recorded the lowest dry root weight respectively under high N rate (Fig. 4.22). However under low N rate, *P005* and *ALT* combination (S2/R2), *Karahidir* and *Helena* combination (S1/R1), *karahidir* and *ALT* combination (S1/R2), *P005* and *Helena* (S2/R1) recorded the highest dry root weight respectively under low N rate whilst *Karahidir* (S1) and *P005* (S2) also recorded the lowest dry root weight respectively under low N rate. With regards to fresh and dry root weight there were significant differences ($P < 0.001$) among grafted combinations and also there was significant difference ($P < 0.001$) among N rate as well as interaction between grafted combinations and nitrogen supply.

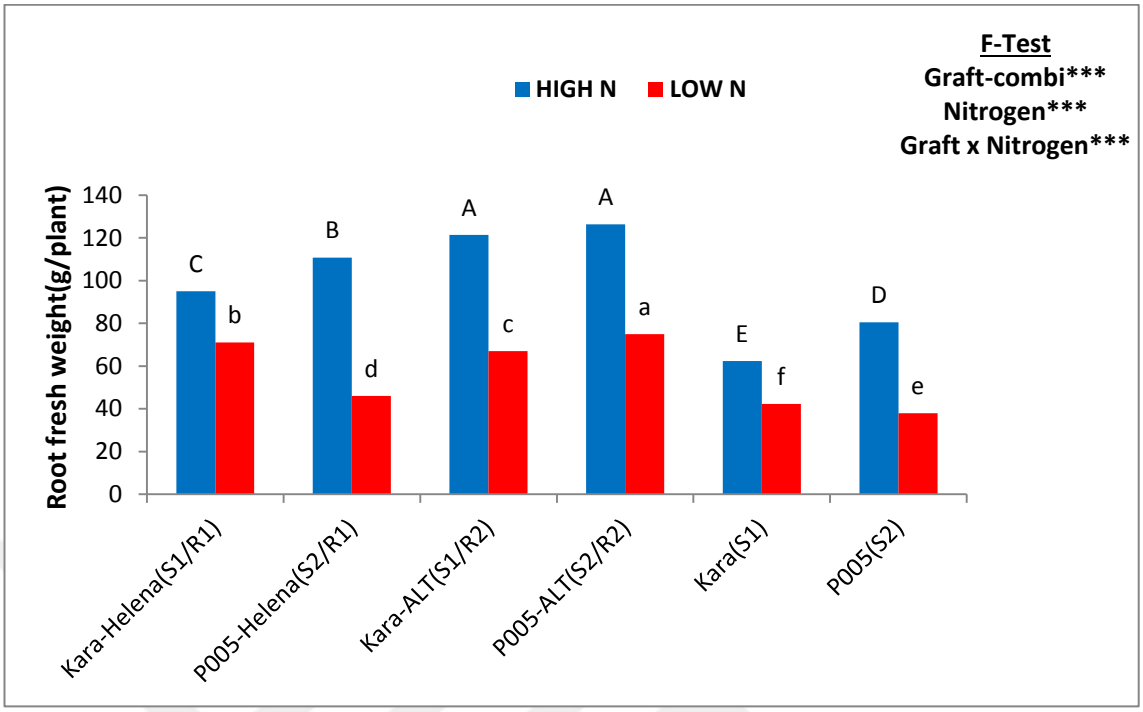


Figure 4.21. Root fresh weight of grafted and non-grafted varieties under high and low N rates.

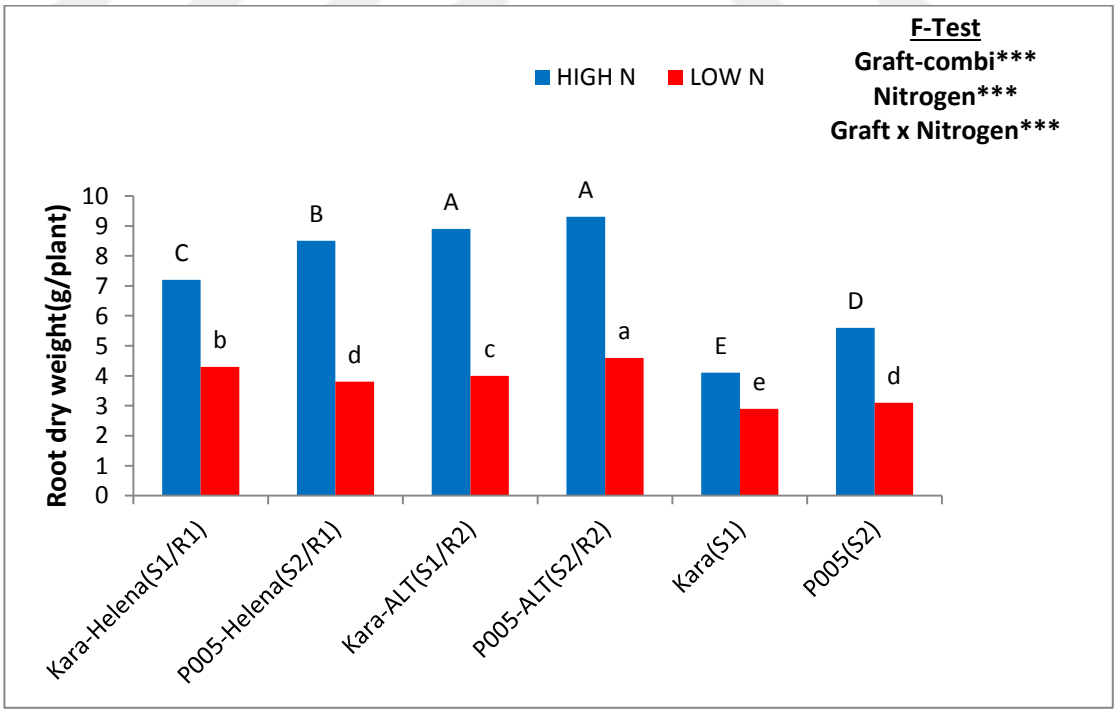


Figure 4.22. Root dry weight of grafted and non-grafted varieties under high and low N rates

4.2.3. Leaf area

Results for the Leaf area analysis indicate that there was significant difference among grafted combinations ($P < 0.001$); also the effect of N rate on leaf area among various grafted combinations was significantly different ($P < 0.001$) (Fig. 4.23). In addition, the interaction between N rate and grafted combinations with respect to leaf area was significant ($P < 0.001$). From Figure 4.23 indicates that, the grafting combination of *karahidir* and *ALT* (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *P005* and *ALT* combination (S2/R2) respectively recorded highest leaf area under high N rate whilst *P005* (S2) and *Karahidir* (S1) recorded the lowest leaf area respectively under high N rate. However under low N rate *P005* and *Helena* combination (S2/R1), *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2), respectively recorded the highest leaf area whilst *P005* (S2) and *Karahidir* (S1) respectively recorded the lowest leaf area under low N rate.

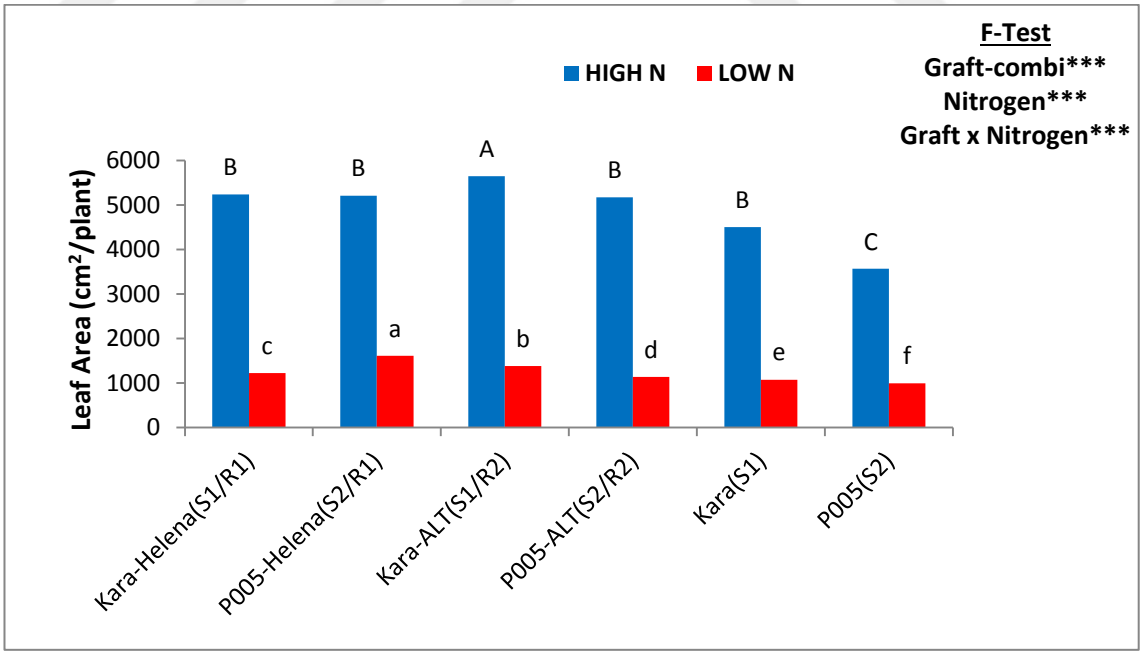


Figure 4.23. The effect of N levels on total leaf area

4.2.4. Root length, volume and diameter

Figure 4.24 indicates that *karahidir* and *ALT* combination (S1/R2), *P005* and *ALT* combination (S2/R2), *P005* and *Helena* combination (S2/R1), *Karahidir* and *Helena* grafting combination (S1/R1) recorded the highest root length whilst *Karahidir* (S1) and *P005* (S2) recorded the lowest root length under high N rate. However under low N rate *P005* and *ALT* combination (S2/R2), *Karahidir* and *Helena* combination (S1/R1), *karahidir* and *ALT* combination (S1/R2), *P005* and *Helena* (S2/R1) recorded the highest root length respectively whilst *Karahidir* (S1) and *P005* (S2) recorded the lowest root length respectively under low N rate. Results of root volume also indicates that *Karahidir* and *ALT* combination (S1/R2), *P005* and *ALT* combination (S2/R2), *Karahidir* and *Helena* combination (S2/R1), *P005* and *Helena* combination (S2/R1) recorded the largest root volume respectively under high N rate whilst *Karahidir* (S1) and *P005* (S2) also recorded the lowest root volume respectively under high N rate (Fig. 4.25). However under low N rate *P005* and *Helena* combination (S2/R1), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2), *karahidir* and *ALT* combination (S1/R2) recorded the highest root volume respectively under low N rate whilst *Karahidir* (S1) and *P005* (S2) recorded the lowest root volume respectively under low N rate. In addition *P005* (S2), *P005* and *ALT* combination (S2/R2), *P005* and *Helena* combination (S2/R1), *Karahidir* and *Helena* combination (S1/R1) recorded the highest root diameter respectively under high N rate whilst *Karahidir* (S1), *karahidir* and *ALT* combination (S1/R2) recorded the lowest root diameter respectively under high N rate. However under low N rate *Karahidir* (S1), *P005* (S2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *P005* and ALT combination (S2/R2) recorded the highest root diameter respectively under low N rate whilst *karahidir* and *ALT* combination (S1/R2) also recoded lowest root diameter respectively under low N rate. Root diameter varied significantly ($P < 0.001$) among grafted combinations (Fig. 4.26). Also the effect of N rate on root diameter was highly significant. In addition there was significant difference ($P < 0.001$) among grafted combinations and nitrogen supply interactions.

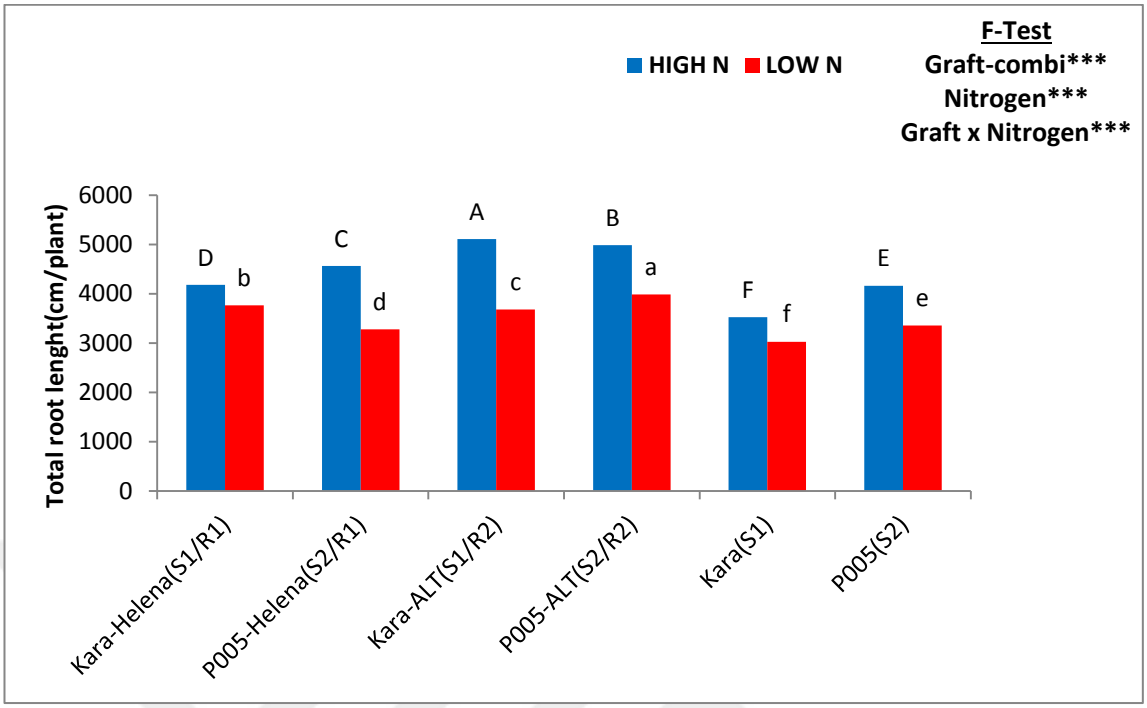


Figure 4.24. Root length. Means with same letter are not significantly different.

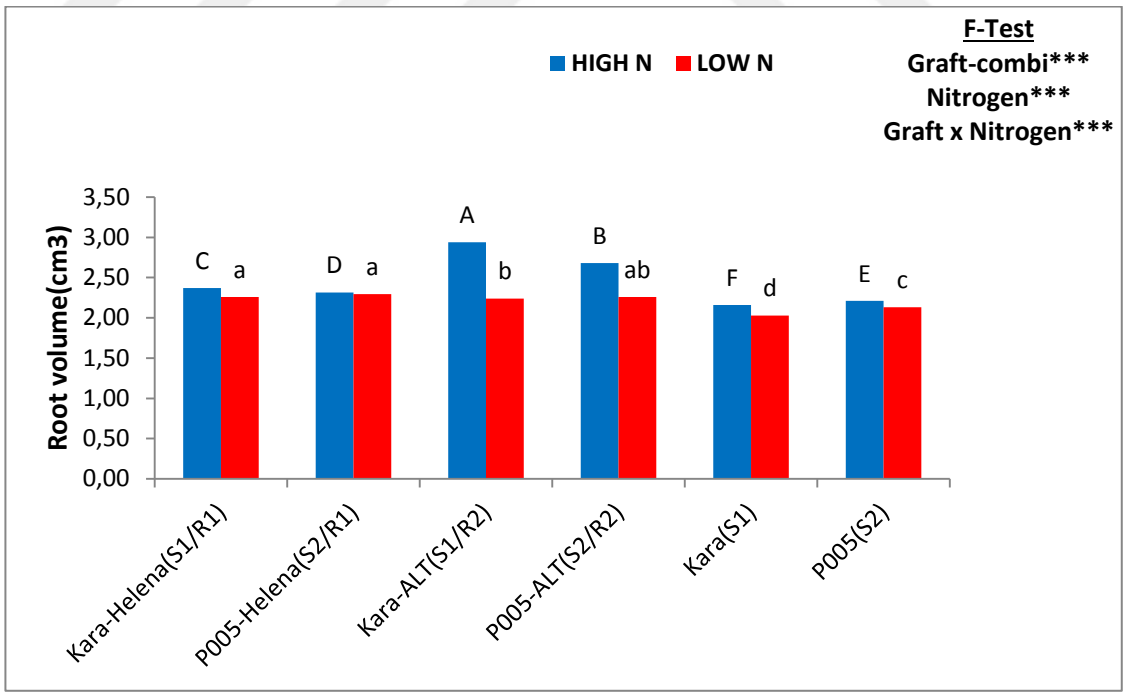


Figure 4.25. Root Volume. Means with same letter are not significantly different.

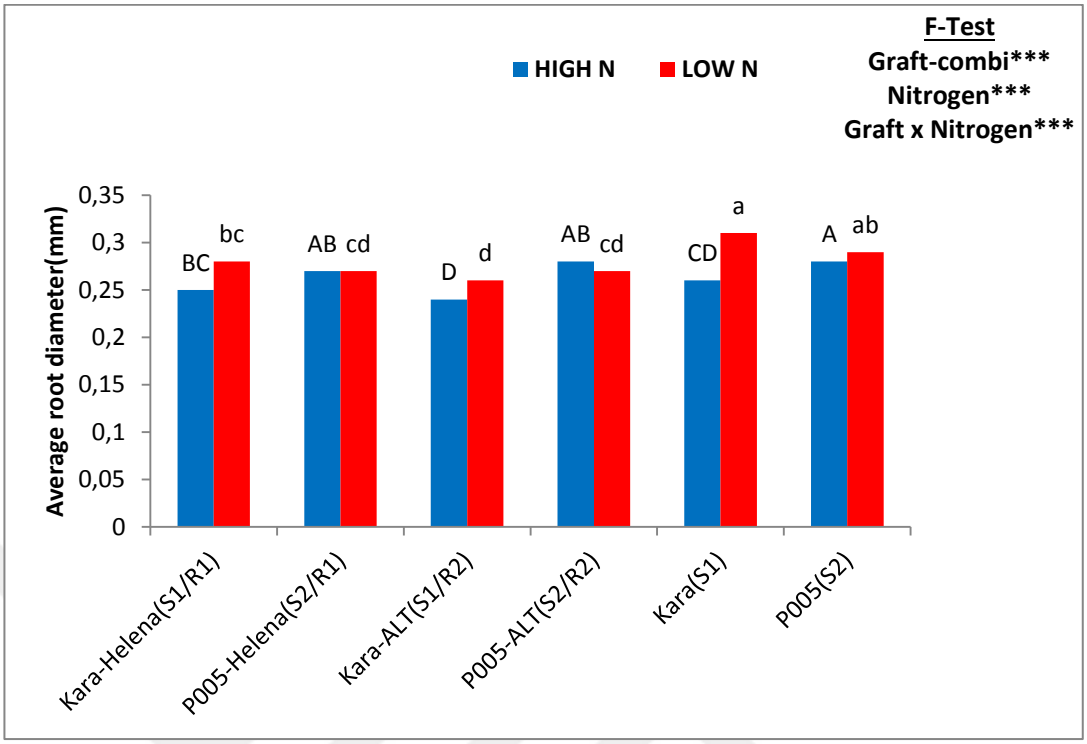


Figure 4.26. Root diameter. Means with same letter are not significantly different.

4.2.5. Nitrate Reductase Activity

Results of NR activity indicates that *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *karahidir* and *ALT* combination (S1/R2), *P005* and *ALT* combination (S2/R2) recorded highest NR activity respectively whilst and *Karahidir* (S1) and *P005* (S2) also recorded the lowest NR activity respectively under high N rate (4.27). However under low N rate *P005* and *ALT* combination (S2/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *karahidir* and *ALT* combination (S1/R2) recorded the highest NR activity whilst *Karahidir* (S1) and *P005* (S2) recorded lowest NR activity respectively. Results of various grafted combinations used for this research shows significant differences ($P < 0.001$) in NR activity. Nitrogen rates also significantly ($P < 0.001$) affected the NR activity. However there was significant interaction ($P < 0.001$) between nitrogen supply and grafted combinations.

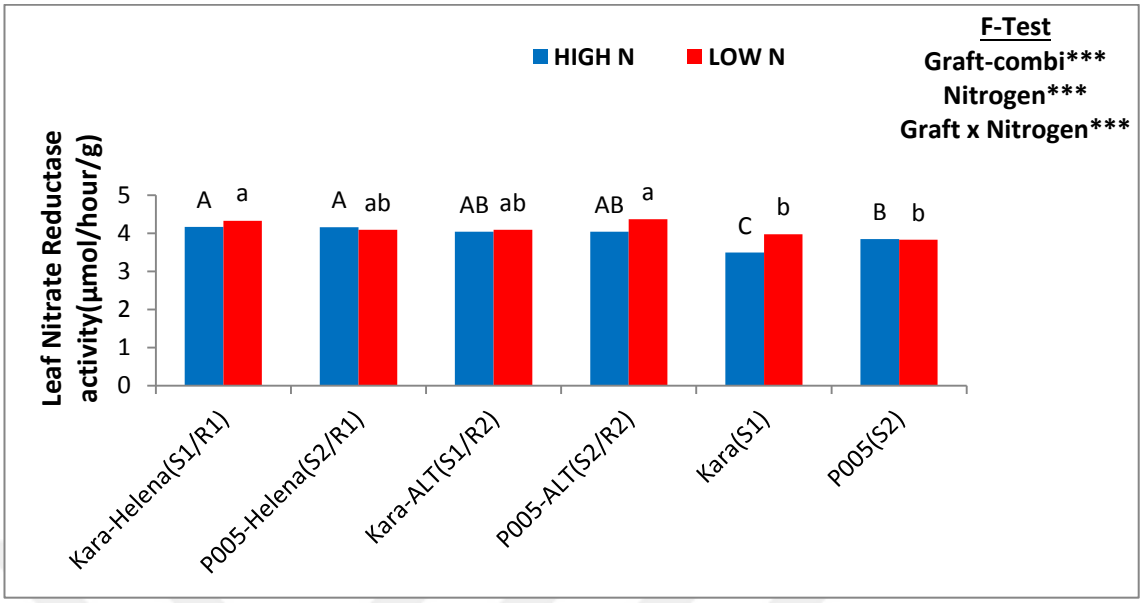


Figure 4.27. Leaf nitrate reductase activity rate.

4.2.6. Shoot and root nitrogen concentration

Results of nitrogen concentration in shoot indicates that *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2) and *Karahidir* (S1) recorded the highest N concentration respectively under high N rate whilst *P005* and *Helena* combination (S2/R1), *P005* (S2) also recorded the lowest N concentration under high N rate (Fig. 4.28). However under low N rate *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *Karahidir* (S1), *P005* and ALT combination (S2/R2) recorded the highest N concentration respectively in shoot whilst *P005* (S2) also recorded the lowest N concentration in shoot. Nevertheless results of N concentration in roots indicates that *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2), *P005* and *Helena* combination (S2/R1) recorded the highest root N concentration under high N rate respectively whilst *P005* (S2) and *Karahidir* (S1) also recorded the lowest N concentration respectively under high N rate respectively (Fig. 4.29). However under low N rate *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *karahidir* and *ALT* combination (S1/R2), *P005* and *ALT* combination (S2/R2) recoded the high root N concentration whilst *P005* (S2) and *Karahidir* (S1) recorded the lowest respectively under low N rate. The shoot and root N concentration saw

significantly (P < 0.001) among the grafted combinations. Again shoot and root N concentration of various grafted combinations were significantly (P < 0.001) affected by N rates. Significant difference (P < 0.001) also existed between nitrogen supply and grafted combinations interaction of both shoot and root of various plants.

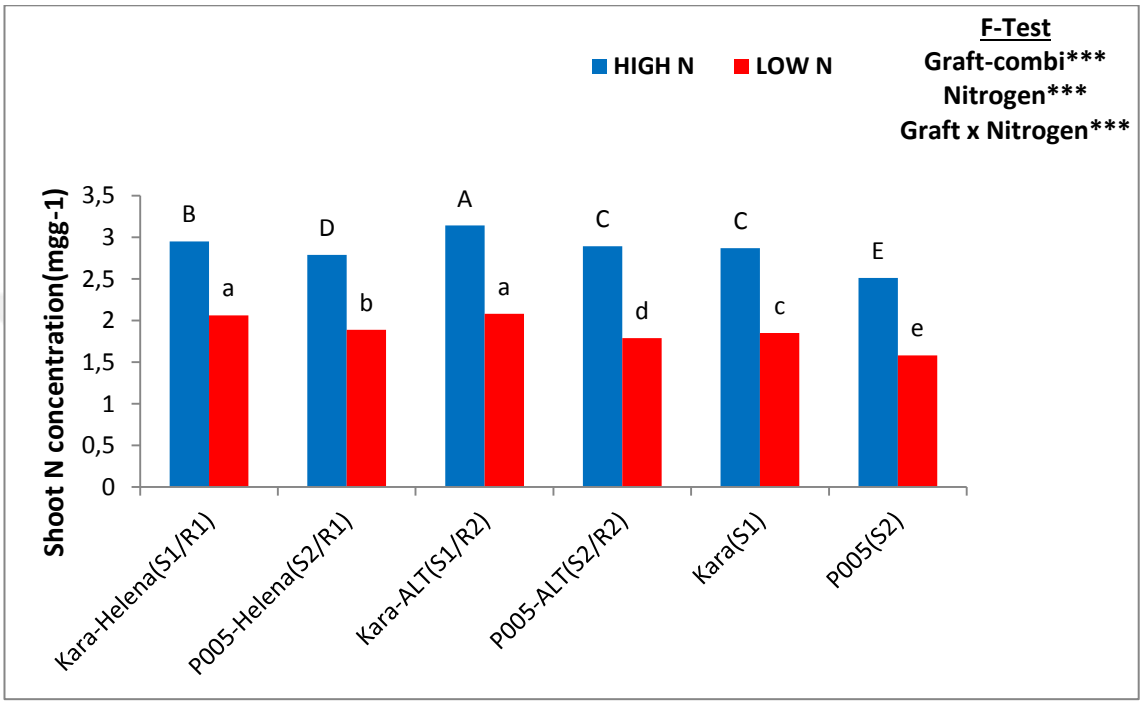


Figure 4.28. Shoot N concentration

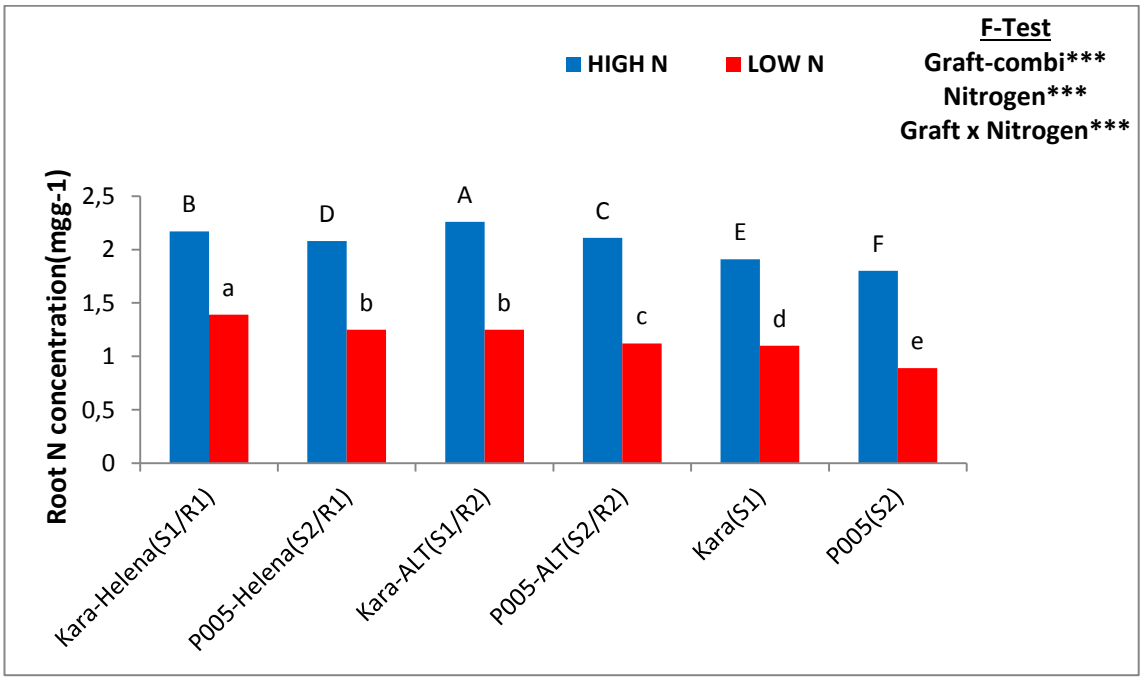


Figure 4.29. Root N concentration

4.2.7. Shoot and Root Total N content

Results of shoot nitrogen uptake indicates that *Karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2), *P005* and *Helena* combination (S2/R1) recorded the highest shoot N uptake respectively under high N rate whilst *Karahidir* (S1) and *P005* (S2) also recorded the lowest shoot N uptake under high N rate (Fig. 4.30). However under low N rate *karahidir* and *ALT* combination (S1/R2), *P005* and *Helena* combination (S2/R1), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2) recorded the high shoot N uptake respectively whilst *Karahidir* (S1) and *P005* (S2) also recorded the lowest shoot N uptake respectively under low N rate. Also the root N uptake also indicates that *P005* and *ALT* combination (RS-S2), *P005* and *Helena* combination (R1-S2), *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1) recorded the highest root N respectively under high N rate whilst *Karahidir* (S1) and *P005* (S2) also recorded the lowest root N uptake respectively under High N rate (Fig. 4.31). However under low N rate *P005* and *ALT* combination (S2/R2), *karahidir* and *ALT* combination (S1/R2), *P005* and *Helena* combination (S2/R1), *P005* (S2), *Karahidir* and *Helena* combination (S1/R1) recorded the highest root N uptake under high N rate whilst *Karahidir* (S1) also recorded the lowest root N uptake under low N rate. Shoot and root N uptake (mg g-1) varied significantly ($P < 0.001$) among the grafted combinations. Also N uptake by shoot and root of various plants were significantly ($P < 0.001$) affected by N rates used for the experiment. Also there exists significant interaction ($P < 0.001$) between nitrogen supply and the grafted combinations.

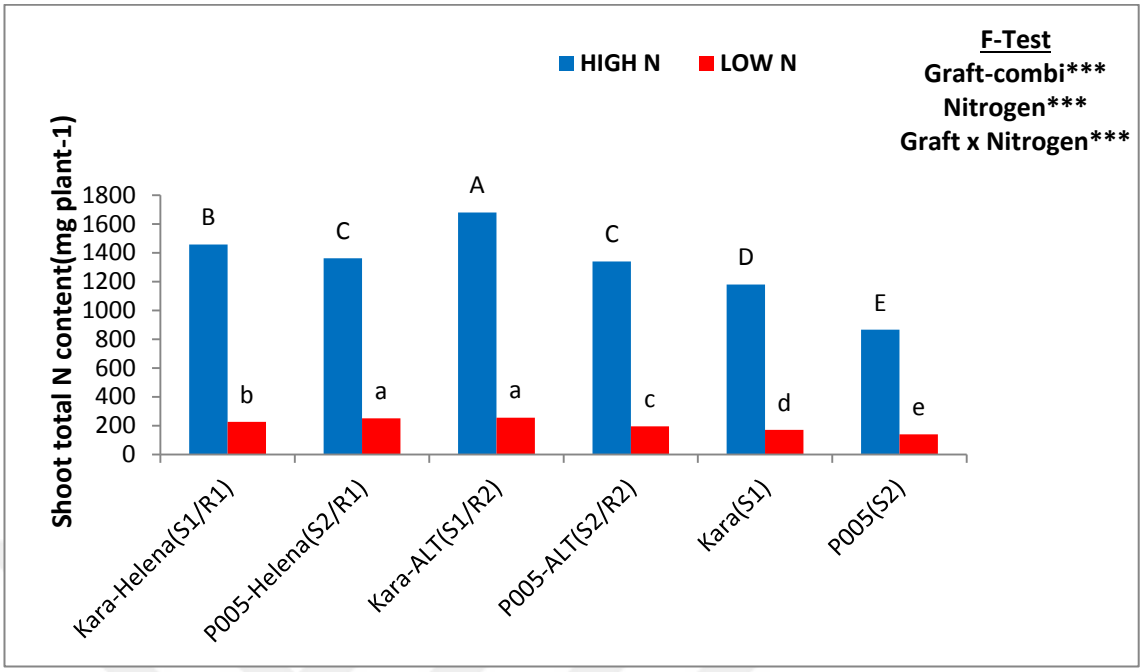


Figure 4.30. Shoot total N content

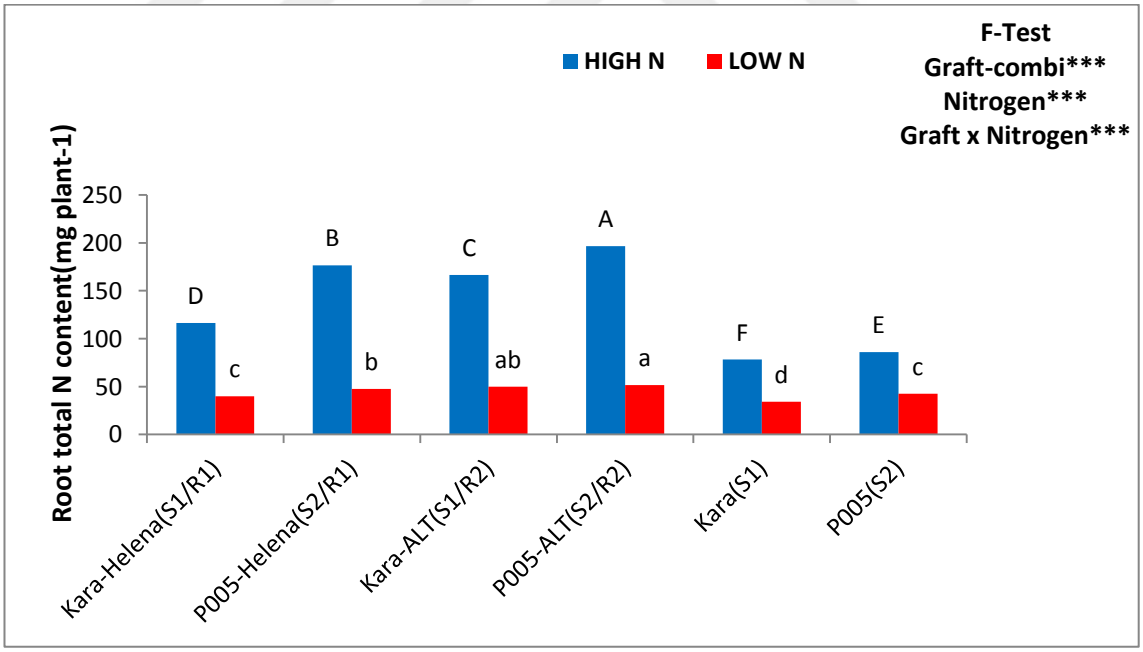


Figure 4.31. Root total N content

4.2.8. Leaf Chlorophyll content (SPAD)

Measurement of the SPAD value indicates that the grafting combination of *Karahidir* and *Helena* (S1/R1) produced the highest SPAD value followed by *karahidir* and *ALT* combination (S1/R2), *Karahidir* (S1), *P005* and *Helena* combination (S2/R1) respectively under high N rate whilst *P005* (S2), *P005* and *ALT* combination (S2/R2) also recorded the lowest under high N rate (Fig. 4.32). However under low N rate *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *Karahidir* (S1) recorded the highest SPAD value respectively whilst *P005* (S2), *P005* and *ALT* combination (S2/R2), *P005* and *Helena* combination (S2/R1) recorded the lowest SPAD value under low N rate. The SPAD value saw a significant different ($P < 0.001$) among grafted combinations and nitrogen rates. There was also significant interaction ($P < 0.001$) between Nitrogen rates and the grafted combinations.

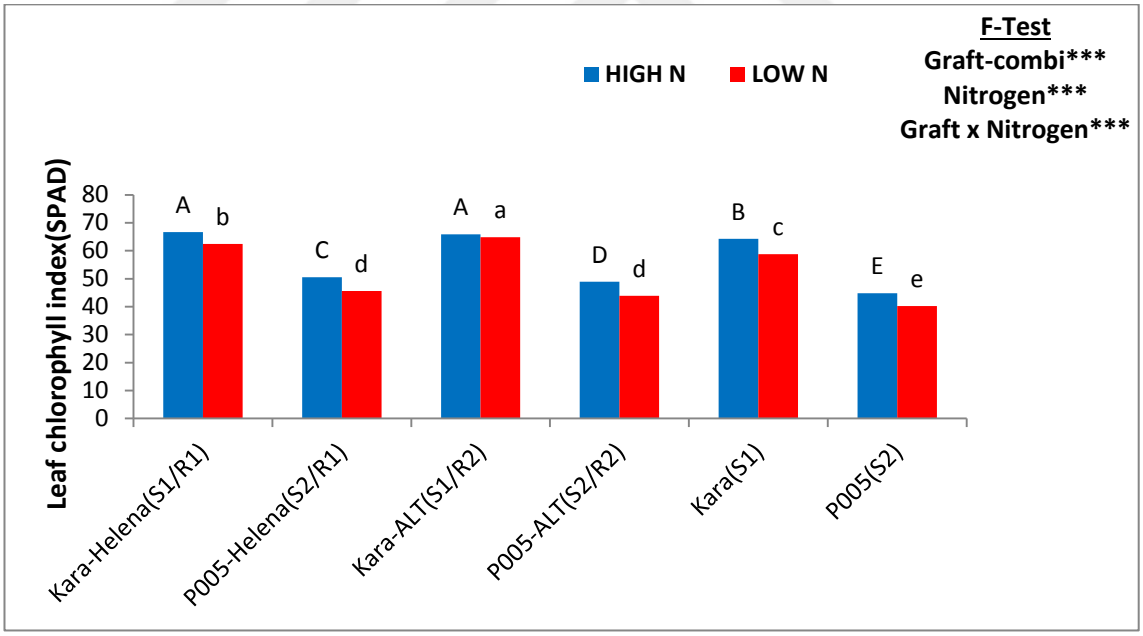


Figure 4.32. Leaf chlorophyll (SPAD). Means with the same letter are not significantly different

4.2.9. Leaf Total Chlorophyll and Carotenoid Content (spectrometric readings)

The spectrometric readings indicated that *karahidir a*nd *ALT* grafting combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *Karahidir* (S1) recorded the highest chlorophyll content respectively under high N rate whilst *P005* (S2) and *P005* and *ALT* combination (S2/R2) recorded the lowest total chlorophyll content respectively under high N rate (Fig. 4.33). However under low N rate *karahidir* and *ALT* grafting combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1), *Karahidir* (S1) recorded the highest total chlorophyll content respectively whilst *P005* (S2), *P005* and *ALT* combination (S2/R2), *P005* and *Helena* combination (S2/R2) recorded the lowest total chlorophyll content respectively. Results of the total carotenoid content also indicates that *karahidir* and *ALT* grafting combination (S1/R2), *Karahidir* and *Helena c*ombination (S1/R1), *P005* and *Helena* combination (S2/R1), *Karahidir* (S1), *P005* and *ALT* combination (S2/R2) recorded highest carotenoid content whilst *P005* (S2) recorded the lowest total carotenoid content under high N rate (Fig. 4.34). However under low N rate *Karahidir* and *Helena* combination (S1/R1), *P005* and *Helena* combination (S2/R1), *karahidir* and *ALT* grafting combination (S1/R2) and *Karahidir* (S1) recorded the highest total carotenoid content whilst *P005* (S2), P005 and *ALT* combination (S2/R2) recorded the lowest total carotenoid content under low N rate. Results of leaf total chlorophyll and carotenoid content (mg/g) revealed that there is a significant different ($P < 0.001$) among the grafted combinations. N rates also significantly ($P < 0.001$) affected total chlorophyll and carotenoid content. Significant interaction ($P < 0.001$) also existed between nitrogen rates and the grafted combinations.

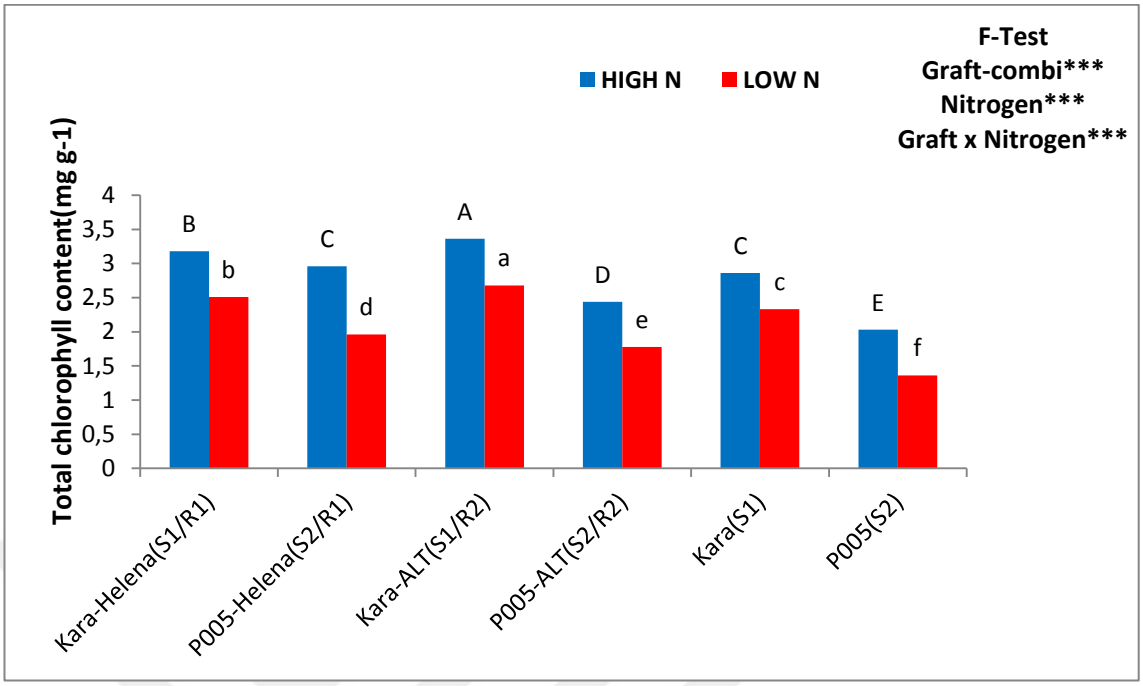


Figure 4.33. Leaf total chlorophyll content. Means with the same letter are not significantly different.

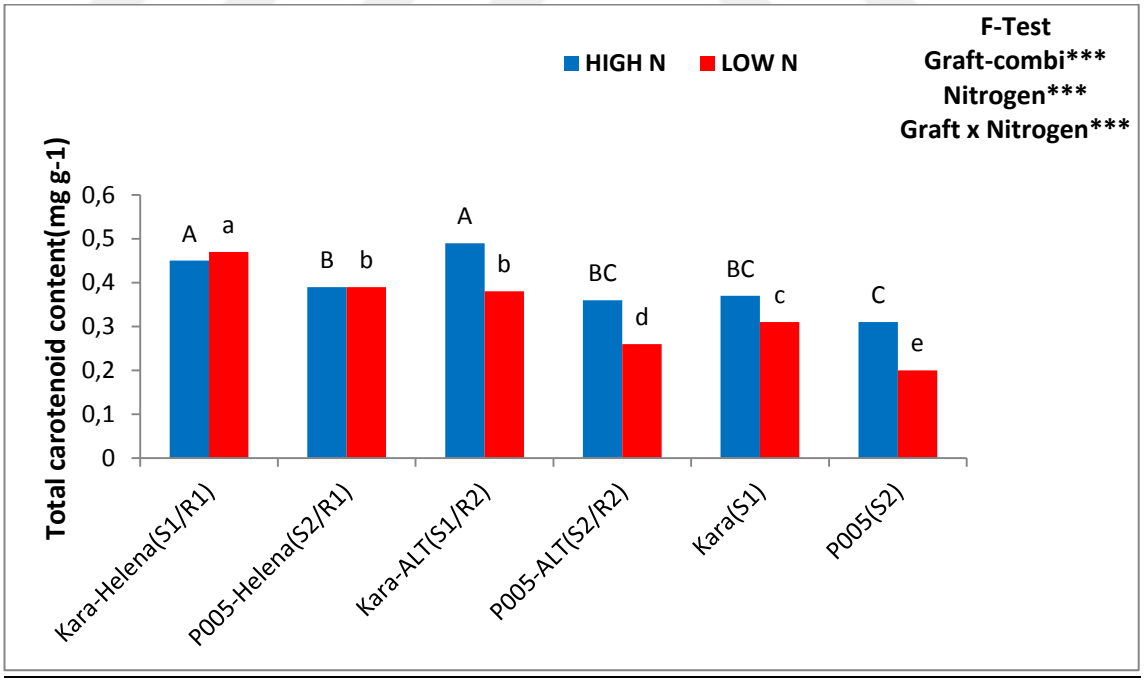


Figure 4.34. Total carotenoid content. Means with the same letter are not significantly different.

4.2.10. Plant Height

The result of plant height was significant among grafted combinations and also the effect of N rate on plant height of various grafted combinations varied significantly ($P < 0.001$) (Fig. 4.35). In addition the interaction between N rate and grafted combinations with respect to plant height saw a significant difference ($P < 0.001$). Measurement of the plant height indicates that the grafting combination *P005* and *Helena* (S2/R1), *P005* (S2), *P005* and *ALT* combination (S2/R2), *karahidir* and *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1) respectively recorded the tallest plant height under high N rate whilst *Karahidir* (S1) recorded shortest plant height also under high N rate. However under low N rate *P005* and *Helena* combination (S2/R1), *P005* and *ALT* combination (S2/R2), *P005* (S2), *karahidir a*nd *ALT* combination (S1/R2), *Karahidir* and *Helena* combination (S1/R1) recorded the tallest plant height under high N rate whilst and *Karahidir* (S1) recorded the shortest plant height respectively.

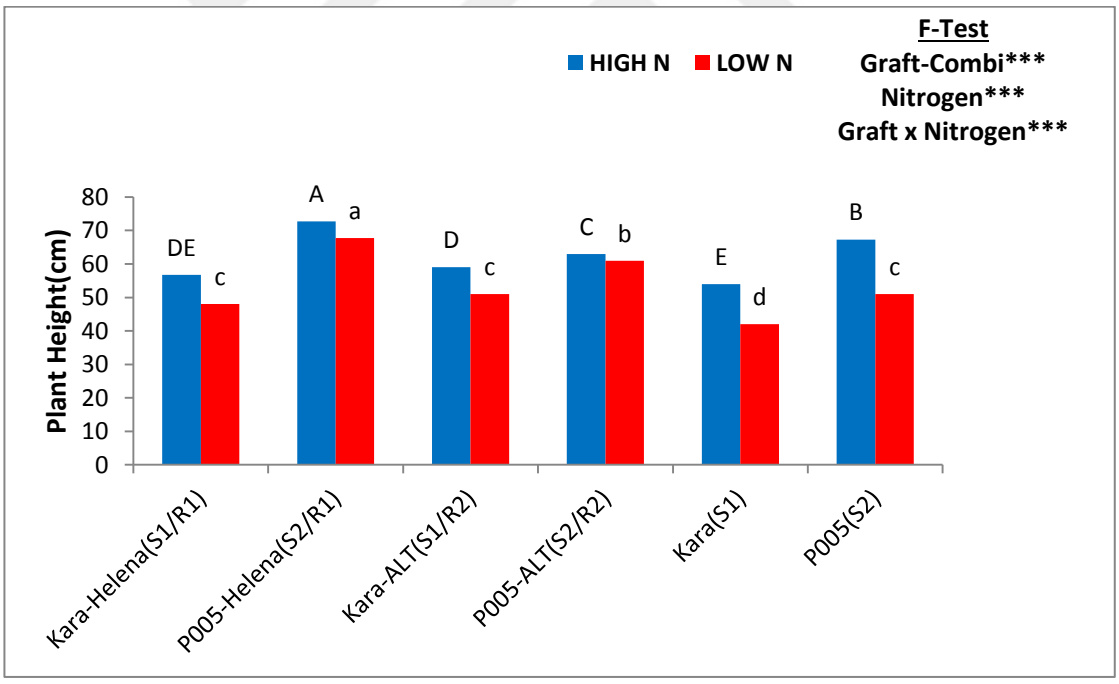


Figure 4.35. Plant height. Means with the same letter are not significantly different.

4.2.11. Photosynthesis

Photosynthesis rate saw a significant difference among grafted combinations and also there was significant difference ($P < 0.001$) among nitrogen rates (Fig. 4.36). Furthermore there was again significant interaction between Nitrogen rates and the grafted combinations. Results of the photosynthetic rate indicate that *Karahidir* and *ALT* grafting combination (S1/R2), *P005* and *Helena* grafted combination (S2/R1), *Karahidir* and *Helena* combination (S1/R1), *P005* and *ALT* combination (S2/R2), *Karahidir* (S1) recorded the highest photosynthetic rate under high N rate respectively. Also *P005* (S2) recorded the lowest photosynthetic rate under high N rate. However under low N rate *Karahidir* and *ALT* grafting combination (S1/R2), *P005* and *Helena* combination (S2/R1), *Karahidir* and *Helena* (S1/R1), *Karahidir* (S1) recorded the highest photosynthetic rate whilst *P005* (S2), *P005* and *ALT* combination (S2/R2) recorded the lowest photosynthetic rate under low N rate respectively.

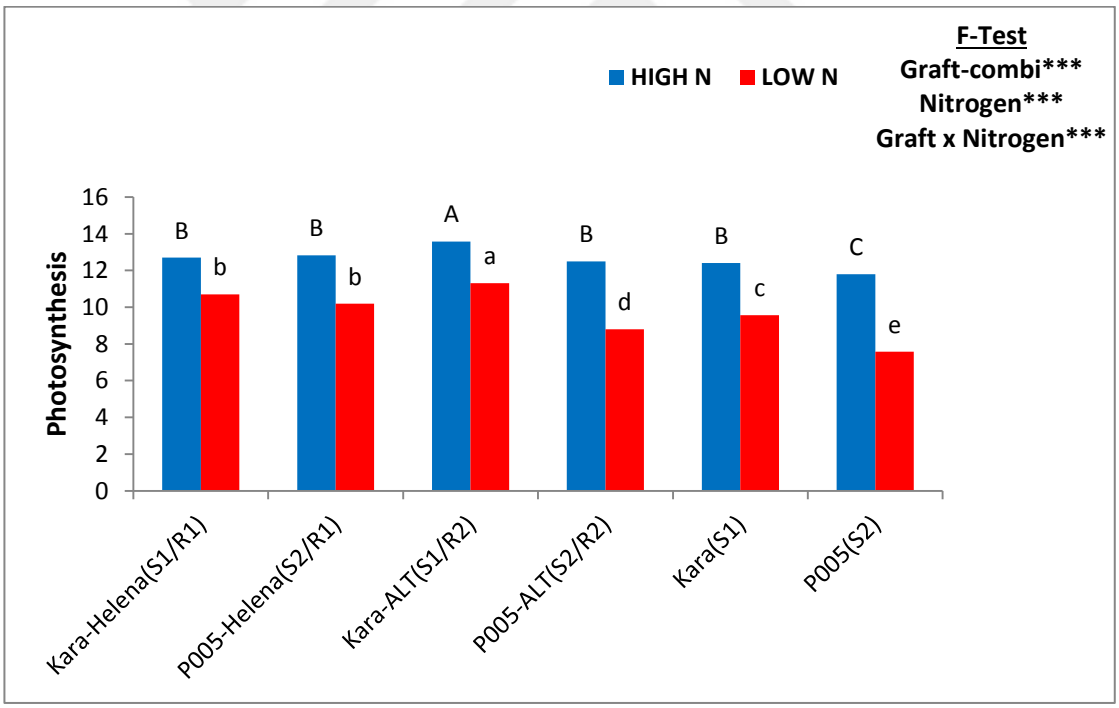


Figure 4.36. Photosynthetic activity. Means with the same letter are not significantly different.

CHAPTER FIVE

DISCUSSION

5.1 Biomass Production

The definition of NUE varies based on preference and objective. However in all contexts the definitions of NUE has similar characteristic in context which is absorbing maximum N by plant for utilization. According to Moll *et al.* (1982) NUE is explained as yield obtained from a crop with regards to amount of N present in the soil or fertilizer. Also according to Lynch (1998) NUE is when the system of a plant has the possibility of transforming taken input into preferable output or in other context reduction of waste during input conversion. Research has shown that increased N supply to crops causes increase in biomass and yield production (Wiedenfeld, 1995; Pandey *et al*., 2001), which simply means that biomass production correlate positive with yield in this regards and so biomass is an important parameter with regards to NUE. In this present research, results of the first experiments indicates that fresh and dry of the genotypes under low and high rate shows significant different among genotypes. Our results correspond with the finding of (Agele *et al*., 2008) from which they observed significant variation in fruit yield and biomass production exist between tomato genotypes with respect to N supply. Also with respect to the first experiment, a positive correlation was found between shoot biomass and root biomass with respect to N rate. This conforms to the finding of Brouwer (1962) from which he observed a linear relation among growth of shoot and root biomass. In addition, the root morphology (root length, volume and diameter) of the first experiment was higher under high N rate than low N rate. This also agrees with finding of by Hebert *et al*. (1992) where they find out amount of N present for plant uptake influenced root biomass production. However with respect to our second results indicates that grafted plants produced much shoot biomass than that of non-grafted plant. This is in agreements with the finding of Ofori

(2015) where he also observed that shoot biomass production was higher in grafted plants than non-grafted plants. Also grafted plants produced large root biomass than not non-grafted plants. Also the root morphology of grafted plants (root length, volume, diameter) was higher than non-grafted plants. Djidonou (2012) also found similar results where total root length, average root diameter, root volume of grafted plants were higher than non-grafted plants and self-grafted plants. Also from our observation we found out shoot and root biomass was enhanced in grafted plants with respect to low N rate. Our results were similar to that of Nawaz *et al.* (2017) where grafting showed enhanced shoot and root biomass production in watermelon and pumpkin grafting combination and also enhanced by nitrogen use efficiency in this grafted combination.

5.2 Total Nitrogen Content and Concentration

According to (Hirel *et al.,* 2007) NUE is explaining as the yield acquired by a crop with respect to N available in a soil or from N fertilizer. NUE involve mechanism in N uptake, translocation, assimilation, redistribution of N efficiently in the plant system. A highly indebt knowledge is needed to understand the genetic mechanism of N uptake and utilization at various stages of plant system (Loudet *et al.,* 2003).The ability of plants to take up N varies genetically which also leads to variation in N concentration. From our first result, indicates variation in N uptake and N concentrate with respect to tomato genotypes were purposely due to differences in ability of genotypes to assimilate and utilize N. Also our first results indicates that N concentration in root under both high and low N correspond positively with the root morphology (root length, root volume and root diameter). This indicates the differences in uptake of N with respect to various tomato genotypes. Our finding was in agreement with observation made by Ruiz and Romero (1998) which they stated that genotypic differences in tomato has impacts on uptake and utilization of N present. The findings of Colla *et al.* (2010) indicate that grafted melon plants have higher N uptake and N utilization than non-grafted melon plants. Similarly the results of our second experiment indicate that N uptake and N concentration in both shoot and root of grafted plants were on the average higher than non-grafted plants under low N rate. Also our second result indicate that under low N rate increase in N uptake and N concentration in grafted plants correspond with increase in root morphology than non-grafted plants. This indicates how rootstock of grafted plant improves in N uptake than non-grafted plants. Our results were in

correspondent to the findings of Nawaz *et al.* (2017) where they observe vigorous root system of pumpkin grafting enhances uptake and accumulation of N in watermelon.

5.3 Photosynthetic activities

In plants, chlorophyll is a green pigment, which is vital as far as photosynthesis is concern; it helps in transforming light energy to chemical energy during the activities of photosynthesis (Şevik, Belkayalı and Aktar 2014).Also the amount of chlorophyll present in a leaf is very paramount to depicts the growth of plants (Bannari *et al.*, 2007). Therefore in crop production chlorophyll is necessary for photosynthetic activities (Ghosh *et al*., 2004).On the other hand, carotenoids aid in light harvesting and also avoiding photo-damage to the photosystem through inter-conversion with xanthophyll molecules (Young et al., 1997; Ort, 2001).With respect to first experiments shows that, N supply at different rates to various genotypes of tomato plants has impact on the leaf SPAD, total chlorophyll, carotenoid content and photosynthetic activities. However, the finding of Zhao *et al.* (2003) indicates that N deficiency causes reduction in leaf chlorophyll concentration which is also related decrease in photosynthesis. Kopsell *et al.* (2007) also found out that increase in N supply in plants also leads to increase in pigmentation of carotenoids. From these two findings indicates that increasing N supply to plants leads increase in chlorophyll, carotenoid and photosynthetic activities which is in conformation to our research. However results of the second experiments also indicate grafted plants on average recorded higher chlorophyll and carotenoid content than non-grafted plants. Our findings were in line with research of Mohammed et al., (2009) where they found grafting enhances chlorophyll and carotenoid content of tomato plants. Also we observed that photosynthetic activities in grafted plants on the average were much greater than non-grafted plant in our second experiments. Also He *et al.* (2003) found similar results which they explain that grafting of tomato plants enhances photosynthetic activities. Nevertheless there are some factors which hinder chlorophyll, carotenoid and photosynthetic activities and they are; extreme temperature, light intensity, water stress, insufficient nutrient supply (Flexas *et al.*, 2004; Chuluun *et al.*, 2014) and also growth stage of leaf, risk of leaf damage and genotypic variation are other factors (Bojović and Stojanović, 2005; Schepers *et al*., 1992). We also observed that increase in total chlorophyll, carotenoid content, leaf SPAD and photosynthetic activities corresponded with increase in biomass. This is in agreement with the findings

of Nasraoui-Hajaji, and Gouia, (2014) whom they stated that decrease in growth of tomato plant (biomass) under N supply is related to inhibition of net photosynthetic activities.

5.4 Leaf Area, Plant Height and Senescence

Increase in Leaf area is an important parameter for plants because it aid in light interception, which is needed for photosynthetic activities and biomass production (Liu *et al.*, 2010). Furthermore Matsuda *et al.* (2011) and Hosoi (2003) observed that in tomato plants leaf area correspond positively with fruit yield. From our first and second experiments, we observed plants under high N rate produces larger leaf area than plant under low N rate. This is in line with research carried by Cardarelli *et al.* (2015) where they found that effect of nitrogen to plant have positive correspondent to leaf area development. Also in both experiments conducted, we observed plants with larger leaf area have corresponding high shoot biomass. This was also in agreements with findings of Natr (1992) who stated that on key parameter that influence biomass production in plants is leaf area developments. Also in both experiment we observed a positive correlation between N rate and plant height. Haque *et al.* (2011) also found similar results where they stated that effect of N rates on plants height of tomato was significant and this was the results of sufficient presence of nitrogen for plant uptake and biomass production. However, leaf senescence leads to reduction of photosynthetic activities in plants (Estrada-Campuzano *et al.,* 2012; Xue *et al.,* 2006). Also leaf senescence also leads to reduction of biomass production and yield in crops (Guo and Gan, 2014). From our first and second experiments, we observed that the plants under low N rate recorded senescence whilst none was recorded in high N rate. Our findings were similar to research conducted by Masclaux-Daubresse *et al.* (2007).

5.5 Nitrate Reductase Activity

In plants nitrate reductase activity (NR activity) is an enzyme which quantify amount of N present in plants and also this enzyme correspond positive with growth and yield (Knobloch, and Tischner, 1989, Nichols *et al.* (1978). Also findings of Kumar *et al.* (2017) indicate that NR activity have positive effect on biomass production. NR activity aid in reducing absorbed nitrate to ammonia in plant system (Aslam *et al.,* 2001).

However, NR activity is regulated by availability of water, temperature, light intensity, amount of nitrate present and level of CO_2 (Beevers and Hageman, 1969). Research has shown that in pepper, cucumber and tomato NR activity is greater in leaves but little or none in roots (Achhireddy *et al.,* 1983, Olday *et al.,* 1976, Teitel *et al.,* 1986). This finding was in conformation with our research work. In our findings with respect to first experiments, we observed that NR activity was present in leaf of plant under both high and low rate but none was present in root of plants. Also with respect to second experiment NR activity was much present in the leaf of graft plants than non-grafted plants in both high and low N rates. There was no NR activity in root of both grafted and non-grafted plants with respect to high and low N rate. This gives us fair knowledge that NR activity in grafted plants is much better than non-graft plants. Our findings was similar to that of (Colla *et al.,* 2011) where they observed that NR activities in grafted plants was higher than non-grafted plants which had positive effect on efficient N uptake and N utilization for shoot biomass production.

5.6 Conclusion and Recommendation

After initial screening 14 local and 4 tomato genotypes in an indoor solution culture experiment in order to identify which genotype has vigorous rootstock and efficient in nitrogen utilization, the results of the first experiments proof *helenaF1* and *ALT* to be much efficient in N uptake and utilization whilst *P005* and *Karahidir* proof to be inefficient in N uptake and utilization. Selection of these efficient and inefficient genotypes was based on parameter discussed above whilst biomass production was the ultimate priority. With respect to the second experiments, grafting was conducted between the selected efficient and inefficient genotypes where efficient genotype was used as rootstock whilst inefficient genotype was use as scion. On the average grafted plants shows to improve NUE than non-grafted plants. With respect to grafted and non-grafted plants used for experiments shows *Karahidir* and *ALT* combination (S1/R1), *Karahidir* and *HelenaF1* combination (S1/R2) proof to be much efficient in N uptake and utilization, which correspond with shoot and root growth compared to other plants. It is there recommends that grafting should be used as tool to improve fruit yield, fruit quality, and shelf life in tomato with respect to NUE.

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CAREER OBJECTIVE

I am looking forward to acquire indebt knowledge in my field of studies from Erciyes University, so that I can contribute to the development of my country.

ACADEMIC QUALIFICATION

Bachelor of Science (Bsc), in Post-Harvest Technology
Kwame Nkrumah University of Science and Technology, Kumasi **(2009-2014)**

West African Senior Certificate Examination (WASSCE)
St. Paul's Senior High School, Hatsukope Aflao 2005-2008

Basic Education Certificate Examination (BECE)
Madina 2 Junior High School, Madina-Accra 2002-2005

WORKING EXPERERIENCE

1. National Service Personnel project farm. (2013-2014)
Key responsibility includes:

- Supervising and management of green house and maize project.

2. Ministry of food and agriculture (Adenta district)
Key responsibility includes:

- Demonstrating new technology to farmers and help in fertilizer subsidy.

WORKSHOPS AND SEMINERS

2012 AGRO-SUMMIT (held at Kumasi)
Training youth for agribusiness, entrepreneurship and creativity.

2014 FOCUS CENTRAL GHANA (held at Accra)
Professional development program on employment skills and career development.

SKILLS AND ABILITIES

Computer Skills: Proficient and experience with Internet Explorer, Firefox Mozilla, Opera, Word, Power Point, Excel and Social Package for Statistical Service.

Leadership and Special Skills: Indebt knowledge in seed production, plant nutrition, soil fertility managements, experimental design. In addition I possess good research skills,

communication and presentation skills; Post harvest management skills, marketing, statistics, sociology, accounting, entrepreneurship and field experimentation.

Abilities: Ability to work in a team as well as individually; Ability to work without supervisor; Ability to work effectively under varied condition and also ability to learn new thing.

HOBBIES

Reading of inspirational books and playing video games**.**

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