1-24-2018

Evaluation of Growth Characteristics, Yield, Marketability and Nitrate Levels of Lettuce (Lactuca sativa L) Cultivars Produced in South Louisiana

William D. Afton
Louisiana State University and Agricultural and Mechanical College, wafton@agcenter.lsu.edu

EVALUATION OF GROWTH CHARACTERISTICS, YIELD, MARKETABILITY AND NITRATE LEVELS OF LETTUCE (*LACTUCA SATIVA*) CULTIVARS PRODUCED IN SOUTH LOUISIANA

A Thesis

Submitted to the Graduate Faculty of the
Louisiana State University and
Agricultural and Mechanical College
in partial fulfillment of the
requirements for the degree of
Master of Science

in

The School of Plant, Environmental and Soil Sciences

by
William Daynne Afton
B.S., Louisiana State University, 2011
May 2018

ACKNOWLEDGMENTS

I would like to express my sincere appreciation for my major professor, Dr. Kathryn “Kiki” Fontenot. I am forever indebted to her for providing me the opportunity to pursue a Master of Science degree in Horticulture. It was through her friendship, guidance, and supervision that helped me to realize my passion to inform everyday people about agriculture and the current industry. I would also like to express my sincere appreciation for my remaining committee members, Dr. Jeff Kuehny and Dr. Carl Motsenbocker. None of this would have been possible without their support and guidance.

In addition to my committee, I would also like to thank others whose help and guidance helped throughout my coursework and research projects. First, I would like to thank my parents, Alan and Marlene Afton for their words of encouragement throughout the graduate school process. I would like to thank all the Burden staff, volunteers, graduate assistants and student workers who helped during the lettuce harvest. I would like to especially thank Keith Lewis, Adley Peltier, Andrew Barker, Mike Steele and Rick Hamilton, Carly Gillett, Reeve LeGendre, Mathew Indest, Mark Wilson, and Julie Nieto. I would also like to thank all the professors and administrative personnel in the JC Miller Hall including: Amy Blanchard, Dr. Charlie Johnson, Dr. Paul Wilson and Dr. Jimmy Boudreaux for all of their advice and encouragement. Last but definitely not least, I would like to thank all of the producers I have met and spoke with while working with Dr. Fontenot. It is for their advancement in which I strive.

TABLE OF CONTENTS

ACKNOWLEDGMENTS ii

LIST OF TABLES iv

ABSTRACT v

CHAPTER 1: INTRODUCTION 1
- History of Lettuce 1
- Classification 2
- Nitrates 2
- Nitrate Accumulation 4
- Objectives of this Experiment 4

CHAPTER 2: LITERATURE REVIEW 5
- Nitrate Accumulation 5
- Variety Trials 7

CHAPTER 3: MATERIALS AND METHODS 9
- Field Preparation 9
- Seed Selection and Sowing 9
- Production Practices 11
- Harvest 11
- Consumer Rating 12
- Nitrate Concentration Analysis 12

CHAPTER 4 RESULTS AND DISCUSSION 13

CHAPTER 5: CONCLUSIONS 22
- Variety Trial 22
- Nitrate Analysis 24

LITERATURE CITED 26

VITA 28

LIST OF TABLES

Table 1 Percent emergence and seed source of 45 lettuce cultivars grown in years 1 and 2 10

Table 2 Growth parameters and nitrate content measured on individual lettuce heads grouped by the four lettuce types evaluated in both years 1 and 2. 13

Table 3. Average field weight (g) of individual lettuce heads evaluated in years 1 and 2. 15

Table 4. Average height (cm) and width (cm) of individual heads evaluated in years 1 and 2. 17

Table 5. Average leaf area (cm^2) of individual heads of lettuce evaluated in years 1 and 2. 20

Table 6.Top performing lettuce cultivars nitrate concentration (ppm) evaluated in years 1 and 2. 21

ABSTRACT

Lettuce (*Lactuca sativa* L.) is an important leafy vegetable crop grown in worldwide food systems with the United States ranking second behind China in total production. In 2009 the USDA Food Economic Research Service reported 1,057,715 hectares of lettuce grown in the world. Lettuce is a common market crop grown across the United States. Nitrate (NO^{3-}) accumulation in lettuce and other leafy vegetables has been found to be a potential health threat and when consumed by humans, potentially causing methaemoglobinaemia and other diseases. There are four main types of lettuce and numerous cultivars within each type. Fresh weight yields and nitrate accumulation may vary significantly between lettuce types and/or cultivars, 45 cultivars of lettuce, representing the four types, were grown in the field under best management practices at the LSU AgCenter Botanic Gardens in Baton Rouge, LA, during Fall 2011 and Fall 2012. Based on field weight, recommended butterhead cultivars were 'Caliente' (21.6 ppm) and 'Harmony' (13.9 ppm). The recommended green leaf varieties were 'Salad Bowl' (10.6ppp) and 'Tango' (14.6 ppm). The recommended red leaf cultivars were 'Red Salad Bowl' (15.2 ppm), 'Red Sails' (15.4 ppm), and 'New Red Fire' (24.0 ppm). The recommended Romaine cultivar was 'Green Towers' (11.2 ppm) and the recommended crisphead varieties were 'Raider' (17.6 ppm) and 'Ithaca' (14.9 ppm). When comparing the highest yielding cultivars from the field weight study, 'New Red Fire' (24.0 ppm) produced the highest nitrate concentration in both years 1 and 2. It made up 3.9% of the RfD (EPA's maximum acceptable oral dose of a toxic substance) for men and 4.59% of the RfD for women. This shows no concern for men or women in the 20-74 age group.

CHAPTER 1: INTRODUCTION

History of Lettuce

Lettuce, *Lactuca sativa* L., is native to the Mediterranean area and inner Asia Minor and was domesticated along the shores of Egypt around 4,500 B.C. It was grown throughout the Mediterranean region and was commonly planted throughout the Roman Empire. During the 7th century, lettuce cultivation was reported in China. Spanish explorers brought the plant to the New World and by the 18th century it was widely used in the Americas (Swiader and Ware, 2002;Rubatzky and Yamaguchi, 1999).

Lettuce is an important leafy vegetable crop in current food systems. It is commonly found on restaurant menus in the United States in the form of fresh salads and serves as a common accompaniment for hamburgers, sandwiches, and tacos. When eaten fresh, it is an excellent source of bulk and fiber (Swiader and Ware, 2002). In 2009, the per capita consumption of lettuce was estimated to be 12.7 kg a year per person (USDA, 2011a). In the U.S., Romaine lettuce and leaf lettuce production have increased 125 percent with 23,755 hectares planted in 1992 to 53,580 hectares planted in 2008 (USDA, 2011b; USDA2011c). Crisp head lettuce production reduced in total acreage by 30 percent from 87,752 hectares planted in 1992 to 61,108 hectares planted in 2008 (USDA, 2011d). Following the potato, lettuce is one of the leading fresh market vegetables in acreage, production, and value with California and Arizona as the leading producers in the United States (Swiader and Ware, 2002). China leads the worldwide agricultural community harvesting 12,855,500 metric tons in 2009 followed by the United States and Spain harvesting 4,104,440 metric tons and 1,000,000 metric tons respectively (USDA, 2011e).

Classification

Lettuce is classified into four groups: crisphead, butterhead, Romaine (Cos), and looseleaf. Crisphead lettuce is also referred to as iceberg lettuce. Crisphead is characterized by having a large, solid head weighing more than 907g and measuring more than 15cm in diameter. The leaves are crisp and brittle with prominent veins and midribs. Crisphead is the more tolerant of shipping and handling than all other types and therefore is the leading type of lettuce grown in the U.S. (Swiader and Ware, 2002) despite a decline in production beginning in 1989 (USDA, 2011d). Romaine lettuce is characterized by long, narrow foliage, upright growth habit, and loose, elongated heads. Butterhead lettuce is characterized by smooth, soft, and pliable leaves forming a loose head. The veins and midribs of butterhead types are not as prominent as in crisphead types and are considered to have better table quality and a more delicate flavor than crisphead types. There are two subgroups of butterhead lettuce: Boston and bibb lettuce. Bibb lettuce is smaller and darker green than Boston lettuce. Looseleaf lettuce is characterized as producing an open rosette of leaves loosely arranged on the stalk. There is a considerable amount of variation in leaf color within looseleaf lettuce, ranging from green and purple to red. There is also variation in looseleaf texture and margin shape (Swiader and Ware, 2002). Although lettuces vary in visual differences all subcategories assimilate and concentrate nitrate (NO^{3-}) in their leaf tissue. Nitrate is incorporated into proteins and other nitrogenous compounds and is used as a terminal electron receptor in the respiratory chain of chloroplasts (Hill, 1996).

Nitrates

Nitrate (NO^{3-}) is an integral component of the nitrogen cycle and is found throughout the environment. The primary routes of entry of nitrates in human diet are drinking contaminated ground water and oral ingestion of leafy vegetables (Hill, 1996). It is estimated that 80% of

dietary nitrates are derived from leafy vegetable consumption (Tamme, 2011). Although nitrate itself at normal levels, below 3.7mg nitrate ion/kg body weight per day in the European Union and below 1.6 mg nitrate nitrogen/kg body weight per day in the United States (EPA, 2012), has not been proven to cause toxicological effects the nitrate metabolites, nitrite (NO^{2-}) and N-nitroso-compounds (NOCs) have profound toxicological effects on humans with some NOCs listed as known carcinogens. Intestinal-type gastric cancer and methaemoglobinaemia are extreme examples of diseases associated with high nitrate exposure (Hill, 1996). There are many types of gastric cancer or stomach cancer that form in the stomach membrane. Adenocarcinoma is the most common worldwide. In the US, 10,340 people died and 21,550 new cases of stomach cancer were reported (NCI, 2011). The sole treatment for the gastric cancer is removal of the stomach (gastrectomy) along with radiation therapy and chemotherapy to reduce reoccurance (NLM, 2011). Methemoglobinemia is a blood disorder where abnormal amounts of methemoglobin are produced in the body. Methemoglobin is a form of hemoglobin but differs by not releasing oxygen to the body. It is commonly referred to as blue baby syndrome. Methemoglobinemia is genetic or induced by the presence of certain drugs, chemicals or foods. There are two forms of the genetic version. Form 1 can be found as Type 1 when red blood cells completely lack the enzyme, cytochrome b5 reductase or Type 2 distinguished by the enzyme being incompatible in the body. Form two is inherited genetically and is known as hemoglobin M disease and is caused by defects to the hemoglobin molecule. Acquired methemoglobinemia is more common, and occurs in people after they are exposed to particular chemicals including nitrates. Infants and children can be vulnerable if they are fed excessive vegetables containing high levels of nitrates such as Swiss chard, spinach, beetroot, celery, and squash (Santamaria et al, 1999). These vegetables typically served in the form of pureed baby food.

Nitrate Accumulation

Nitrate accumulation in plant leaf tissue is influenced by the concentration in soil, environmental conditions and by genotype. Reinink and Eenink (1988) report, low light conditions increase nitrate concentration within leaf tissue. Lettuce genotypes with lower nitrate concentration levels have been determined and are utilized in plant breeding programs. Reinink (1991) introduced the concept of two genotype by environment (GE) interactions, daily variations and annual variations. Nitrate assimilation in plants is the process by which nitrate is converted and incorporated into carbon compounds within the plant such as pigments, lipids, nucleic acids, or amino acids. Nitrate reductase catalyzes of the reaction for nitrate assimilation and is regulated by several factors, one being light (Taiz and Zeiger, 2006). Thus nitrate concentration may be determined by the duration and intensity of light during the fall and spring crop cycles.

Objectives of this Experiment

1. Evaluate 45 lettuce cultivars to determine the highest yielding cultivars of each lettuce type under typical commercial production standards in Louisiana.
2. Determine nitrate concentration in the foliage of the highest yielding lettuce types and cultivars.

CHAPTER 2: LITERATURE REVIEW

Nitrate Accumulation

Nitrate content in leaf tissue of lettuce is influenced by genotype. Reinink et al. (1987) evaluated 135 genotypes of lettuce for nitrate concentration. Two experiments were conducted. In experiment 1 plants were grown on recirculating nutrient film and in experiment 2 plants were grown in large pots containing potting soil. Nitrate content was determined using a Skalar Autoanalyzer (Breda, The Netherlands). In both cultivated and wild lettuce genotypes, differences in nitrate values were found. The highest nitrate values were in wild lettuce ranging from 1.6 g/kg to 5.1g/kg fresh weight and the lowest nitrate levels were found in butterhead types of cultivated lettuce ranging from 1.3g/kg to 3.7g/kg fresh weight. Ranges in plant dry matter values were observed with wild lettuce genotypes averaging the greatest (8.7% and 9.4% dry matter in experiment 1 and 2) and crisphead types of cultivated lettuce averaging the lowest (6.9% and 6.8% dry matter in experiment 1 and 2. A negative correlation between nitrate content and plant dry matter was observed in all lettuce types and the highest was observed within butterhead lettuce types. As plant dry matter increased, nitrate content decreased. (Reinink et al, 1987).

In an attempt to determine differences in nitrate accumulation between leaf tissue and root tissue, Reinink and Eenink (1988) conducted a study between nine different lettuce cultivars grown in three different nitrate concentrations: 6.9, 10.5, and 13.5 $mmol^{-3}$. Two Romaine cultivars and seven butterhead cultivars were represented. Plant shoot tissue (leaves) and root tissue were harvested and analyzed for nitrate content using a Skalar Autoanalyzer. Results indicated that roots accumulated nitrate (nitrate content in root tissue was 4-9 times higher than the nutrient solution) and the nitrate content of roots is closely correlated to the nitrate content of

shoots. However, differences in nitrate content observed among different cultivars were greater in shoots than in roots. Thus, research focuses on nitrate content in shoots because lettuce roots are generally not consumed. In 1991, Reinink studied two genotype by environment (GE) interactions in Butterhead type lettuce: interactions related to daily variations in light intensity and those related to annual variations in light intensity. Daily variations in nitrate content influenced by light intensity were not detected however significant annual variations in nitrate content caused by changes in light intensity were found. Burns et al. (2010) grew 24 cultivars of lettuce from all four types including wild types inside a glasshouse during both winter and summer months. Similar effects of genotype and light intensity were found. Nitrate content was found to be higher in winter crops more so than summer grown crops.

In 2009, Novaes et al. analyzed nitrate content in lettuce between crops grown in soil and crops grown in hydroponic conditions. The lettuce plants harvested from the hydroponic system contained higher levels of nitrates (71.5 g/kg dry weight) than the field grown lettuce (29.8 g/kg dry weight). Salomez and Hofman (2009) investigated nitrogen (N) nutrition and its effects on shoot nitrate concentration in butterhead type lettuce grown in soil. The soil's mineral N content had a direct effect on shoot nitrate concentration. By using the lower suggested rates of 60 and 80 kg N/ha, nitrate concentration was reduced in 17 of 24 experiments and only decreased field weight in 2 of the 24 experiments. Weightman et al. (2006) studied light level, time of harvest and field position and their relation to nitrate concentration in lettuce. Short term shading had no effect on nitrate accumulation but there were significant differences in time-of-day harvested in contrast to Reinink in 1991. In 2010, Tamme et al. studied nitrate content of many leafy vegetables and herbs including lettuce. Nitrate content of winter grown lettuce was 22% higher than summer grown lettuce.

Hanafy et al. (2000) studied the effect of 4 bio-fertilizers on growth, yield, chemical composition and nitrate accumulation of lettuce. All treatments received 100kg/fed (kg/0.41ha) ammonium sulphate (20.5% n), 150kg/fed calcium superphosphate (15.5% P_2O_5), and 50kg/fed potassium sulphate (48% K_2O).The four biofertilizers were Rhizobactriena (commercial product containing *Azospirillium sp.* and *Azotobacter sp.*), Microbien (commercial product containing N-fixing + phosphorous dissolving bacteria), Nitrobien (commercial product containing *Azospirillium sp*. and *Azotobacter sp*.), and Biogien commercial product containing *Azotobacter sp*.). All treatments were compared to a control which did not receive an application of any biofertilizer. Differences related to plant growth were not found in crops treated with Rhizobactrien or Biogien. Crops treated with Microbian recorded significant decreases in plant growth. Significant decreases in nitrate content were observed in all treatments especially in crops treated with Nitrobien, Biobien, and Rhizobactrien.

Variety Trials

Commercial producers are constantly search for better performing lettuce cultivars. Researchers in Kentucky studied seventeen varieties of Romaine lettuce including ‘Green Towers’, ‘Ideal’ and ‘Paris Island’, in order to select the optimum cultivar for production. ‘Jericho’ and ‘Ideal’ had the greatest field weight (Spalding and Coolong, 2008). Oklahoma State University evaluated nine leaf lettuce cultivars and found that ‘Tropicana’ and ‘Green Star’ had the greatest field weight. Kemble et al, 2012, conducted a field weight study on Romaine, bibb and leaf lettuce varieties including ‘Starfighter’, ‘Bergam’s Green’, ‘Nevada’, ‘New Red Fire’ and ‘Northstar’, and found ‘Starfighter’ was rated the greatest in one location and ‘North Star’ was the greatest cultivar in the other location. Eight Romaine lettuce cultivars including ‘Green Towers’, ‘Musena’, and ‘Ideal’ were also evaluated at the same locations. The cultivar

‘Ideal’ was rated greatest field weight at location one and ‘Green Forest’ was rated greatest field weight at location two. Purdue University in Indiana conducted a variety trial of several lettuce types and cultivars in order to identify cultivars to be used in a future replicated study. ‘Pirat’ had the greatest field weight among bibb cultivars. ‘Green Forest’ was rated greatest field weight among Romaine cultivars, and ‘Tango’ had a greater field weight among other leaf cultivars (Maynard, 2013).

CHAPTER 3: MATERIALS AND METHODS

Field Preparation

A lettuce trial was planted at the Louisiana State University AgCenter Botanic Gardens located at 4560 Essen Lane, Baton Rouge, Louisiana 70809 using cultivation practices from the Louisiana Commercial Vegetable Production Recommendations, publication number 2433 (Boudreaux, 2009). Forty-five varieties of lettuce were planted in a randomized complete block design on a 0.28 hectare field over two consecutive growing seasons 2011-2012.

Glyphosate was applied as a burn down, non-selective herbicide at a rate of 3pt active ingredient per acre (Boudreaux, 2009; Kemble, 2011) 4 weeks prior to transplanting. The field was disked and shaped into 121.9 cm rows. A 13-13-13 fertilizer (5.1% ammoniacal nitrogen, 7.9% urea nitrogen) was applied as a pre-plant application at a rate of 90.8kg/ha and black plastic mulch (Agriculture Solutions, LLC, Strong, ME) and drip-tape (Netafilm, Fresno, CA) (emitters on 30.5cm centers) were installed. Kerb™ (Dow AgroSciences, Indianapolis, IN) was applied for pre-emergent weed control on the row middles at a rate of 3.5kg active ingredient per hectare. Herbicide was not applied under the plastic.

Seed Selection and Sowing

Forty-five varieties of lettuce representing four types of lettuce (Romaine, butterhead, leaf, crisphead) were selected for study. Seed was sourced from several companies (Table 1). Recommended varieties for the southeast U.S. (Kemble, 2011) influenced the varieties chosen. All seeds were sown into 98ct plug trays (T.O. Plastics, Inc., Clearwater, MN), city, state) filled with media (Sunshine Mix #3, Agawam, MA), 35 d prior to transplanting in year 1 and 31 d prior to transplanting in year 2. Seedlings were grown in a greenhouse with average temperatures between 30°C and 13.9°C in year 1. Average temperatures in year 2 were between 31.6°C and

15.5°C. Plants were monitored daily and watered as needed by hand. Cotyledon emergence and first true leaf appearance was observed and recorded every 7 d for 28 d (Table 1). Transplants were hardened for 7 d prior to transplanting in the field.

Table 1. Percent emergence and seed source of 45 lettuce cultivars grown in years 1 and 2.

Cultivar	% Emergence Year 1	% Emergence Year 2	Source
Caliente	96	100	Siegers
Harmony	98	82	Siegers
Sylvesta	97	99	Johnny's
Adriana	92	97	Johnny's
Buttercrunch	96	98	Johnny's
Esmeralda	50	100	Territorial
Drunken Woman	60	94	Territorial
Frizzy Headed Skyphos	97	100	Siegers
Summer Bibb	95	91	Siegers
Ithaca	98	97	Harris
Raider	98	98	Siegers
Great Lakes	74	96	Rupp
Keeper	96	90	Siegers
Bergman's Green	97	99	Siegers
Cherokee	100	99	Siegers
Grand Rapids	64	71	Rupp
Green Vision	89	88	Siegers
Lolla Rossa	78	99	Harris
New Red Fire	36	97	Siegers
Northstar	11	92	Siegers
Oakleaf	81	96	Rupp
Panisse	35	97	Johnny's
Red Sails	89	99	Johnny's
Red Salad Bowl	17	98	Rupp
Salad Bowl	85	97	Territorial
Sierra	98	98	Rupp
Slobolt	48	98	Harris
Starfighter	97	53	Siegers
Tango	97	99	Johnny's
Tehama	100	99	Siegers
Two Star	97	90	Johnny's
Waldmann's Green	18	99	Rupp

N=294 for emergence rate

Production Practices

The field was planted in a randomized complete block design. Five plots per variety (40 plants per plot) were planted. Plots were 12.2 m long. Two rows of lettuce plants were planted on 121.9cm wide rows with 0.91m between plots and 1.52m alleyways between each block (Figure 1) on Nov 11 in year 1 and Oct 29 in year 2. Varieties were randomly assigned to plots within each of the five blocks. The weather conditions on both planting days, Nov 11 in year 1 and Oct 29 in year 2 were sunny with a high temperature of 19.4°C in year one and 18.8°C in year two. Irrigation was automated at 30min/d beginning at 7:00am. Calcium nitrate fertilizer (14.5% nitrate nitrogen, 1.0% ammoniacal nitrogen) was injected through irrigation lines at 20lb N/acre in the form of $CaNO_3$ per week for 3 weeks starting 28 d and 25 d, respectively, after planting in years 1 and 2.

Harvest

Leaf lettuce was harvested 40 d after planting in year 1 and 39 d in year 2. Plants were cut 3.81cm above the mulch layer. Plant material was weighed in the field (field weight) and placed inside a (0.95 L) plastic bag. Yield was determined from field weight. Ten lettuce plants were harvested from the middle of each plot. Bibb lettuce was harvested 63 d after planting in year 1 and 60 d after in year 2. Both Romaine and crisphead types were harvested at the same time with 67 prior to planting in year and 67 d in year 2. Height, two perpendicular widths and fresh weight were measured from each lettuce sample. Plants were cut even with the mulch layer, weighed, and placed into individually labeled bags. Immediately after harvest, lettuce was stored in a walk-in cooler at 4.4°C. Leaf surface area was measured on a subsample using a Li-Cor surface area instrument (LI-COR Environmental, Lincoln, NE) and then extrapolated to determine total leaf area. All plant shoot material was then dried for 168 h at 60° C.

Consumer Rating

One sample of each cultivar of average size and appearance was harvested from the field and brought to Louisiana's State University's main campus in both years 1 and 2. Each specimen was given a random number and placed on tables. Faculty, staff and students throughout the University were asked to rate their top three cultivars by visual appearance. Eighty-seven people participated in year 1 and 96 people participated in year 2.

Nitrate Concentration Analysis

Leaf nitrate content analysis was conducted in the Louisiana State University's Department of Agricultural Chemistry laboratories. Cultivars with the heaviest field weight along with the cultivars with the most votes in the consumer rating experiment in year 1 were analyzed. Five dried, random samples of each tested variety were ground and passed through a #40 mesh sieve with 0.40mm openings. Two grams of each pulverized sample were added to 250ml volumetric flask along with 50ml of deionized water. Flasks were then placed into a hot water bath at 29.4°C and shaken for 45 min. An additional 200ml of deionized water was then added to each flask. Flasks were shaken and solution was poured through a Whatman 11cm folded paper filter into 16x125mm polystyrene test tubes. The filtrate was diluted by 1/5 with deionized water. The diluted filtrate was then transferred to 4.0mL Fisherbrand™ polystyrene sample cups (Thermo Fisher Scientific, Pittsburg, PA and loaded into a SEAL AutoAnalyzer 3 - Automated Segmented Flow Analyzer (Seal Analytical, Mequon, WI) to determine total nitrate content using the Kjeldahl total nitrogen method (EPA, 1993).

CHAPTER 4 RESULTS AND DISCUSSION

There were four types of lettuce, butterhead, crisphead, leaf and Romaine representing 45 cultivars evaluated in this study. Significant differences in average fresh weight by lettuce type were found (Table 2).

Table 2. Growth parameters and nitrate content measured on individual lettuce heads grouped by the four lettuce types evaluated in both years 1 and 2.

Lettuce type	Field Weight (g)	Height (cm)	Width (cm)	Leaf Area (cm^2)	Nitrate (ppm)
Butterhead	369.4[w] c	15.0[x] c	28.7[y] b	5619.8[z] b	17.8 a
Crisphead	416.7 b	13.5 d	25.7 c	4343.3 c	16.2 a
Leaf	178.4 d	15.8 b	28.8 b	3015.4 d	16.9 a
Romaine	462.5 a	26.5 a	32.5 a	7134.3 a	8.2 b

[w] Field weight is the fresh weight of individual harvested lettuce heads measured in the field prior to removal of wrapper leaves.
[x] Plant height was calculated from the top of the soil line to the upper most point of growth on each lettuce head.
[y] Average width was calculated by measuring the lettuce head on a y and x axis and averaging the two widths.
[z] Average leaf area was calculated using a Li-COR leaf area instrument (LI-COR Environmental, Lincoln, NE).
Means in columns followed by the same letter are not significantly different as determined by Tukey grouping, $P \leq 0.05$.

Romaine lettuce was the heaviest grown in the study followed by crisphead and butterhead with leaf lettuces being the lightest of all lettuce types grown in this study. Although differences occurred between lettuce types, differences in weight between cultivars within each type did not always occur (Table 3). Romaine lettuce was the tallest lettuce type grown in both years 1 and 2 followed by leaf, butterhead, and crisphead being the shortest. Although height differences occurred between lettuce types, differences in height between cultivars within type did not always occur (Table 4). Romaine lettuce was the widest lettuce of all types grown in both years 1 and 2. There were no differences in width between leaf and butterhead lettuce types. However all three lettuce types were wider than crisphead lettuce. Although differences occurred between lettuce types, differences in width between cultivars within type did not always occur

(Table 4). The measurements of weight or height and width of lettuce serve as different standards for determining lettuce quality and therefore price. However, lettuce producers often sell lettuce by the pound where greater weights lead to greater revenue over lower weighing types. Some producers sell lettuce by the head. In this instance, consumers often gravitate to larger sized lettuce heads (height and width) under the assumption that a larger head is a greater bargain. Average leaf area quantified in this experiment provided an additional measurement of lettuce quality. Romaine lettuce had the greatest leaf area of all lettuce types evaluated followed by butterhead and crisphead types. Leaf lettuce had the smallest leaf area of all lettuce types trialed. Although differences occurred between lettuce types, differences in leaf area between cultivars within type did not always occur (Table 5).

While growth characteristics are an important measurement to determine lettuce size and potential consumer preference, nitrate content is also important when determining lettuce quality. Therefore, cultivars with the greatest field weight within each of the four types of lettuce trialed were evaluated for nitrate concentration. Romaine had the lowest nitrate levels of all four lettuce types in both years 1 and 2 (Table 2). There were no differences found in nitrate concentration between butterhead, crisphead and leaf lettuce types. Romaine type cultivars had a lower nitrate concentration than all of types of lettuce tested.

Table 3. Average field weight (g) of individual lettuce heads evaluated in years 1 and 2.

Lettuce Type	Lettuce Cultivar	Field Weight (g)
Butterhead	Adriana	376.9 b-h
	Buttercrunch	377.6 b-h
	Caliente	460.9 a-d
	Drunken Woman Frizzy Headed	318.5 f-l
	Esmeralda	321.8 e-k
	Harmony	472.0 a-d
	Skyphos	324.0 e-j
	Summer Bibb	270.9 g-m
	Sylvesta	401.4 b-g
Crisphead	Great Lakes	402.2 b-g
	Ithaca	441.2 a-f
	Keeper	374.7 c-h
	Raider	448.6 a-f
Leaf	Bergam's Green	195.4 j-o
	Cherokee	113.6 no
	Grand Rapids	203.0 i-o
	Green Vision	170.3 m-o
	Lolla Rossa	80.0 o
	Nevada	158.9 m-o
	New Red Fire	187.6 j-o
	Northstar	184.5 i-o
	Oakleaf	250.6 h-m
	Panisse	222.4 i-o
	Prizehead	190.3 i-o
	Red Sails	171.3 m-o
	Red Salad Bowl	145.1 m-o
	Salad Bowl	229.1 i-n
	Sierra	174.8 m-o
	Sloblot	141.5 m-o
	Starfighter	191.5 j-o
	Tango	187.3 k-o
	Tehama	210.5 i-o
	Two Star	182.0 l-o
	Waldmann's Green	154.6 m-o
Romaine	Bambi	348.6 d-i
	Cimmaron Red	379.0 b-h
	Cuore	486.9 a-d
	Flashy Trout Back	388.4 b-g
	Green Towers	508.8 a-b
	Ideal	501.9 a-c
	Musena	476.1 a-e
	Parris Island	485.4 a-d
	Red Eye	424.1 a-f
	Ridgeline	550.4 a
	Tall Guzmaine Elite	537.4 a-f

Mean comparison within columns by SAS Proc Mixed with Tukey at $P \leq 0.05$. Means with the same letter do not differ at the 5% significance level.

Romaine lettuce was the heaviest lettuce type grown followed by crisphead and butterhead types. Leaf lettuces were the lightest of all lettuce types produced in this study. However, differences occurred between cultivars within lettuce types. For example, within the Butterhead type, 'Caliente' and 'Harmony' were heavier than 'Drunken Woman Frizzy Headed', 'Esmeralda', 'Skyphos' and Summer Bibb. However, 'Caliente' and 'Harmony' did not have heavier field weights than the other butterhead cultivars tested. Within the crisphead category, all cultivars were the same in terms of field weight. 'Great Lakes', 'Ithaca' and 'Raider' were heavier than all of the leaf lettuce trialed. Within leaf types, 'Oakleaf' is heavier than 'Cherokee' and 'Lolla Rosa'. 'Salad Bowl' is heavier than 'Lolla Rosa'. However, 'Oakleaf' and 'Salad Bowl' do not differ in field weight from any of the other leaf types. Within Romaine types, 'Ridgeline' is heavier than 'Cimmaron Red' and 'Flashy Trout Back'. There were no other field weight differences within the Romaine type lettuces.

Romaine is generally the heaviest type of lettuce grown, however some lettuce cultivars within other types in this study were equal to Romaine cultivars. 'Caliente' and 'Harmony' did not differ in field weight from 'Cuore', 'Green Towers', 'Ideal', 'Musena', 'Parris Island', 'Red Eye', 'Ridgeline' and 'Tall Guzmaine Elite'. 'Raider', a crisphead type also produced equal field weight to 'Cuore', 'Green Towers', 'Ideal', 'Musena', 'Parris Island', 'Red Eye', 'Ridgeline' and 'Tall Guzmaine Elite.'

Table 4. Average height (cm) and width (cm) of individual heads evaluated in years 1 and 2.

Lettuce Type	Lettuce Cultivar	Height (cm)	Width (cm)
Butterhead	Adriana	14.9 f-m	30.0 a-i
	Buttercrunch	17.8 d-h	27.0 c-j
	Caliente	15.0 f-l	31.1 a-i
	Drunken Woman Frizzy Headed	19.6 de	31.5 a-i
	Esmeralda	12.9 k-m	29.1 a-i
	Harmony	16.0 e-l	30.5 a-i
	Skyphos	14.0 i-m	30.3 a-i
	Summer Bibb	11.3 m	20.6 j
	Sylvesta	13.4 j-m	28.0 b-j
Crisphead	Great Lakes	13.0 j-m	25.6 d-j
	Ithaca	13.3 j-m	25.2 f-j
	Keeper	13.7 i-m	26.5 c-j
	Raider	14.2 h-m	25.4 e-j
Leaf	Bergam's Green	14.8 f-m	27.6 b-j
	Cherokee	14.8 f-m	25.1 f-j
	Grand Rapids	20.7 d	31.4 a-i
	Green Vision	18.2 d-f	30.3 a-i
	Lolla Rossa	12.4 lm	23.5 ij
	Nevada	14.4 g-m	24.0 h-j
	New Red Fire	15.5 f-l	32.4 a-h
	Northstar	13.3 i-m	27.0 b-j
	Oakleaf	16.0 e-l	27.1 c-j
	Panisse	12.0 lm	26.5 c-j
	Prizehead	18.9 d-g	31.7 a-i
	Red Sails	16.7 e-j	32.3 a-g
	Red Salad Bowl	15.4 e-m	33.7 a-g
	Salad Bowl	15.3 f-l	34.0 a-d
	Sierra	14.7 f-m	24.3 g-j
	Slobolt	16.5 e-k	28.2 b-j
	Starfighter	15.3 f-l	27.6 b-j
	Tango	13.9 i-m	28.6 b-j
	Tehama	17.8 d-h	30.9 a-i
	Two Star	17.4 d-i	28.3 b-j
	Waldmann's Green	18.4 d-h	30.6 a-i
Romaine	Bambi	15.2 f-m	23.9 f-j
	Cimmaron Red	26.9 bc	34.1 a-c
	Cuore	24.9 c	34.0 a-d
	Flashy Trout Back	25.0 c	32.8 a-f
	Green Towers	27.7 bc	35.7 ab
	Ideal	30.5 b	31.9 a-h
	Musena	25.5 c	32.1 a-i
	Parris Island	26.9 bc	33.8 a-e
	Red Eye	26.7 c	34.4 a-c
	Ridgeline	35.1 a	37.3 a
	Tall Guzmaine Elite	27.4 bc	27.7 a-j

Mean comparison within columns by SAS Proc Mixed with Tukey at $P = 0.05$. Means with the same letter do not differ at the 5% significance level.

Romaine lettuce was the tallest lettuce types grown fallowed by leaf and Butterhead types. Crisphead lettuces were the shortest types grown in this study. However, there were some differences in cultivars between each type. For instance, within Butterhead types, 'Drunken Woman Frizzy Headed' was taller than 'Adriana', 'Caliente', 'Esmeralda', 'Skyphos', 'Summer Bibb', and 'Sylvesta'. However, 'Drunken Woman Frizzy Headed' was not taller than other cultivars within the same lettuce type. Within the crisphead lettuce type there were no differences in height. Within the leaf lettuce types 'Grand Rapids' was the tallest cultivar and was taller than 'Bergrams's Green', 'Cherokee', 'Lolla Rossa', 'Nevada', 'Oakleaf', 'Panisse', 'Red Sails', 'Red Salad Bowl', 'Salad Bowl', 'Sierra', 'Slobolt', 'Starfighter', and 'Tango'. Within Romaine lettuce types 'Ridgeline' was the tallest and was taller than 'Ideal', 'Cuore', 'Flashy Trout Back', 'Musena', 'Red Eye', and 'Bambi'. 'Bambi' was the shortest Romaine grown in this study.

Lettuce width was measured on two axes and then averaged together. Romaine lettuce was the widest type grown followed by butterhead and leaf types. There were no differences between butterhead and leaf types. Crisphead lettuce types were the narrowest type grown in the study. Despite these differences in lettuce types, there were also differences amongst lettuce cultivars between each type. For instance, butterhead cultivars 'Adriana', 'Caliente', 'Drunken Woman Frizzy Headed', 'Esmeralda', 'Harmony', and 'Skyphos' were wider than 'Summer Bibb'. There were no differences between the crisphead lettuce types. Within the leaf lettuce types 'Salad Bowl' was wider than 'Lolla Rossa', 'Nevada', and 'Sierra'. 'New Red Fire' was also greater than 'Lolla Rossa' but there is no difference between 'New Red Fire' and 'Salad Bowl'. Within Romaine lettuce types 'Cimmaron Red', 'Cuore', 'Green Towers', 'Parris Island', 'Red Eye', and 'Ridgeline' are all wider than 'Bambi'.

In general, Romaine lettuce types are taller than all other lettuce types. However, the butterhead cultivar, ‘Drunken Woman Frizzy Headed’, is taller than all crisphead cultivars but not taller than the leaf cultivars: ‘Grand Rapids’, ‘Green Vision’, ‘Prizehead’, ‘Red Sails’, ‘Red Salad Bowl’, ‘Slobolt’, ‘Tehama’, ‘Two Star’, ‘Waldmann’s Green’ and the Romaine cultivar, ‘Bambi’. Romaine lettuce types are also the widest types grown in this study, but only ‘Ridgeline’ is wider than the butterhead variety, ‘Buttercrunch’, and the leaf varieties, ‘Lolla Rossa’, ‘Nevada’, ‘Northstar’, ‘Oakleaf’, ‘Panisse’, ‘Sierra’, ‘Slobolt’, ‘Starfighter’, ‘Tango’, and ‘Two Star’. When producers are considering types of lettuce to grow, lettuce weight is important for economic return, but in some instances, consumer preferences may take part in the final decision. The overall size of the product may help persuade customers to make their final decision.

Romaine lettuce types were the largest types based on total leaf surface area followed by butterhead, and crisphead types (Table 5). Leaf type lettuces displayed the lowest leaf surface area. However, there were some differences in cultivars between each type. For instance, within butterhead types, ‘Harmony’ was larger than ‘Skyphos’, ‘Summer Bibb’, and ‘Buttercrunch’. However, ‘Sylvesta’ was larger than ‘Summer Bibb’. There were no differences between Crisphead cultivars. Within leaf lettuce types, ‘Oakleaf’ was larger than ‘Lolla Rossa’. Within Romaine types ‘Bambi’ was the smallest of all cultivars.

In general, Romaine lettuce types exhibited the largest leaf surface areas amongst all other lettuce types. However, several butterhead types including: ‘Adriana’, ‘Caliente’, ‘Drunken Woman’, ‘Harmony’, and ‘Sylvesta’ were larger than the Romaine cultivar ‘Bambi’. When producers are making decisions on which lettuce cultivars to grow, leaf surface area can be important characteristic to look at when choosing cultivars for fullness and appearance.

Table 5. Average leaf area (cm^2) of individual heads evaluated in years 1 and 2.

Lettuce Type	Lettuce Cultivar	Leaf Area (cm^2)
Butterhead	Adriana	5811.2 b-g
	Buttercrunch	4683.5d-k
	Caliente	6938.2 a-d
	Drunken Woman Frizzy Headed	5891.3 a-f
	Esmeralda	5160.9 c-j
	Harmony	7382.5 a-c
	Skyphos	4710.4 d-k
	Summer Bibb	3491.8 g-m
	Sylvesta	6508.5 a-e
Crisphead	Great Lakes	3579.7 f-m
	Ithaca	3942.5 f-l
	Keeper	4401.2 e-l
	Raider	5449.7 c-i
Leaf	Bergam's Green	3005.7 j-m
	Cherokee	2189.7 lm
	Grand Rapids	3105.6 i-m
	Green Vision	2628.0 k-m
	Lolla Rossa	1451.9 m
	Nevada	2491.9 k-m
	New Red Fire	3747.9 f-m
	Northstar	3190.5 g-m
	Oakleaf	3842.9 f-l
	Panisse	3367.6 f-m
	Prizehead	3612.2 f-m
	Red Sails	3712.0 f-m
	Red Salad Bowl	3217.0 f-m
	Salad Bowl	3714.3 f-m
	Sierra	2262.8 lm
	Sloblot	2441.5 k-m
	Starfighter	2954.8 j-m
	Tango	3010.3 j-m
	Tehama	3676.2 f-m
	Two Star	2722.2 k-m
	Waldmann's Green	2977.9 h-m
Romaine	Bambi	3103.8 h-m
	Cimmaron Red	6962.8 a-d
	Cuore	7999.5 ab
	Flashy Trout Back	6962.2 a-d
	Green Towers	8223.7 a
	Ideal	8146.2 ab
	Musena	7205.3 a-d
	Parris Island	7346.9 a-c
	Red Eye	7489.1 a-c
	Ridgeline	7912.9 ab
	Tall Guzmaine Elite	7125.3 a-h

Mean comparison within columns by SAS Proc Mixed with Tukey at $P \leq 0.05$. Means with the same letter do not differ at the 5% significance level.

Table 6. Top performing lettuce cultivars nitrate concentration (ppm) evaluated in years 1 and 2.

Lettuce Type	Lettuce Cultivar	Nitrate-N Content (ppm)
Butterhead	Caliente	21.6 ab
	Harmony	13.9 bc
Crisphead	Ithaca	14.9bc
	Raider	17.6 a-c
Leaf	Cherokee	21.9 ab
	New Red Fire	24.0 a
	Red Sails	15.4 a-c
	Red Salad Bowl	15.2 a-c
	Salad Bowl	10.6 cd
	Tango	14.6 bc
	Two Star	15.9 a-c
Romaine	Green Towers	11.2 cd
	Ridgeline	5.2 d

Mean comparison within columns by SAS Proc Mixed with Tukey at $P \leq 0.05$. Means with the same letter do not differ at the 5% significance level.

The Romaine lettuce types accumulated the least amount of nitrate (Table 2). Within butterhead and crisphead types there were no differences in nitrate accumulation. Within leaf lettuce, ‘Cherokee’ and ‘New Red Fire’ had higher nitrate content than ‘Salad Bowl’ while the other two red lettuce cultivars ‘Red Sails’ and ‘Red Salad Bowl ‘Ridgeline’ a Romaine lettuce type was the lowest nitrate accumulating lettuce of all the four types but it did not accumulate any less nitrate content than ‘Green Towers’, a Romaine lettuce or ‘Salad Bowl’, a leaf lettuce (Table 6).

CHAPTER 5 CONCLUSIONS

Variety Trial

Lettuce producers strive to sell a high quality product to ensure a good return on investment. Wholesale lettuce is sold on a by-weight basis whereas fresh market lettuce can be sold either on a weight-basis or as a set price per individual head. Identifying lettuce cultivars that produce heavier yields by fresh weight is useful in advising local producers who sell products through the wholesale market on what cultivars will lend heavier yields and thus return greater profit. Romaine types yielded the highest weight (Table 2). Of the Romaine cultivars, produced in this study, 'Ridgeline', produced the highest field weight (550.4g).

The results of this study indicate that Louisiana producers should grow Romaine lettuce when weight dictates price. However, if consumer preference is for lettuce types other than Romaine, producers should grow other lettuce types. Crisphead type lettuce cultivars had the second highest field weight. The butterhead lettuce type ranked third in field weight and leaf lettuce types ranked last in field weight (Table 2).

Among Romaine cultivars of lettuce grown in the current study, 'Green Towers' had the highest field weight with similar results to Kemble et al (2012). However, the weight of 'Green Towers' was similar to the field weight of 'Ridgeline'. Spalding and Coolong at (2008), also found that 'Green Towers' produces one of the heaviest field weights but was similar in weight to all other Romaine cultivars included in the study. The results in an Indiana study were similar where 'Green Towers' produced some of the highest field weights.. (Maynard, 2013).

Within the crisphead type of lettuce grown in years 1 and 2 of the study, 'Raider' had the greatest field weight but it was no different than the three other cultivars tested (Table 3).The butterhead types, 'Harmony' and 'Caliente' had the first and second highest field weights,

respectively out of all the other varieties tested but were similar in weight to ‘Adriana’, ‘Buttercrunch’, and ‘Sylvesta’. ‘Harmony’ and ‘Caliente’ were not included in the Indiana study but ‘Adriana’ and ‘Sylvesta’ were recommended for further testing (Maynard, 2013).
Leaf lettuce cultivars ‘Oakleaf’ and ‘Salad Bowl’ produced the greatest field weight. Although ‘Oakleaf’ and ‘Salad Bowl’ were not included in the Alabama study, ‘Bergam’s Green’, ‘Nevada’, ‘New Red Fire’, ‘Northstar’, and ‘Starfighter’ were included. ‘Starfighter’ and ‘Northstar’ had the heaviest field weights (Kemble et al, 2012) and were not different than ‘Oakleaf’ and ‘Salad Bowl’ (Table 3) in the LSU AgCenter study. In the Indiana study ‘Panisse’ peoduced some of the highest field weights of the cultivars tested (Maynard, 2013) and interesting enough it recorded a very similar field weight in both the Indiana study and the current study (Table 3).

Producers looking for specific cultivars with the greatest average field weight within the Romaine type should use ‘Ridgeline’ and ‘Green Towers’. For crisphead type lettuce, producers should grow ‘Raider’ and ‘Ithaca’. For butterhead types lettuce, producers should grow ‘Caliente’ and ‘Harmony’ while ’Oakleaf’ and ‘Salad Bowl’ should be grown if leaf lettuce is desired.

Some producers prefer to sell individual heads of lettuce, therefore, total size of the product may be the biggest determinant of overall sales. When looking at each type of lettuce grown, the Romaine type were both wider and taller than all other types grown in this study (Table 2). Leaf lettuces came in second while Crisphead types came in fourth and butterhead types came in third tallest at 15.0cm and with no difference to the leaf types (Table 2).

Producers looking for overall product size should refer to cultivars within each lettuce type that show significant differences in size. Producers seeking tall heads may consider producing ‘Ridgeline”, ‘Ideal Cos’, ‘Tall Guzmaine Elite’, ‘Cimmeron Red’, ‘Green Towers’, and ‘ Parris Island’ (Table 4). Producers interested in tall heads of lettuce should select ‘Grand Rapids’, ‘Prizehead’, ‘Waldmann’s Green’, and ‘Green Vision’. For those looking for tall butterhead varieties they should grow ‘Drunken Woman Frizzy Headed’, ‘Buttercrunch’, ‘Harmony’, or ‘Caliente’. For producers looking for the tallest growing crisphead types they should produce either ‘Raider’ or ‘Keeper’.

If producers prefer the widest cultivars they should choose the Romaine cultivars ‘Ridgeline’, ‘Green Towers’, and ‘Cimmaron Red’. For those looking for the widest leaf lettuce types they may select ‘Salad Bowl’, ‘Red Salad Bowl’, and ‘New Red Fire’. The widest butterhead cultivars to grow include ‘Caliente’, ‘Skyphos’’, and ‘Harmony’. The widest crisphead types to grow are ‘Keeper’ or ‘Great Lakes.’

Nitrate Analysis

Leaf nitrate concentration was analyzed on cultivars that yielded the heaviest field weights. A total of 12 cultivars were analyzed for nitrate concentration (Table 6). To determine comparable levels of nitrate in cultivars within different lettuce types, the nitrate analysis data was converted from ppm nitrate to ppm nitrate-nitrogen using a conversion factor of 4.4 (EPA, 1993). The average weights of US males and females ages 20-74 are 86.8 and 74.7kg respectively (CDC, 2004). Using the EPA’s oral reference dose, or RfD, of 1.6mg NO_3-N/kg body weight/day the RfD for males would amount to 138.88mg NO_3-N/day. The RfD for females amounts to 119.52mg NO_3-N/day. The leaf lettuce cultivar, ‘New Red Fire’ recorded the highest NO_3-N content with a mean of 24.0 mg NO_3-N, accounting for 3.92% of the RfD for

men and 4.56% of the RfD for women (Table 6). The lowest concentrated lettuce cultivar was 'Ridgeline', a Romaine type. Its NO_3-N concentration accounted for 1.18% of the RfD for men and 0.99% of the RfD for women (Table 6).

For a segment of the U.S. population that is concerned with nitrate levels in food including vegetables, the nitrate data of the current study is of value to determine difference in cultivar uptake of the nitrate molecule. In this study, all of the cultivars received the same fertilization schedule. The difference in nitrate uptake is presumably due to genetic differences. It is interesting to note, red leafed cultivars generally had higher nitrate content. These recommendations are dependent on the fact that farmers take soil test from their field and apply fertilizer at recommended rates based on these tests.

LITERATURE CITED

Boudreaux, J. 2009. Louisiana commercial vegetable production recommendations. Louisiana Cooperative Extension Service. Pub. 2433.

Burns, I., K. Zhnag, M. Turner, M. Meachum, K. Al-Redhiman, J. Lynn, M. Broadley, P. Hand, D. Pink. 2010. Screening for genotype and environment effects on nitrate accumulation in 24 species of young lettuce. J. Sci. Food Agric. 91:553-562.

EPA, 1993. US Environmental Protection Agency. Method 351.2, Revision 2.0: Determination of Total Kjeldahl Nitrogen by Semi-Automated Colorimetry. Cincinnati, OH (updated 1993; accessed July 2017). Available from: http://www.epa.gov/sites/production/files/2015-08/documents/method_351-2_1993.pdf

Hanafy Ahmed, A., J. Mishriky, and M. Khalil. 2000. Reducing nitrate accumulation in lettuce (Lactuca sativa L.) plants by using different biofertilizers.

Kemble, J., E. Vinson, R. Akridge, J. Burkett. 2012., Lettuce Trials Conducted in South and Central Alabama. p11-12. Fall 2011 Commercial Fruit and Vegetable Variety Trials. Auburn University, Auburn, AL.

Kemble, J., K. Ivors, F. Louws, K. Jennings, and J. Walgenbach. 2011. Southeastern Vegetable Crop Handbook. Vance, Lincolnshire, IL.

Hill, M.J. 1996. *Nitrates and Nitrites in Food and Water*. Woodhead Publishing Series in Food Science, Technology and Nutrition No. 7. P p131-162.

Maynard, E. 2013. Lettuce Cultivar Observation Trial 2013, p41-50. Midwest Vegetable Trial Report for 2013. Purdue University, W. Lafayette, IN.

Novaes, H., D. Vaitsman, and P. Dutra. 2009. Determination of nitrate in lettuce by ion chromatography after microwave water extraction. Quim Nova 32(6):1647-1650.

Reinink, K., R. Groenwold, and A. Bootsma. 1987. Genotypical differences in nitrate content in *Lactuca sativa* L. and related species and correlation with dry matter content. Euphytica 36: 11-18.

Reinink, K. and A.H. Eenink. 1988. Genotypical differences in nitrate accumulation in shoots and roots of lettuce. Scientia Horticulturae 37: 13-24.

Reinink, K. 1991. Genotype x environment interaction for nitrate concentration in lettuce. Plant Breeding 107: 39-49.

Rubatzky, V. and M. Yamaguchi. 1999. World Vegetables: Principles, Production, and Nutritive Values. 2nd ed. Aspen Publishers, Inc. Gaithersburg, Maryland.

Salomez, J. and G. Hofman. 2009. Nitrogen nutrition effects on nitrate accumulation of soil-grown greenhouse butterhead lettuce. Commun. In Soil Sci. and Plant Analysis 40:620-632.

Spalding D., T. Coolong. 2008. Romaine Lettuce Cultivar Trial, p. 28-29. In: E. Maynard. Midwest Vegetable Trial Report for 2008. Purdue University, W. Lafayette, IN.

Swiader, J. and G. Ware. 2002. Producing Vegetable Crops. 5th ed. Interstate, Danville, IL.

Taiz, L., Zeiger, E. 2006. *Plant Physiology*. Sinauer Associates, Inc. Sunderland, Massachusetts.

Tamme, T., M Reinink, M. Roasto, K. Meremae, and A. Kiis. 2010. Nitrate in leafy vegetables, culinary herbs, and cucumber grown under cover in Estonia: content and intake. Food Additives and Contaminants 3(2):108-113.

Temme L., S. Vandevijvere, C. Vinkx, I. Huybrechts, L. Goeyens, et al.. Average daily nitrate and nitrite intake in the Belgian population older than 15 years. Food Addit Contam Part A Chem Anal Control Expo Risk Assess. 2011;28:1193–204.

USDA. 2011a. U.S. Lettuce: Per capita use, 1960-2010. USDA, Economic Research Service. 9 Dec 2011 <http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?documentID=1576>

USDA. 2011b. U.S. RomaineRomaine lettuce: Acreage, yield, production, and value, 1992-2009. USDA, Economic Research Service. 9 Dec 2011 <http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?documentID=1576>

USDA. 2011c. U.S. leaf lettuce: Acreage, yield, production, and value, 1992-2009. USDA, Economic Research Service. 9 Dec 2011 <http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?documentID=1576>

USDA. 2011d. U.S. head lettuce: Acreage, yield, production, and value, 1950-2009. USDA, Economic Research Service. 9 Dec 2011 <http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?documentID=1576>

USDA. 2011e. Lettuce, all types: Production, by selected state, 1960-2009. USDA, Economic Research Service. 12 Dec 2011. <http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?documentID=1576>

Weightman, R., C. Dyer, J.Buxton, and D. Farington. 2006. Effects of light level, time of harvest, and position in the field on the variability of tissue nitrate concentration in commercial crops of lettuce (*Lactuca sativa*) and endive (*Chichorium endive*). Food Additives and Contaminants 23(5):462-469.

VITA

William D. Afton is the second child of Alan and Marlene Afton, born in December 1984. He was born in the town of Bemidji located in Beltrami County Minnesota, but raised in Baton Rouge, Louisiana. He graduated Baton Rouge Magnet High School in 2002 and started taking college classes in 2009 at LSU. He graduated with a bachelor's of science degree in Spring 2011 and pursued graduate studies later in the fall. In between classes, Mr. Afton worked as supervisor at a local plant nursery and retail center, Clegg's Nursery, accruing 11 years of service in the retail horticulture industry. Mr. Afton is currently employed by the Louisiana Cooperative Extension Service and the LSU AgCenter as the County Agent in St. Tammany Parish. Mr. Afton is also a contributing writer for Louisiana Gardener Magazine and volunteers his time as a shotgun instructor for the St. Tammany Shooting Sports Association.