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EFFECTS OF SOIL SILICON AMENDMENT AND NITROGEN LEVELS ON RICE INSECT PEST COMPLEX

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 Department of Entomology

by James Michael Pabicon Villegas B.S., Ateneo de Manila University, 2012 December 2017 To my wife, Merry My son, Leon My mom, Julie My dad, Jimmy My brothers Jaime and Jaimar My sisters Dem and Queencel My grandmother, Elsa.

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Abstract

Rice plants are consistently subjected to various pressures by insect pests throughout the growing season. The main insect pest complex reported in Louisiana are the rice water weevil (major early-season insect pest), fall armyworm (sporadic early-season pest), sugarcane borer and rice stalk borer (long- established but sporadic stem boring pests), Mexican rice borer (an invasive stem-boring pest), and rice stink bug (major late-season pest). Soil silicon amendment has been shown to enhance plant resistance against herbivorous pests. Rice is a typical siliconaccumulating graminaceous crop. Field and greenhouse experiments were conducted from 2015 to 2017 to investigate the effects of soil silicon amendment and nitrogen levels on the rice insect pest complex In Louisiana. In the greenhouse, we found that force required to penetrate rice stem was higher on silicon-treated rice plants compared to untreated plants. Total phenolic content on the other hand, decreased when nitrogen rate was increased. In the field experiment, the effects of soil silicon amendment on rice water weevil densities, whitehead incidences, stink bug population, and yields were found to be weaker than the effects of nitrogen fertilization. Furthermore, separate field experiments were conducted in 2016 and 2017 to investigate the effects of silicon fertilization and rice cultivars on the rice insect pests. Results showed reductions in weevil larval densities in silicon treated plots compared to untreated plots in one core sampling in 2016. Similarly, higher yields were observed in silicon treated plots compared to untreated plots in 2016. Silicon amendment did not affect whitehead incidences and rice stink bug densities in both years. The levels of infestations of rice water weevil, stem borers, and rice stink bugs were also were also found to vary among the rice cultivars evaluated in the study. Despite the weak effect of silicon on insect pests in this study, silicon could still play an

important role in rice production considering the positive effects on yield and documented effects on disease suppression.

Chapter 1. General Introduction

Rice (*Oryza sativa* L) is one of the most important crops globally. It is a staple for nearly half of the world's seven billion people (Mohanty 2013). The United States is a major rice producer worldwide with a total production of 14.4 million tons in 2016 (USDA-ERA 2017). Louisiana is the third largest rice-producing state in the U.S. with a total production of 1.6 million tons of rice in 2016 (USDA-NASS 2017). Production of rice in Louisiana is concentrated in the southwestern and northeastern part of the state. The majority of the rice in Louisiana is drill-seeded and grown under flood for the majority of the growing season.

Both biotic and abiotic factors can contribute to yield reductions in rice production. Specifically, damage by insect pests is a major limiting factor worldwide for rice production (Pathak and Khan 1994). In the United States, the rice water weevil, Lissorhoptrus oryzophilus Kuschel (Coleoptera: Curculionidae), is consistently the most destructive and widespread earlyseason pest of rice (Way 1990, Aghaee and Godfrey 2014). This pest poses a global threat to rice production, having recently invaded rice-producing regions of Asia and Europe (Saito et al. 2005). Adult weevils overwinter in leaf litter, bunch grasses, and stubble in and around rice fields, and emerge in early spring (Shang et al. 2004). Typically, adult rice water weevils feed on young rice leaves, leaving longitudinal scars along the leaf blades (Stout et al. 2002). Injury from adult feeding is generally not economically important. Flooding of rice fields triggers female weevils to oviposit, primarily in leaf sheaths beneath the water surface (Stout et al. 2002a). Rice water weevil larvae feed on the rice leaves and stems after eclosion but soon move down to the roots to feed until pupation. Root pruning by the soil-dwelling larvae can cause extensive injury to the root systems causing yield losses due to reduction in tillers and grains per panicle at maturity (Zou et al. 2004, 2004a). Infestations by rice water weevil larvae can result in yield

losses exceeding 25% when fields are left untreated and can be higher under heavy weevil pressure (Zou et al. 2004a, Reay-Jones et al. 2008).

The fall armyworm, *Spodoptera frugiperda* Smith (Lepidoptera: Noctuidae), is also an early-season pest but only sporadically infests rice fields in Louisiana. Female fall armyworm oviposit on young rice leaves and larvae can defoliate entire rice plants when densities are high (Pantoja et al. 1986, Stout et al. 2009). It has also been reported that increased infestation levels of fall armyworm resulted in reductions on plant and panicle density and reductions in rice yields (Pantoja et al. 1986).

Stem borers are important pest worldwide that attack rice plants from the seedling stage to maturity (Akinsola 1984). A complex of stem boring lepidopteran pests has been reported to attack Louisiana rice. The sugarcane borer, *Diatraea saccharalis* F. (Lepidoptera: Crambidae), and, less commonly, the rice stalk borer, *Chilo plejadellus* Zincken (Lepidoptera: Crambidae), have been reported to be the most economically important stem boring pests of rice in Texas and Louisiana (Bowling 1967, Roe et al. 1981). The Mexican rice borer, *Eoreuma loftini* Dyar (Lepidoptera: Crambidae), has invaded Louisiana in the past decade (Wilson et al. 2015, 2017) and is predicted to cause significant economic yield losses once fully established (Reay-Jones et al. 2008). Generally, adult stem borers lay overlapping eggs in clusters of 2-100 eggs on rice leaf blades. Upon hatching, larvae descend toward the base of the plant and crawl between the leaf sheath and the stem. The stem borer larvae will feed within the leaf sheath for a few days before boring into the stem. When feeding occurs during the vegetative stage of the rice plant, the central leaf whorl does not unfold, but turns brownish and dries off and the affected tillers dry out without bearing panicles, a condition known as a deadheart (Pathak and Khan 1994). When feeding occurs at reproductive stages of the rice plant, injury on the growing plant parts from the

base dries the panicles which may emerge but do not produce grains and remain straight and are whitish, a condition known as a whitehead (Pathak and Khan 1994). Stem borer activity in rice fields is often measured through whitehead incidence because it was reported that there appears to be a negative correlation between the number of whiteheads and crop yield (Way, Reay-Jones, and Reagan 2006).

The rice stink bug, *Oebalus pugnax* F. (Hemiptera: Pentatomidae) is considered the major late season insect pest of rice in Louisiana. Rice stink bugs feed on rice at the flowering, grain filling, milk, and dough stages of grain development. Feeding by the rice stink bug does not cause significant yield losses under most circumstances but greatly affects the quality of rice (Tindall et al. 2005, Espino 2007). Stink bugs also feed on other graminaceous species like sorghum, wheat, oats, rye, barley, barnyardgrass, broomsedge, Johnson grass, bearded sprangle, and broadleaf signal grass but it prefers rice to other hosts (Douglas 1939, Odglen and Warren 1962, Tindall et al. 2005).

Control strategies for these rice insect pests rely heavily on chemical insecticides (Way 1990, Johnson et al. 2003, Reay-Jones et al. 2007). Seed treatments such as the anthranilic diamide chlorantraniliprole (Dermacor X-100, DuPont Crop Protection, Wilmington, DE) have been the most effective management tactic in reducing rice water weevil larval densities (Hummel and Stout 2010, Way 2010a, 2010b, Stout et al. 2011) and can also reduce performance of stem borers and densities of whiteheads in the field (Way 2010b, Sidhu et al. 2014). Another control tactic for managing stem borer infestations and yield losses in Texas has been the use of foliar applications of pyrethroids at the reproductive and late boot or early heading phases of rice development (Reay-Jones et al. 2007, Way and Espino 2010). Furthermore, surveys conducted by Blackman et al. (2014) show that λ -cyhalothrin (Karate

Zeon®, Syngenta Crop Protection, Greensboro, NC) and z-cypermethrin (Mustang® Maxx, FMC, Research Park Triangle, NC) are the primary products used against rice stink bugs in Louisiana and Texas. However, the use of insecticides has several negative consequences such as pest resurgence, health hazards to users, environmental contamination, and costs associated with multiple applications (Chelliah and Bharathi 1994).

Cultural methods that involve weed control, field draining, delayed flooding, and early planting have been utilized to manage rice water weevil, stem borers and rice stink bugs (Hesler et al. 1992, Kendig et al. 2003, Tindall et al. 2005, Stout, Harrell, et al. 2009). Host-plant resistance has also been targeted as an alternative and sustainable approach to manage rice insect pests. Some levels of resistance and tolerance traits have been observed in rice cultivars against the rice water weevil (Stout et al. 2001). In Texas, screening of rice cultivars against stem borers under field conditions have shown varying levels of injuries and yield losses among the cultivars (Way et al. 2006). Moreover, some levels of resistance to rice stink bug infestations also exist among rice cultivars but there is a negative correlation between resistance and yield (Bernhardt et al. 2004).

Fertilization can also affect the physiology of the plant which may lead to alterations in resistance to insect pests (Slansky 1990, Altieri and Nicholls 2003). Nitrogen is an essential nutrient for plant growth and development and is often a limiting factor in non-leguminous crop production systems. The effects of nitrogen on host plant nutritional quality can increase both host plant suitability for insect herbivores and tolerance of the plant to herbivore injury (McNeil and Southwood 1978, Rubia et al. 1996). The effective acquisition and utilization of nitrogen from a host plant is vital to the growth and development of phytophagous insects, and plant nitrogen level is one of the most essential factors affecting insect performance (Awmack and

Leather 2002). Nitrogen is typically found in plants at levels much lower than insect nutritional requirements (McNeil and Southwood 1978). Numerous studies have shown that increasing amounts of nitrogen fertilizer application on crops can influence plant-insect interactions and potentially increase food consumption, growth rates, reproductive rates, survival, and population densities of insect pests (Chang et al. 1985, Way et al. 2006, VanWeelden et al. 2016). In contrast, deficiency of nitrogen may modify plant metabolism and trigger insect resistance (Comadira et al. 2015). Nitrogen is the most used nutrient applied to U.S. rice to achieve the high-yield potential of modern rice cultivars (Norman et al. 2003). In Louisiana, recommendations for nitrogen fertilization on drill-seeded rice are to apply two thirds of nitrogen before permanent flood on dry ground and the remaining nitrogen at internode elongation (Louisiana State University AgCenter 2017).

Soil silicon amendment has been proposed as a tactic to augment plant resistance against phytophagous insect pests (Reynolds et al. 2009); thus, silicon fertilization can be potentially integrated into management programs for rice insect pests in rice growing regions of the United States. Rice is a typical silicon-accumulating graminaceous crop (Ma et al. 2006, Zhao et al. 2010). Although silicon is not yet classified as an essential plant nutrient, the International Plant Nutrition Institute, Georgia, USA, recently listed silicon as a 'beneficial substance' (International Plant Nutrition Center 2015). Despite the prevalence of silicon in soil, it primarily exists as silica (SiO₂) which is not readily available for plant uptake. Silicon must be in the water soluble form of monosilicic acid (H₂SO₄) to be taken up by plants (Raven 1983). Monosilicic acid is taken up by plants via active, passive, and rejective mechanisms (Cornelius et al. 2011) and is transported to the shoot where it is deposited as solid amorphous silica known as pytoliths (Yoshida et al. 1962, Jones and Handreck 1967). Once silicon is deposited, it is immobile and is not

redistributed in the plant (Epstein 1994). The role of plant-assimilated silicon has long been debated; however, the beneficial effects of silicon on plant resistance to both abiotic and biotic stresses are well established (Reynolds et al. 2009). Biotic stressors may come in the form of vertebrate animals, arthropod herbivores, and plant pathogens (Reynolds et al. 2016). Benefits from silicon fertilization, especially on soils low or limiting in this element, include increases in yields and resistance to insect pests and diseases (Alvarez and Datnoff 2001, Ma, Miyake, et al. 2001).

Silicon fertilization has been proposed to augment plant resistance via two different mechanisms. One mechanism involves deposition of amorphous silica in plant tissues that contributes to the thickening of epidermal layer, thus forming a mechanical barrier. This mechanical barrier results in increased rigidity and abrasiveness of plant tissues, thereby reducing the palatability and digestibility of plants to insect herbivores (Ma 2004, Massey and Hartley 2009). A second mechanism involves the ability of silicon to stimulate biochemical pathways related to specific plant chemical defenses and to prime hormone-mediated defense responses via the jasmonate (JA) signaling pathway (Gomes et al. 2005, Ye et al. 2013). Economically important crops such as sugarcane, rice, and barley are classified as high accumulators of silicon containing 10-100 g kg⁻¹ Si in dry weight (Ma, Goto, et al. 2001, Ma and Takahashi 2002, Liang et al. 2007).

Ideally, management of rice insect pest complex should incorporate combinations of control tactics that complement each other. A study by Villegas et al. (2017) evaluated the effects of soil silicon amendment when combined when combined with insecticide treatment and rice varieties for the management of rice insect pests. They reported that the effect of soil silicon amendment on rice water weevil densities and whitehead incidences was weaker than the effects

of insecticide treatment and variety. Despite the weaker effects of silicon on insects, rice yields were significantly improved by silicon amendment. Considering the effects of silicon on yield, amending soils with silicon slag could still play and important role in rice production.

1.1. Objectives of the study

This research was designed to investigate the independent and combined effects of soil silicon amendment, nitrogen fertilization, and rice cultivars on the rice insect pest complex. The specific objectives of the study were:

- To evaluate the effects of nitrogen rates and soil silicon amendment on rice water weevil densities, whitehead incidences, rice stink bug densities, and yields under field conditions.
- 2.) To evaluate the effects of nitrogen rates and soil silicon amendment on stem hardness, total phenolic concentration, fall armyworm relative growth rates, and sugarcane borer performance under greenhouse conditions.
- 3.) To evaluate the effects rice cultivars and soil silicon amendment on rice water weevil densities, whitehead incidences, rice stink bug densities, and yields under field conditions.

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Chapter 2. Effects of nitrogen levels and soil silicon amendment on rice insect pest complex 2.1. Introduction

Plant resistance to phytophagous insects is linked to the physiology of the plant and thus any factors that affect the physiology of the plant may lead to alterations in resistance to insect pests (Slansky 1990, Altieri and Nicholls 2003). The morphological and physiological changes in crop plants in response to fertilization, such as accelerated or delayed maturity, tissue nutrient concentrations, growth rates, size of plant parts, and thickness and hardness of epicuticle, can influence the success of many pest species in utilizing the host (Altieri and Nicholls 2003).

Nitrogen is an essential nutrient for plant growth and development and is often a limiting factor in non-leguminous crop production systems. The effects of nitrogen on host plant nutritional quality can increase both host plant suitability for insect herbivores and tolerance of the plant to herbivore injury (McNeil and Southwood 1978, Rubia et al. 1996). The effective acquisition and utilization of nitrogen from a host plant is vital to the growth and development of phytophagous insects, and plant nitrogen level is one of the most essential factors affecting insect performance (Awmack and Leather 2002). Nitrogen is typically found in plants at levels much lower than insect nutritional requirements (McNeil and Southwood 1978). Numerous studies have shown that increasing amounts of nitrogen fertilizer application on crops can influence plant-insect interactions and potentially increase food consumption, growth rates, reproductive rates, survival, and population densities of insect pests (Chang et al. 1985, Way et al. 2006, VanWeelden et al. 2016). In contrast, deficiency of nitrogen may modify plant metabolism and trigger insect resistance (Comadira et al. 2015). Nitrogen is the most used nutrient applied to U.S. rice to achieve the high-yield potential of modern rice cultivars (Norman et al. 2003). In Louisiana, recommendations for nitrogen fertilization on drill-seeded rice are to apply two thirds

of nitrogen before permanent flood on dry ground and the remaining nitrogen at internode elongation (Louisiana State University AgCenter 2017).

Silicon is the second most abundant element in soil. Although silicon is not yet classified as an essential plant nutrient, the International Plant Nutrition Institute, Georgia, USA, recently listed silicon as a 'beneficial substance' (International Plant Nutrition Center 2015). Despite the prevalence of silicon in soil, it primarily exists as silica (SiO_2) which is not readily available for plant uptake. Silicon must be in the water soluble form of monosilicic acid (H₂SiO₄) to be taken up by plants (Raven 1983). Monosilicic acid is taken up by plants via active, passive, and rejective mechanisms (Cornelius et al. 2011) and is transported to the shoot where it is deposited as solid amorphous silica known as pytoliths (Yoshida et al. 1962, Jones and Handreck 1967). Once silicon is deposited, it is immobile and is not redistributed in the plant (Epstein 1994). The role of plant-assimilated silicon has long been debated; however, the beneficial effects of silicon on plant resistance to both abiotic and biotic stresses are well established (Reynolds et al. 2009). For instance, the co-deposition of silicon and metals (e.g. Al, Mn, Cd) in the plant results in reduced concentrations of toxic levels of metal ions in plants (Richmond and Sussman 2003, Tubana et al. 2016). Similarly, silicon fertilization can enhance plant resistance to biotic stresses in the form of plants pathogens and animals (vertebrate and arthropod herbivores) (Datnoff et al. 2009, Reynolds et al. 2009).

Silicon fertilization has been proposed to augment plant resistance against herbivorous pests via two different mechanisms. One mechanism involves deposition of amorphous silica in plant tissues that contributes to the thickening of epidermal layer, thus forming a mechanical barrier. This mechanical barrier results in increased rigidity and abrasiveness of plant tissues, thereby reducing the palatability and digestibility of plants to insect herbivores (Ma 2004,

Massey and Hartley 2009). A second mechanism involves the ability of silicon to stimulate biochemical pathways related to specific plant chemical defenses and to prime hormonemediated defense responses via the jasmonate (JA) signaling pathway (Gomes et al. 2005, Ye et al. 2013). Economically important crops such as sugarcane, rice, and barley are classified as high accumulators of silicon containing 10-100 g kg⁻¹ Si in dry weight (Ma et al. 2001, Ma and Takahashi 2002, Liang et al. 2007).

Rice is one of the most important crops worldwide. It is a staple for nearly half of the world's seven billion people (Mohanty 2013). The United States is one of the largest global producers of rice with an acreage of 1.27 million hectares and production of 14.4 million tons in 2016 (USDA-ERA 2017). Approximately 1.6 M tons of rice were produced in Louisiana in 2016 (USDA-NASS 2017). Rice plants are consistently subjected to various pressures by pests throughout the growing season. The rice water weevil, Lissorhoptrus oryzophilus Kuschel (Coleoptera: Curculionidae), is the most destructive early-season pest of rice in the United States (Way 1990). Adult weevils can inflict minor damage by feeding on leaf tissues, leaving longitudinal scars along the leaf blades (Stout et al. 2002). The majority of the injury, however, is caused by the soil-dwelling, root feeding larvae (Shang et al. 2004). The injury by larvae can result in yield losses exceeding 25% when fields are left untreated (Zou et al. 2004, Reay-Jones et al. 2008). The fall armyworm, Spodoptera frugiperda (Lepidoptera: Noctuidae), is also an early-season pest that periodically infests rice in Louisiana. Female fall armyworm oviposit on young rice leaves and larvae can defoliate entire rice plants when densities are high (Pantoja et al. 1986, Stout et al. 2009).

Stem boring lepidopteran pests also attack rice from seedling to maturity (Akinsola 1984) but infestations are sporadic in Louisiana. The sugarcane borer, *Diatraea saccharalis* F

(Lepidoptera: Crambidae), is a major agronomic pest in the southern United States and an economically important pest in sugarcane, corn, rice, and sweet sorghum (Roe et al. 1981). Rice stalk borer, *Chilo plejadellus* Zincken (Lepidoptera: Crambidae), is a sporadic but a long-established stem-boring pest in Louisiana. Moreover, another stem boring species, the Mexican rice borer, *Eoreuma loftini* Dyar (Lepidoptera: Crambidae), has invaded Louisiana crops in the past decade (Wilson et al. 2015, 2017) and is predicted to cause significant economic losses once fully established (Reay-Jones, Wilson, et al. 2008).

Lastly, rice stink bug, *Oebalus pugnax* F. (Hemiptera: Pentatomidae) is considered the primary late season pest of rice in Louisiana. Rice stink bugs can cause damage to rice by feeding on developing grains at the flowering, grain filling, milk, and dough stages of grain development. Feeding by the rice stink bug does not cause significant yield losses under most circumstances but greatly affects the quality of rice (Tindall et al. 2005, Espino 2007).

In the literature, most studies assess the effect of silicon on an individual pest or agronomic trait. There's also a need to consider the effects of silicon under field condition because a lot of these studies were done extensively in the greenhouse and laboratory (Reynolds et al. 2016). This study investigated the independent and combined effects of soil silicon amendment and nitrogen fertilization on injury by rice insect pest complex and direct effect on the morphology (stem hardness) of rice plants. More specifically, experiments conducted in the field were performed to characterize the effects of nitrogen levels, soil silicon amendment, and variety on rice water weevil densities, whitehead incidences, rice stink bug densities, and yields. In greenhouse experiments, effects on stem hardness, total phenolic concentrations, fall armyworm relative growth rates, and sugarcane borer performance were evaluated. This is one of

the few studies that have examined the effects of soil silicon amendment on multiple pests in rice both in natural field infestations and greenhouse experiments.

2.2. Materials and Methods

2.2.1. Field experiments

Field experiments were conducted from 2015 to 2017 at the H. Rouse Caffey Rice Station in Crowley, Louisiana (30°14'22.406''N, 92°20'46.195''W, 7 m asl) to investigate the interactive effects of nitrogen levels, soil silicon amendments, and variety on rice water weevil densities, whitehead incidences, rice stink bug densities, and yields. All experiments employed a randomized complete block design with four replications. In 2015 and 2016, each block consisted of 16 plots subjected to factorial combinations of two varieties (Cocodrie and CL161), four levels of nitrogen (0, 67, 101 and 135 kg N/ha), and two levels of Ca₂SiO₄ slag as a silicon source (0 and 5000 kg/ha). In 2017, each block consisted of 18 plots subjected to factorial combinations of two varieties (Jupiter and CL261), three nitrogen levels (70, 135, and 200 kg N/ha), and three levels of Ca₂SiO₄ slag (0, 3000, and 5000 kg/ha).

In 2015, rice seeds were water-seeded into 1.5 m x 3 m plots at a rate of 170 kg/ha. In 2016 and 2017, rice was drill-seeded into 1.2 m x 5.5 m plots at a rate of 70 kg/ha. For plots assigned to the silicon treatment, calcium silicate slag was evenly spread on the soil surface of the plots immediately after planting, except in 2015, when slag was applied a few weeks after flood-seeding to allow the water to drain and for the soil to dry. After planting, fields were surface irrigated as needed until permanent flooding to facilitate plant emergence. Table 2.1 lists the dates of planting and slag application for the three years of the study. Nitrogen was applied in the form of urea four to five weeks after planting (Table 2.1). Urea was evenly hand-distributed on each plot at full rate (no-split) on dry soil before establishment of permanent flood. No

insecticides were sprayed on the plots for the duration of the experiment to facilitate natural infestations of rice water weevil, stem borers, and rice stink bugs.

At grain maturity, entire plots were harvested by a mechanical harvester. Grain yields from each plot were adjusted to 12% moisture

Table 2.1. Field Activities and corresponding dates									
Activity	Year								
	2015	2016	2017						
Planting	27 May	6 April	28 April						
Silicon Slag Application	18 June	6 April	28 April						
Nitrogen Fertilization	23 June	4 May	19 June						
Permanent Flood	24 June	5 May	20 June						
1 st Core Sampling	13 July	30 May	5 July						
2 nd Core Sampling	20 July	6 June	11 July						
Whitehead counts	4 September	5 July	16 August						
Stinkbug counts	18 August	8 July	-						
Harvest	_	11 August	19 September						

2.2.1.a. Rice water weevil densities

Densities of rice water weevil (larvae and pupae) were determined using a metal soil/root core sampler with a 10 cm diameter and a 10 cm depth. Flooding triggers female weevils to oviposit (Stout et al. 2002); thus, core samplings were performed approximately three and four weeks after permanent flood (Table 2.1). For each sampling date, two to three core samples were taken from the interior portion of each plot. Every core sample contained a minimum of one rice plant with intact roots. Core samples were processed individually by washing the soil from roots in 40-mesh screen sieve buckets. Larvae were counted as they floated in the sieve buckets when dipped on basins with salt solution (N'guessan and Quisenberry 1982). The larval density in each plot was estimated by calculating the average number of larvae from two to three core samples from each plot.

2.2.1.b. Whitehead incidence

Whiteheads resulting from infestations by stem borer larvae in rice at the reproductive stages was selected as an indicator of stem boring infestation. Total numbers of whiteheads were counted at 100 percent heading in each plot. Whiteheads were transported to a laboratory and dissected to identify stem borer species. Sugarcane borer was found to be the major cause of whiteheads in 2015 and Mexican rice borer was the major cause of whiteheads in 2016 and 2017. 2.2.1.c. Rice stink bug densities

Rice stink bugs feed on the sap of developing rice grains. Insect sweep nets (38 cm diameter) were used to determine total number of stink bugs in a given area. In each plot, ten 180° sweeps of rice canopy were performed at grain filling stage. Densities of stink bug (number of stink bug per plot) were estimated by counting the total number of adults and nymphs from ten sweeps in each plot.

2.2.1.d. Analysis of field data

Rice water weevil larval densities, whitehead incidences, rice stink bug densities, and yields in 2015, 2016, and 2017 were analyzed separately as factorial RBD experiments with block as random effect and variety, nitrogen rates, soil silicon amendment, and their interactions as fixed effects using mixed model analysis of variance in PROC MIXED of SAS (SAS 2013). Larval densities from the two core sampling dates in each year were also analyzed separately. Means were separated using Tukey's HSD test (Tukey 1953). Residuals were also analyzed for normality using PROC UNIVARIATE (SAS 2013). Transformations to normalize data distribution to satisfy statistical normality assumptions were performed as necessary, but untransformed data are presented. Transformations were performed on the following data sets using either square or cube root: core 1 and core 2 in 2015; core 2, whitehead incidence, and rice

stink bug densities in 2016. Kenward-Rogers adjustment for degrees of freedom in mixed model was applied in all analyses (Littell et al. 2002).

2.2.2. Greenhouse studies

Greenhouse experiments were conducted on the campus of Louisiana State University, Baton, Rouge, LA to investigate the effects of nitrogen level, soil silicon amendment, variety, and their interactions on stem hardness, total extractable phenolic compounds in rice leaves, fall armyworm relative growth rates, sugarcane borer relative growth rates, and boring success. Planting were staggered every two weeks to ensure a continuous supply of plant materials at desired growth stages. All greenhouse experiments employed a randomized complete block design with five replications. Each block consisted of 16 pots subjected to factorial combinations of two varieties (Cocodrie and CL 161), four levels of nitrogen (0, 67, 101 and 135 kg N/ha), and two levels of Ca₂SiO₄ slag (0 and 5000 kg/ha). The 15 cm (diameter) pots were filled with mixed soil (2:1:1, top soil: peat moss: sand) and five to six rice seeds were sown on each pot. After sowing, silicon slag was evenly spread on the soil surface. Five days after emergence, pots were thinned to one plant per pot. Nitrogenx was applied 20 days after emergence. Plants received tap water as often as necessary. Plants were grown to desired developmental stages as required for each experiment.

2.2.2.a. Culm strength

The force required to penetrate rice stems was measured using a handheld digital force gauge (Dillon GL, Athens, Texas). The digital force gauge was mounted to a test stand with a wheel rotation capacity (Dillon CT, Athens, Texas) to ensure accurate measurement. Rice plants at maximum tillering stage (approximately 60 days after sowing) were used in this experiment. Intact plants were collected from the greenhouse and brought back to the laboratory. The primary

tiller was marked at three different points: 5, 10, and 15 cm above soil line. A metal cone point accessory was attached to the force gauge and was aligned to the marked point on the stem. The force gauge was slowly lowered until the metal cone point pierced halfway through the stem. Reading at each point was recorded as the force required (kg·m·s⁻²) to penetrate the stem. This experiment was repeated twice in 2015. Data from the two replicate experiments were pooled. 2.2.2.b. Total phenolic compounds

Total phenolic compounds in rice leaves were estimated using the Folin-Ciocalteu method described by Singleton et al. (1999). Rice leaves were collected at the tillering stage (30-45 days after sowing) from field and greenhouse plants in 2015. About three to five leaves were randomly collected from each treatment in field and greenhouse rice plants and transported to the laboratory. Leaves were cut into two-cm pieces and were placed in scintillation vials with five mL 50% MeOH and were left for 24-48 h to extract phenolic compounds from leaves. For the colorimetric assay, 100 μ L of phenolic extract was placed in a 20 mL test tube. Deionized water was added to the test tube to a final solution volume of 2.75 mL. The solution was oxidized with 500 μ L 1:1 Folin-Ciocalteu reagent dissolved in water (Sigma Aldrich, St. Louis, Missouri). After eight minutes, the reaction was neutralized with 500 μ L Na₂CO₃. The solution was gently mixed in a vortex mixer and was let to stand for one hour. Solutions were transferred to disposable cuvettes and absorbance at 720 nm was measured with a VMR UV-6300PC Double Beam Spectrophotometer at 720 nm. A standard curve was generated using ferulic acid. 2.2.2.c. Fall armyworm relative growth rates

Fall armyworm larvae used in this experiment were obtained from a colony maintained continuously on artificial diet (Southland Products Inc., Lake Village, Arkansas) in laboratory at Louisiana State University. Larvae collected from the rice fields and pastures at Crowley, LA

were added annually to the laboratory colony to maintain genetic variability and vigor. Rice leaves used in the feeding assays were collected from the greenhouse at early tillering stage (30-45 days after sowing). About three random leaves were collected from each pot and were brought back to laboratory immediately. Fall armyworm were stage-synchronized to third instars and larvae of similar sizes were selected and starved for three hours and then weighed (initial weight). A single larva was then left to feed for seven days on leaves that were placed in petri dishes lined with moist filter paper. Leaves were replaced every two to three days. After seven days, larvae were taken off food and starved for another three hours to clear digestive tracts prior to weighing (final weight). Relative growth rates of fall armyworm larvae were calculated using the formula:

$$RGR = \frac{\text{Larval Final weight - Larval Initial weight}}{\left(\frac{\text{Final weight + Initial weight}}{2}\right) \times \text{Number of feeding days}}$$

(Waldbauer 1968). The feeding assay was repeated five times in 2015.

2.2.2.d. Sugarcane borer performance

Sugarcane borer larvae used in this experiment were obtained from a colony maintained continuously in a laboratory at Louisiana State University following the methods of Martinez et al. (1988). The colony originated from larvae collected in rice fields from Crowley, LA and eggs purchased from Benzon Inc. (Carlisle, PA). Sugarcane borer performance was measured by relative growth rates and boring success. Rice plants collected from the greenhouse at maximum tillering (45-60 days after sowing) were used for the assays. For feeding assay, stage-synchronized third instar sugarcane borer larvae of similar sizes were selected and starved for three hours and then weighed (initial weight). The primary tiller from each pot were collected and cut into 10-cm pieces and placed on petri dish lined with moist filter paper. A single larva

was left to feed on the cut stem. After seven days, the stem was dissected to remove the larva and the larva was starved for three hours to clear digestive tracts prior to weighing (final weight). Relative growth rates were calculated using the same formula used for the fall armyworm. For the boring success assay, a primary tiller was collected from each pot, cut to 25 cm, and placed in 50-mL test tubes. Both ends of primary tillers were covered with parafilm to prevent sugarcane borer to enter through the cut ends. Ten third instar sugarcane borer larvae were then released inside the test tubes and were left to bore for 72 hours. To prevent escape, the opening of test tube was covered with parafilm. After 72 hours, entry holes with frass coming out of them were counted on each stem. Stem diameter was also measured using a digital caliper (General tools, Secaucus, New Jersey).

2.2.2.e. Analysis of greenhouse data

Stem hardness, total phenolic compounds, fall armyworm relative growth rates, sugarcane borer relative growth rates, and boring success were analyzed separately as factorial RBD experiments with block as random effect and variety, nitrogen rates, soil silicon amendment, and their interactions as fixed effects using the mixed model analysis of variance in PROC MIXED (SAS 2013). Means were separated using Tukey's HSD test (Tukey 1953). Residuals were analyzed for normality using PROC UNIVARIATE (SAS 2013). Transformations to normalize data distribution were performed as necessary, but untransformed data are presented. Transformations were performed on the following data sets using either square or cube root: stem hardness, fall armyworm relative growth rates, sugarcane borer relative growth rates, and boring success. Kenward-Rogers adjustment for degrees of freedom in mixed model was applied in all analyses (Littell et al. 2002). The relationship between stem diameter

and number entry holes were also examined using Pearson correlation coefficients in PROC CORR (SAS 2013).

2.3. Results

2.3.1. Field studies

2.3.1.a. Effects on rice water weevil densities

Core sampling was performed three and four weeks after permanent flooding in 2015, 2016, and 2017. Data were analyzed separately by core sampling and year. Effects of nitrogen levels on weevil larval densities were significant (P<0.05) both in the first and second core samplings in 2015, 2016, and 2017 (Table 2.2). Higher larval densities were observed in nitrogen treated plots compared to untreated plots in 2015 and 2016 (Figure 2.1A, D). In 2017, larval densities were found to be lower in plots with a nitrogen rate of 70 kg N/ha compared to 135 and 200 kg N/ha (Figure 2.2A). The effect of variety on weevil densities was only significant in two sampling dates (P<0.05), the first core sampling in 2015 and second core sampling in 2017 (Table 2.2). On those sampling dates, the long-grain variety 'Cocodrie' and the medium-grain variety 'Jupiter' supported higher densities of weevil larvae (Figures 2.1C and 2.2C) than the long-grain Clearfield variety 'CL161' and the medium-grain Clearfield variety 'CL261', respectively. In contrast, soil silicon amendment did not affect weevil larval densities (P>0.05) on any sampling dates (Table 2.2; Figures 2.1B, E and 2.2B). In 2015, significant interaction was observed between variety and nitrogen (Table 2.2). This interaction might have been caused by high numbers of weevil larvae on nitrogen treated plots in 'Cocodrie' compared to 'CL161'. In 2017, interaction between nitrogen and silicon was significant (Table 2.2). Highest density of weevil larvae was observed on plots treated by both silicon at a rate of 3000 kg slag/ha and nitrogen at a rate of 200 kg N/ha.

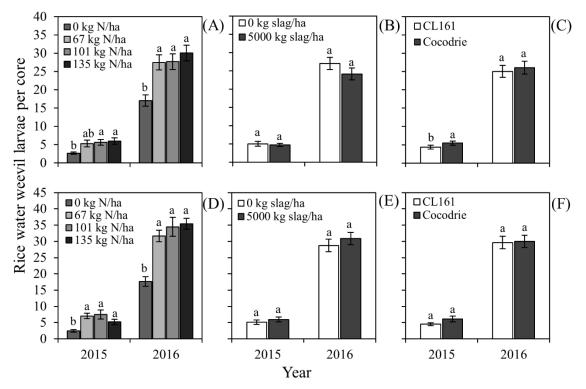


Figure 2.1. Main effects of nitrogen rates, soil silicon amendment, and variety on rice water weevil larval densities in 2015 and 2016, Crowley, LA. Core 1 (A, B, and C) and Core 2 (D, E, and F) sampling were performed three and four weeks, respectively, after permanent flooding. (A and D), main effect of nitrogen rates; (B and E), main effect of soil silicon amendment; (C and F), main effect of variety. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

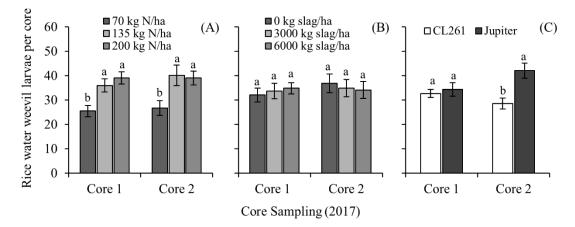


Figure 2.2. Main effects of nitrogen rates, soil silicon amendment, and variety on rice water weevil larval densities in 2017, Crowley, LA. Core sampling were performed three and four weeks after permanent flooding. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

Table 2.2. Statistical comparisons of the effects of variety (Var), soil silicon amendment (Sil), nitrogen rates (N), and their interactions on rice water weevil larval densities. First and second sampling was performed approximately three and four weeks after permanent flooding in 2015, 2016, and 2017. Analysis were separated between year and core sampling.

	1 st Core Sampling									
Treatment	2015			2016			2017			
	df	F	Р	df	F	Р	df	F	Р	
Var	1, 47	4.12	0.0482*	1, 47	0.30	0.5888	1, 53	0.38	0.5390	
Sil	1, 47	0.04	0.8437	1, 47	2.25	0.1400	2, 53	0.36	0.6966	
Var x Sil	1, 47	2.32	0.1341	1, 47	1.45	0.2345	2, 53	2.67	0.0785	
Ν	3, 47	5.56	0.0024*	3, 47	9.36	0.0001*	2, 53	9.47	0.0003*	
Var x N	3, 47	5.31	0.0031*	3, 47	1.14	0.3428	2, 53	0.52	0.5968	
Sil x N	3, 47	0.39	0.7633	3, 47	0.87	0.4624	4, 53	3.59	0.0115*	
Var x Sil x N	3, 47	2.86	0.0470*	3, 47	0.14	0.9376	4, 53	1.04	0.3933	
	2 nd Core Sampling									
Var	1, 47	3.73	0.0596	1, 47	0.03	0.8637	1, 53	19.55	0.0001*	
Sil	1, 47	1.14	0.2908	1, 47	1.40	0.2431	2, 53	0.28	0.7560	
Var x Sil	1, 47	0.52	0.4764	1,47	0.00	0.9527	2, 53	0.84	0.4367	
Ν	3, 47	8.82	0.0001*	3, 47	21.75	0.0001*	2, 53	7.86	0.0010*	
Var x N	3, 47	1.07	0.3719	3, 47	2.58	0.0643	2, 53	1.16	0.3226	
Sil x N	3, 47	0.50	0.6873	3, 47	0.44	0.7279	4, 53	1.60	0.1871	
Var x Sil x N	3, 47	0.18	0.9080	3, 47	0.84	0.4811	4, 53	1.84	0.1349	
*Significant at P<0.05										

2.3.1.b. Effects on whitehead incidences

Whiteheads were used as indicator of stem borer activity in the field. Densities of whiteheads (whiteheads per plot) were assessed and were subsequently collected and dissected to identify stem borer species. Sugarcane borer and Mexican rice borer were the predominant cause of whiteheads in the field. Infestation of stem borers in 2017 was remarkably high compared to past years. Approximately 50 whiteheads were collected per plot on average in 2017. In this study, effects of nitrogen rates on whitehead densities were not significant in 2015 (F = 0.80; df = 3, 31; P = 0.5055) and 2016 (F = 2.16; df = 3, 47; P = 0.1057) but were marginally significant in 2017 (F = 3.22; df = 2, 26; P = 0.0562) (Figures 2.3A and 2.4A). Although a lower incidence of whiteheads was observed in plots with nitrogen rate of 70 kg N/ha than in plots with nitrogen rates of 135 kg N/ha and 200 kg N/ha, post-hoc analysis (Tukey's HSD) revealed no significant

differences between nitrogen rates (Figure 2.3A). Significantly higher whitehead incidence was also observed in plots planted with 'Cocodrie' compared to plots planted with 'CL161' in 2015 (F = 39.00; df = 1, 31; P <0.0001) but not in 2016 (F = 1.78; df = 1, 47; P = 0.1883) (Figure 2.3C). Soil silicon amendment did not affect whitehead densities in 2015 (F = 3.18, df = 1, 31; P = 0.0842), 2016 (F = 0.38; df = 1, 47; P = 0.5426), or 2017 (F = 0.04; df = 2, 26; P = 0.9645) (Figures 2.3B and 2.4B). Due to poor development of the medium-grain variety 'Jupiter' in the field caused by South American rice miner and fall armyworm attack and uneven flooding, the effect of variety on whiteheads was not included in the 2017 analysis. There were no significant interactions between nitrogen rates, soil silicon amendment, and variety (data not shown).

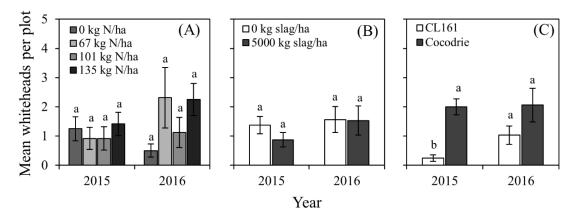


Figure 2.3. Main effects of nitrogen rates, soil silicon amendment, and variety on whiteheads in 2015 and 2016, Crowley, LA. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

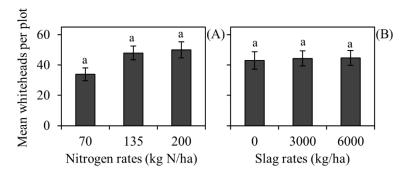


Figure 2.4. Main effects of nitrogen rates and soil silicon amendment on whiteheads in 2017, Crowley, LA. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

2.3.1.c. Effects on rice stink bug densities

Densities of rice stink bugs (total number of stink bugs per plot) were assessed by using insect sweepnets in 2015 and 2016. Numbers of adults and nymphs were combined for analysis since both developmental stages feed on the sap of the developing rice grain. Nitrogen rates did not affect densities of stinkbugs in 2015 (F = 0.84; df = 3, 32; P = 0.4829) or 2016 (F = 1.92; df = 3, 47; P = 0.1399) (Figure 2.5A). Similarly, no significant differences were observed among silicon treatments for stinkbug densities in 2015 (F = 0.07; df = 1, 32; P = 0.7986) or 2016 (F = 0.22; df = 1, 47; P = 0.3486) (Figure 2.5B). Significantly higher stinkbug density was observed in plots planted with 'Cocodrie' compared to plots planted with 'CL161' in 2016 (F = 14.78; df = 1, 47; P = 0.0004) but not in 2015 (F = 0.26; df = 1, 32; P = 0.6104) (Figure 2.5C). There were no interactions observed between nitrogen rates, soil silicon amendment, and variety (data not shown).

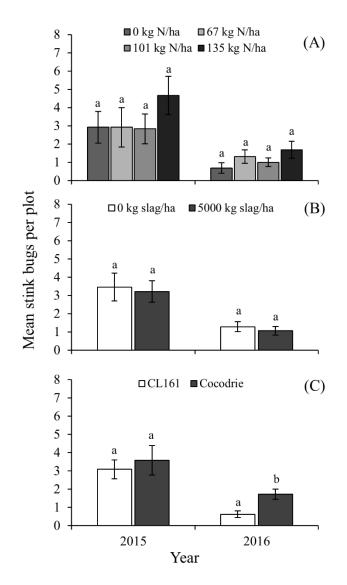


Figure 2.5. Main effects of nitrogen rates, soil silicon amendment, and variety on rice stink bugs densities in 2015 and 2016, Crowley, LA. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

2.3.1.d. Effects on rice yield

Effects of nitrogen rates on rice yields were significant in 2016 (F = 4.30; df = 3, 47; P = 0.0092) but not in 2017 (F = 2.84; df = 2, 26; P = 0.0765). In 2016, yields increased with increasing nitrogen rates (Figure 2.6). In contrast, soil silicon amendment did not affect yields in 2016 (F = 0.35; df = 1, 47; P = 0.5554) or 2017 (F = 1.63; df = 2, 26; P = 0.2146) (Figure 2.6).

Variety also did not affect rice yields in 2016 (F = 3.09; df = 1, 47; P = 0.0851) (data not shown). Effects of variety on yields in 2017 were not included in the analysis due to poor development of the medium-grain variety 'Jupiter' as mentioned earlier. Yields from 2015 were not included in the analysis because it was manually harvested late in the season. There were no interactions observed between nitrogen rates and soil silicon amendment (data not shown).

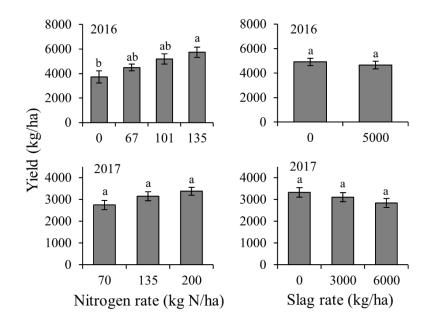


Figure 2.6. Main effects of nitrogen rates and soil silicon amendment on rice yields in 2016 and 2017, Crowley, LA. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

2.3.2. Greenhouse studies

2.3.2.a. Effects on stem toughness

Stem borers can penetrate rice stems just a few days after feeding on the leaf sheaths.

This experiment was conducted to quantify the force required to penetrate rice stem at maximum

tillering in three marked positions (5, 10, and 15 cm distances above soil). Data analysis was

performed separately for each position. Nitrogen rate did not affect the force required to

penetrate rice stems at 5 cm (F = 1.53; df = 3, 143; P = 0.20970, 10 cm (F = 2.33; df = 3, 144; P = 0.0769), or 15 cm (F = 1.78; df = 3, 144; P = 0.1540) (Figure 2.7A). In contrast, there was a significant effect of soil silicon amendment on force required to penetrate rice stems at 10 cm (F = 9.87; df = 1, 144; P = 0.0020) and 15 cm (F = 5.92; df = 1, 144; P = 0.0162) but not at 5 cm (F = 1.51; df = 1, 43; P = 0.2217). Higher force was required to penetrate rice stems when soils were amended with silicon slag compared to untreated soils (Figure 2.7B). Higher force was required to penetrate the stem for 'CL161' compared to 'Cocodrie' at 5 cm (F = 16.06; df = 1, 143; P = 0.0001) and 10 cm (F = 4.34; df = 1, 44; P = 0.0391) but not at 15 cm (F = 0.50; df = 1, 44; P = 0.4795) (Figure 2.7C). There were no significant interactions observed between nitrogen rates, soil silicon amendment, and variety (data not shown).

2.3.2.b. Effects on total phenolic concentration

Significantly higher concentrations of phenolic compounds were consistently observed in rice leaves grown with no added nitrogen compared to nitrogen treated plants in the field (F = 5.98; df = 3, 175; P = 0.0007) and greenhouse (F = 10.19; df = 3, 140; P < 0.0001) (Figure 2.8A). Total phenolic compounds were also significantly higher in 'CL161' compared to 'Cocodrie' in the field (F = 7.29; df = 1, 175; P = 0.0076) but not in the greenhouse (F = 0.49; df = 1, 140; P = 0.4839) (Figure 2.8C). In contrast, soil silicon amendment did not affect concentrations of phenolic compounds in rice leaves collected in the field (F = 2.98; df = 1, 175; P = 0.0860) or greenhouse (F = 0.15; df = 1, 140; P = 0.6975) (Figure 2.8B). There were no interactions of nitrogen rates, soil silicon amendment, and variety (data not shown).

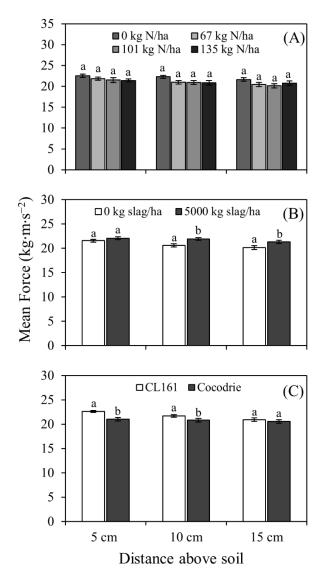


Figure 2.7. Main effects of nitrogen rates, soil silicon amendment, and variety on the force required to penetrate the rice stems at a distance of 5, 10, and 15 cm above soil. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety in 2015. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

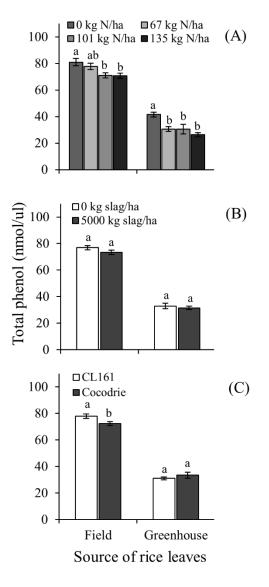


Figure 2.8. Main effects of nitrogen rates, soil silicon amendment, and variety on total phenolic compounds in rice leaves collected from the field and greenhouse. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

2.3.2.c. Effects on fall armyworm relative growth rates

Relative growth rates of fall armyworm were determined through feeding assays. Assays were performed in 2016 and 2017 and data from these assays were analyzed separately by year. Relative growth rates of fall armyworm were significantly lower on plants without nitrogen

compared to nitrogen treated plants in 2015 (F = 21.66; df = 3, 219; P < 0.0001) and 2016 (F = 14.35; df = 3, 45; P < 0.0001) (Figure 2.9A). Higher relative growth rates were also observed on fall armyworm feeding on 'Cocodrie' compared to 'CL161' in 2016 (F = 5.00; df = 1, 45; P = 0.0304) but not in 2015 (F = 0.10; df = 1, 219; P = 0.7563) (Figure 2.9C). In contrast, soil silicon amendment did not affect relative growth rates in 2015 (F = 0.07; df = 1, 219; P = 0.7945) or 2016 (F = 1.05; df = 1, 45; P = 0.3102) (Figure 2.9B). There were no significant interaction between nitrogen rates, soil silicon amendment, and variety (data not shown).

2.3.2.d. Effects on sugarcane borer performance

Assays were performed to determine the performance of sugarcane borer by investigating relative growth rates and boring success. Relative growth rates were assessed through feeding assays and data were analyzed separately by year. Relative growth rates of sugarcane borer that fed on rice plants without nitrogen were significantly lower compared to plants with nitrogen in 2015 (F = 30.1; df = 3, 56; P < 0.0001) and 2016 (F = 20.76; df = 3, 46; P < 0.0001) (Figure 2.10A). In 2016, relative growth rates increased when levels of applied nitrogen were also increased (Figure 2.10A). Unexpectedly, relative growth rates were significantly higher on silicon treated plants compared to untreated in 2016 (F = 8.29; df = 1, 46; P = 0.0060) but not in 2015 (F = 2.05; df = 1, 56; P = 0.1577) (Figure 2.10B). Moreover, variety did not affect relative growth rates in 2015 (F = 0.30; df = 1, 56; P = 0.5856) or 2016 (F = 0.24; df = 1, 46; P = 0.6236) (Figure 2.10C).

Performance of sugarcane borer was also measured through boring success. Entry holes were used as positive indicator of boring success. The number of second instar larvae that bored into the stem within 72 h (indicated by entry holes) differed significantly by nitrogen level (F = 9.94; df = 3, 64; P < 0.0001) but not soil silicon amendment (F = 1.67; df = 1, 64; P = 0.2012) or

variety (F = 1.19; df = 1, 64; P = 0.2796) (Figure 2.11). Stem diameter was also measured to evaluate correlation between plant diameter and boring success. Results revealed a positive linear relationship between entry holes and stem diameter (r = 0.54; P < 0.0001) (Figure 2.12).

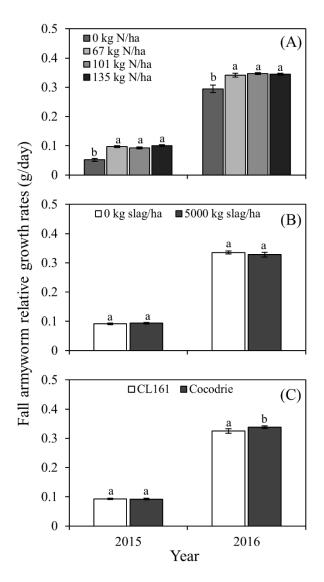


Figure 2.9. Main effects of nitrogen rates, soil silicon amendment, and variety on the relative growth rates of fall armyworm in 2015 and 2016. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

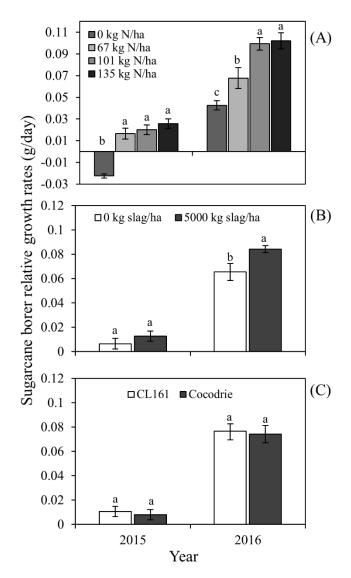


Figure 2.10. Main effects of nitrogen rates, soil silicon amendment, and variety on the relative growth rates of sugarcane borer in 2015 and 2016. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety. For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

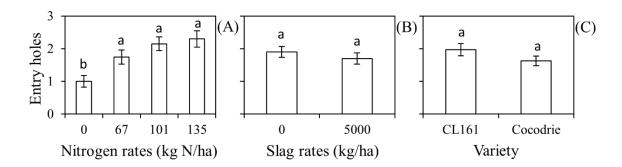


Figure 2.11. Main effects of nitrogen rates, soil silicon amendment, and variety on the boring success of sugarcane borer on cut stems. (A), main effect of nitrogen rates; (B), main effect of soil silicon amendment; (C), main effect of variety in 2016 For each year, bars accompanied by the same letter represent means that do not significantly differ (P>0.05; Tukey's HSD). Error bars represent standard error of the means (SEM).

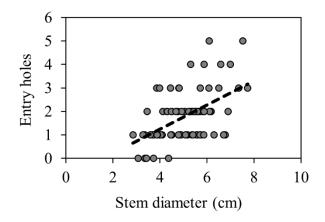


Figure 2.12. Linear relationship between the diameter of rice stems and sugarcane borer entry holes. Correlation was examined using Pearson correlation coefficients (r = 0.54; p < 0.0001).

2.4. Discussion

Louisiana rice is consistently subjected to complexes of pests at different developmental stages. The insect pest complex reported in Louisiana are the rice water weevil (major early-season insect pest), rice stink bug (major late-season pest), sugarcane borer and rice stalk borer (long- established but sporadic stem boring pests), Mexican rice borer (an invasive stem-boring pest), and fall armyworm (sporadic early-season pest). This study was conducted to investigate the effects of nitrogen levels, soil silicon amendment, variety, and their interactions on the insect

pest complex in rice under both field and greenhouse conditions. This study is among the few to investigate the effects of soil silicon amendment on multiple insect pests under natural infestations under field conditions. There is also an apparent lack of studies on the effect of silicon on below-ground herbivores (Reynolds et al. 2016), which this study attempted to address.

In this study, we found that high nitrogen fertilization levels significantly increased the infestations of the root-feeding rice water weevil larvae in the field in 2015, 2016 and 2017 (Figures 2.1 and 2.2), consistent with the observations made in previous studies (Bowling 1963, Way et al. 2006b). Effect of variety on the density of weevil larvae was also observed in two instances (Table 2.2). Although we only used two varieties in each of the experiment, previous studies have shown that some rice varieties commonly grown in Louisiana do differ in their susceptibility to rice water weevil infestations and their tolerance of weevil feeding; however, none of the varieties possess high level of resistance (Stout et al. 2001).

On the other hand, soil silicon amendment did not affect the densities of rice water weevil larvae in the field (Table 2.2). A recent study conducted in Texas reported that the effect of silicon on densities of rice water weevil larvae was weaker than the effects of variety or seed treatments in field experiments (Villegas et al. 2017). The lack of effect of soil silicon amendment on weevil larval densities in this study might be attributed to the phenology of attack by weevils on rice.

Typically, older plants have higher silicon content compared to younger plants due to continual uptake and immobility of silicon once deposited in plant tissues (Ishizuka 1964, Epstein 1994). Drill-seeded rice fields are permanently flooded when the rice reaches the four- to five-leaf stages and flooding triggers the oviposition of rice water weevil (Stout et al. 2002).

Since rice water weevil attack early in the planting season, accumulation of silicon on rice plants may not have reached the level that would consistently influence the attack. Furthermore, soil properties such as pH, organic matter, and clay content can highly influence the amount of monosilicic acid released from calcium silicate slag (Tubana et al. 2016). We did not estimate the nitrogen levels or silicon levels in the plant tissues in all of our experiments; thus, amounts of silicon assimilated by the rice plants are unknown. In contrast, a study conducted in a glasshouse chamber reported reductions on the performance of the root-feeding larvae 'canegrubs' in response to silicon treatment on sugarcane plants (Frew et al. 2016).

Results obtained from the field and greenhouse experiments also revealed significant effects of nitrogen fertilization on whitehead incidence and sugarcane borer performance. To our knowledge, this study is the first to report an effect of nitrogen fertilization on sugarcane borer and Mexican rice borer in U.S. rice. In the field, where both Mexican rice borer and sugarcane borer were identified as the major cause of whiteheads, increases in nitrogen fertilization led to marginally significant increases in whitehead incidences in 2017 (Figure 2.4). The absence of effect of nitrogen on whitehead incidences in 2015 and 2016 might have been influenced by extremely low natural infestations of stem boring pests in the field during those years. In greenhouse experiments, relative growth rates and boring success of sugarcane borer were also significantly affected by nitrogen rates. Higher rates of nitrogen applied to rice plants increased the relative growth rates of sugarcane borer in 2015 and 2016 (Figure 2.10). Similarly, entry holes were significantly higher on rice plants when nitrogen was applied (Figure 2.11). Earlier reports on the effects of nitrogen on the stem boring pests were conducted on bioenergy sorghum and sugarcane but not in rice. Higher rates of nitrogen led to higher injuries from Mexican rice borer on bioenergy sorghum (VanWeelden et al. 2016). Likewise, increasing rates of nitrogen in

sugarcane also increased the injuries from sugarcane borer (Lopez et al. 1983). Effects of variety on stem borer activity were more pronounced in the field compared to the greenhouse experiments. Whitehead incidence was consistently lower on 'CL161' than 'Cocodrie' but this effect was only significant in 2015 (Figure 2.3). In contrast, variety did not affect the relative growth rates of sugarcane borer in 2015 and 2016 (Figure 2.10) nor did it affect boring success (Figure 2.11). Results also showed a positive linear relationship between entry holes and rice stem diameter (Figure 2.12). Previous field studies reported the presence of varying levels of injury and yield losses from stem borer infestations on multiple rice cultivars (Way et al. 2006a). Moreover, studies conducted in Louisiana also reported variation in larval performance and oviposition preference of sugarcane borer on commonly grown rice varieties (Hamm et al. 2011, Sidhu et al. 2013).

Soil silicon amendment did not affect whitehead incidences in the field in 2015, 2016, or 2017 (Figures 2.3, 2.4). Similarly, effects of soil silicon amendment on relative growth rates and boring success of sugarcane borer in greenhouse experiments were not significant (Figure 2.10 and 2.11). In contrast, force required to penetrate rice stems was significantly increased by soil silicon amendment (Figure 2.7). This is one of the few studies that has quantified the effects of silicon on mechanical strength of plant tissues (Massey et al. 2006). The deposition of silica in shoots and plant epidermis has been shown to enhance the plant's mechanical strength and protective layer (Massey et al. 2006, Massey and Hartley 2009) and was proposed as one of the mechanisms that augment plant resistance to insect herbivores (Reynolds et al. 2009). The uptake and deposition of silica to plant tissues contributes to the thickening of the epidermal layer and increases the rigidity and abrasiveness of plant tissues, which reduces the palatability

and digestibility of plants to insect herbivores and promote the resistance of plants to pathogens (Datnoff et al. 2009, Massey and Hartley 2009).

The lack of effect of soil silicon amendment on whitehead incidence and stem borer performance was unexpected because other studies have shown effects of silicon on stem borers in rice. The first report of silicon-induced resistance was associated with resistance against the rice stem borer, Chilo simplex (Sasamoto 1953). Silicon was also documented to enhance plant resistance to several stem boring pests such as yellow stem borer (Panda et al. 1975), Asiatic rice borer (Hou and Han 2010), and sugarcane borer (Sidhu, Stout, Blouin, et al. 2013). In fact, soil silicon amendment reduced the weight gain and stem damage and prolonged the penetration time and larval development of a destructive stem boring rice pest in Asia, the Asiatic rice borer, Chilo suppressalis Walker (Hou and Han 2010). Greenhouse experiments conducted in Louisiana also reported reductions in relative growth rates and boring success of sugarcane borer on rice plants amended with silicon slag (Sidhu et al. 2013). Several factors could be responsible for the lack of effect of soil silicon amendment on stem borers in this study. To begin with, varieties used in the experiments may not have taken up sufficient silicon levels to influence infestations and performance by stem borers. Deren (2001) reported that some rice varieties differ in their ability to accumulate silicon. Relatively low populations in 2015 and 2016 and uneven distributions of stem borers in the field may have also prevented the detection of silicon effects. Moreover, stem borer complex present in a field environment is potentially less sensitive to silicon-induced changes. In the field, it was observed that the sugarcane borer was found to be the major cause of whiteheads in 2015 and the Mexican rice borer in 2016 and 2017

Population of rice stinkbugs were also evaluated in the field. Although feeding of rice stink bugs on developing rice grains does not cause yield losses under most circumstances, it

affects the quality of rice (Tindall et al. 2005, Espino 2007). Results showed no significant differences on stinkbug populations among nitrogen levels and silicon treatments in 2015 and 2016 (Figure 2.5). Although there was no observed effects of soil silicon amendment on stink bug population in rice, this study is the first attempt to investigate potential effects of silicon on rice stink bug density. Higher number of stinkbugs were observed in 'Cocodrie' compared to 'CL161' in 2016 but not in 2015 (Figure 2.5). Studies have shown that resistance to rice stink bugs exist between rice varieties but correlation between resistance and yield appears to be negative (Bernhardt et al. 2004). Studies indicated that rice resistance to stink bug increases with grain length and the rice varieties least affected by stink bug feeding were not the highest yielding (Bernhardt et al. 2004).

Nitrogen fertilization had a significant effect on relative growth rates of fall armyworm. Higher relative growth rates were observed on fall armyworm that were fed on rice plants fertilized with nitrogen compared to untreated rice (Figure 2.9). This is one of the first reports on the effects of nitrogen fertilization on fall armyworm in rice. Fall armyworms remain a sporadic pest of rice in Louisiana but a major pest in other row crops. Damage caused by fall armyworm on drill-seeded rice typically occur at early developmental stages of rice, when nitrogen fertilizer often has not yet been applied. Previous studies have reported that additions of nitrogen increased the susceptibility of corn and other grasses to fall armyworm larval feeding (Wiseman et al. 1973, Chang et al. 1985). Nitrogen levels were also found to affect the phenolic compounds when measured in rice leaves at the same developmental stages used in relative growth rates experiment. There are over 9000 phenolic-based compounds across the plant kingdom in both leaves and roots and these compounds play an essential role in plant defense against herbivores (Mithofer and Boland 2012). Higher phenolic compounds were extracted in rice leaves of plants not fertilized with nitrogen compared to fertilized rice plants from the field and greenhouse (Figure 2.8). In contrast, effects of soil silicon amendment on the relative growth rates of fall armyworm and phenolic concentrations in rice leaves were not significant (Figure 2.9 and 2.8). Since fall armyworm attack early in the season in field condition, we replicated this in the greenhouse by using younger rice plants at early tillering stage (4-6 weeks after sowing). Accumulation of silicon at early tillering stage might not have reach the level that will affect the feeding of fall armyworm. Study conducted by Massey et al. (2006) reported reductions on the relative larval growth rates of African armyworm when left to feed on high silica grasses compared to low silica grasses. However, for their feeding experiments, they used grasses grown at 12-15 weeks which might have been long enough for silicon to accumulate at sufficient levels to influence an effect on the growth rates of African armyworm.

Despite increases in the densities and performance of rice insect pests at higher rates of fertilization, higher yields were associated with higher nitrogen rates. In 2016, the highest yield was obtained by applying nitrogen fertilizer at a rate of 135 kg/ha (Figure 2.6) which was the highest rate used in the field that year. Recommended nitrogen fertilization rates for rice production in Louisiana is 100-145 kg N/ha depending on rice cultivar and planting methods (LSU Agcenter 2016). Decreasing nitrogen rates below recommended rates resulted to lower yields while increasing nitrogen rates above recommended rates did not necessarily translate to higher yields. We failed to observe significant effects of soil silicon amendment on rice yields in 2016 and 2017 (Figure 2.6), although numerous studies have shown increases on crop yields due to silicon amendment. For example, 4.4-6.6 percent increases in yields were observed on silicon amended soils in Texas (Villegas et al. 2017). Moreover, silicon fertilization resulted in yield

increases of up to 17 percent in rice in Japan (Ma and Takahashi 2002) and 9.3 percent in wheat in China (Liang et al. 1994).

In this study, we did not observe any interactions among nitrogen rates and soil silicon amendment on pest densities, rice yields, or plant morphology under field and greenhouse conditions. Recently, a study conducted by Wu et al. (2017) reported interactions between nitrogen and silicon in rice and the subsequent effects on plant resistance toward the brown planthopper. Based on their results, high nitrogen fertilization levels reduced the accumulation of silicon on rice plants due to decreased expression of the major transporter genes (*OsLsi1* and *OsLsi2*) responsible for silicon uptake and transport. Silicon amendment also enhanced rice resistance to brown planthopper (Wu et al. 2017). Moreover, reductions in borer survival and stalk damage were observed when silicon was applied at different nitrogen levels in sugarcane under field conditions (Keeping et al. 2014).

It is important to understand the effects of fertilization and its implications for plant resistance against insect pests. The use of soil silicon amendment can be potentially integrated to pest management programs in rice but should be evaluated in combination with other control strategies. In this study we assessed the effects of nitrogen fertilization, soil silicon amendment, and variety on multiple pests under field conditions and using more specific assays in the greenhouse. This study is among the first to investigate the role of silicon in augmenting plant resistance against natural infestations of multiple insect pests of rice under field conditions. This study is also one of the first to quantify the effects of silicon on mechanical strengths of plant tissues. In this study, the effects of soil silicon amendment were found to be weaker than both the effects of nitrogen levels and variety. Despite the weak effects of soil silicon amendment, it is important to understand the role of silicon amendment as potential component of pest

management in U.S. rice. Silicon could still play an important role in rice production considering documented effects on yield improvement and disease suppression. Based on the results from this study, there is a possibility that effects of soil silicon amendment depend on rice variety and plant-available silicon present in the soil. Further studies that include estimation of silicon and nitrogen levels in rice plant tissues at different developmental stages should be conducted to help interpret mechanisms of the effects of nitrogen and silicon on insect pests and, identify conditions under which soil silicon amendment might be helpful. For future studies, the number of rice varieties for field and greenhouse experiments should also be increased to encompass varieties with different levels of resistance to insect pests and to assess silicon accumulation on current rice varieties.

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Chapter 3. Effects of soil silicon amendment and rice cultivars on rice insect pest complex in drill-seeded rice

3.1. Introduction

Amendments of soil with silicon has been proposed as a tactic to augment plant resistance against phytophagous insect pests (Reynolds et al. 2009). Silicon is the second-most abundant element in soil. Rice is a typical silicon-accumulating graminaceous crop (Ma et al. 2006, Zhao et al. 2010). In the soil solution, silicon is present in the form of silicic acid, H4SiO4, the form of water soluble silicon that can be absorbed by plants. Silicon is taken up by plants by the lateral roots and is transported to the shoot as monosilicic acid, where it is deposited as solid, amorphous, hydrated silica (SiO₂.nH₂O) (Yoshida et al. 1962, Jones and Handreck 1967). The deposition of silicon in plant tissues leads to formation of a thick epidermal cell layer that can make plants less susceptible to biotic and abiotic stresses (Ma 2004). Biotic stressors may come in the form of vertebrate animals, arthropod herbivores, and plant pathogens (Reynolds et al. 2016). Benefits from silicon fertilization, especially on soils low or limiting in this element, include increases in yields and resistance to insect pests and diseases (Alvarez and Datnoff 2001, Ma et al. 2001).

Most plants contain some level of silicon; however, silicon concentrations in the shoot greatly varies among plant species (Ma and Takahashi 2002, Hodson et al. 2005). The difference in silicon accumulation of various plant species has been attributed to the ability of the roots to take up silicon (Mitana and Ma 2005). The variation in silicon concentration within a plant species has been reported in several important crops. For instance, silicon concentration in the shoot of sugarcane grown in the field varied with the variety (Deren 2001). A survey of about 400 cultivars of barley also revealed a large variation of silicon concentration in the grain on

different cultivars (Ma et al. 2003). In rice, *japonica* varieties usually have higher concentration of silicon than *indica* varieties (Deren et al. 1992, Ma et al. 2007). In a field experiment, 18 rice cultivars varied significantly in their percentage of silicon in rice straw (Deren et al. 1992).

In the United States, Louisiana is the third largest rice-producing state with a total production of 1.6 million tons of rice in 2016 (USDA-NASS 2017). Rice plants are vulnerable to biotic and abiotic stresses and are subjected to various pressures by pests and diseases throughout the growing season. Consistently, the rice water weevil, *Lissorhoptrus oryzophilus* Kuschel (Coleoptera: Curculionidae), is the most damaging and widely distributed early-season insect pest of rice in the United States (Way 1990, Aghaee and Godfrey 2014). This pest has recently invaded rice-producing regions of Asia and Europe and thus poses a global threat to rice production (Saito et al. 2005). Adult weevils feed on leaf tissues, leaving longitudinal scars along the leaf blades (Stout et al. 2002). Injury from adult feeding is generally not economically important. However, root pruning by the soil-dwelling larvae can cause extensive injury to the root systems causing yield losses due to reduction in tillers and grains per panicle at maturity (Zou et al. 2004, 2004a). Weevil larval infestations can result in yield losses exceeding 25% when fields are left untreated and can be higher under heavy weevil pressure (Zou et al. 2004a, Reay-Jones et al. 2008).

Complexes of stem boring Lepidopteran pests also attack rice plants from seedling to maturity (Akinsola 1984). The sugarcane borer, *Diatraea saccharalis* F. (Lepidoptera: Crambidae), and, sporadically, the rice stalk borer, *Chilo plejadellus* Zincken (Lepidoptera: Crambidae), have been reported to be economically important pests of rice in Texas and Louisiana (Bowling 1967, Roe et al. 1981). Furthermore, the Mexican rice borer, *Eoreuma loftini* Dyar (Lepidoptera: Crambidae), has invaded Louisiana in the past decade (Wilson et al. 2015,

2017) and is predicted to cause significant economic yield losses once fully established (Reay-Jones et al. 2008). Generally, stem borer larvae will feed on the leaf sheath for a few days before boring to the stem. When feeding occurs at reproductive stages of the rice plant, injury on the growing plant parts from the base dries the panicles which may emerge but do not produce grains and remain straight and are whitish, a condition known as a whitehead (Pathak and Khan 1994). Stem borer activity in the rice field is often estimated by counting numbers of whiteheads.

The primary late-season insect pest of rice in Louisiana is the rice stink bug, *Oebalus pugnax* F. (Hemiptera: Pentatomidae). Feeding by rice stink bugs on developing rice grains increases the incidence of unfilled, broken, and discolored grains known as "pecky' rice in milled rice (Patel et al. 2006, Espino 2007). Injury from rice stink bugs does not cause significant yield losses under most circumstances but greatly affects the quality of rice (Tindall et al. 2005, Espino 2007).

Control strategies for these rice insect pests rely heavily on chemical insecticides (Way 1990, Johnson et al. 2003, Reay-Jones et al. 2007). However, the use of insecticides has several negative aspects such as pest resurgence, hazards to users, environmental contamination, and costs associated with multiple applications (Chelliah and Bharathi 1994). Cultural methods that involve weed control, field draining, delayed flooding, and early planting have been utilized to manage rice water weevil, stem borers and rice stink bugs (Hesler et al. 1992, Kendig et al. 2003, Tindall et al. 2005, Stout et al. 2009). Host-plant resistance has also been targeted as an alternative and sustainable approach to manage rice insect pests. Some levels of resistance and tolerance traits have been observed in rice cultivars against the rice water weevil (Stout et al. 2001). In Texas, screening of rice cultivars against stem borers under field conditions has shown varying levels of injury and yield loss among cultivars (Way et al. 2006).

Ideally, management of rice insect pest complex should incorporate combinations of control tactics that complement each other. Silicon fertilization can be potentially integrated as a component of control strategies for rice insect pests. In this study, we evaluated the interactive effects of soil silicon amendment and rice cultivar on rice water weevil densities, whitehead incidences, rice stink bugs densities, and rice yields under field conditions in 2016 and 2017. Ten commonly grown rice cultivars in Louisiana were selected in this study. These cultivars do differ in their susceptibility to rice water weevil and stem borer infestations but their ability to take up silicon has not been investigated.

3.2. Materials and Methods

3.2.1. Field establishment

Field experiments were conducted in 2016 and 2017 at the H. Rouse Caffey Rice Station in Crowley, Louisiana (30°14'22.406"N, 92°20'46.195"W, 7 m asl) to investigate the interactive effects of rice cultivars and soil silicon amendments on rice water weevil densities, whitehead incidences, rice stinkbug densities, and yields. The selected rice cultivars are commonly planted in Louisiana. These cultivars provide a good representation of grain types and characteristics (long-grain, medium grain, special purpose, and Clearfield) (Table 3.1). All experiments employed a randomized complete block design with four replications. In each year, each block contained 20 plots subjected to factorial combinations of ten varieties and two levels of Ca₂SiO₄ slag as silicon source (0 and 5000 kg/ha).

Rice seeds were drill-seeded in 1.2 m x 5.5 m plots at a seeding rate of approximately 70 kg/ha for all cultivars in both years. Calcium silicate slag was evenly distributed on the soil surface of the plots assigned to the silicon treatment immediately after planting in 2016. In 2017, slag was applied a week later due to a heavy rain that followed drill-seeding. Fields were surface

irrigated as needed until permanent flooding to facilitate plant emergence. Table 3.2 lists the dates of field activities. Urea was used as source of nitrogen and was evenly distributed on each plot at full rate (no-split application) five to six weeks after planting (Table 3.2). Plots were fertilized at rates of 150 kg N/ha in 2016 and 135 kg N/ha in 2017. Insecticides were not used for the duration of the field experiments to facilitate natural infestations of rice water weevil, stem borers, and rice stink bug.

Entire plots were harvested by a mechanical harvester at grain maturity. Grain yields from each plot were adjusted to 12% moisture.

Cultivar	Rice Type	Year	
		2016	2017
CL151	Long grain Clearfield	Х	Х
CL161	Long grain Clearfield	Х	Х
CL261	Medium grain Clearfield	Х	Х
Caffey	Medium grain	Х	Х
Cheniere	Long grain	Х	Х
Cocodrie	Long grain	Х	Х
Cypress	Long grain	Х	Х
Jazzman	Specialty long grain	Х	Х
Jupiter	Medium grain	Х	Х
Mermentau	Long grain	Х	Х

Table 3.1. Rice cultivars used in field experiments on 2016 and 2017

 Table 3.2. Field activities and corresponding dates

Activity	Year		
	2016	2017	
Planting	6 April	29 May	
Silicon Slag Application	6 April	6 June	
Nitrogen Fertilization	17 May	5 July	
Permanent Flood	18 May	29 June	
1 st Core Sampling	10 June	20 July	
2 nd Core Sampling	16 June	28 July	
Whitehead counts	22 July	6 September	
Stinkbug counts	27 July	13 September	
Harvest	24 August	25 September	

3.2.2. Rice water weevil densities

Densities of immature rice water weevils (larvae and pupae) were estimated using a metal soil/root core sampler with a 10 cm diameter and a 10 cm depth. Core samplings were performed approximately three and four weeks after permanent flood (Table 3.2) because female weevils oviposit primarily after permanent flood is established (Stout et al. 2002). For each sampling date, two to three core samples with a minimum of one rice plant with intact roots in every core sample were taken from the interior portion of each plot. Core samples were processed individually by washing the soil from the roots in 40-mesh screen sieve buckets. Larvae were counted as they floated in the sieve buckets when dipped on basins with salt solution (N'guessan and Quisenberry 1982). The larval density in each plot was estimated by calculating the average number of larvae from two to three core samples from each plot.

3.2.3. Whitehead incidence

When feeding of stem borer larvae occur at reproductive stages, severance of the growing plant parts from the base dries the panicle, which may not emerge. Panicles that do emerge do not produce grains but remain straight and are whitish, a condition known as a whitehead (Pathak and Khan 1994). In this experiment, whiteheads were selected as an indicator of stem borer infestations in the field. Total numbers of whiteheads were counted at 100 percent heading in each plot. Whiteheads were collected and subsequently dissected to identify stem borer species. Mexican rice borer was the major cause of whiteheads in 2016 and 2017.

3.2.4. Rice stink bug densities

Rice stink bugs feed on the sap of developing rice grains; thus, stink bugs were sampled at the grain filling developmental stage of rice. Insect sweep nets (38-cm diameter) were used to determine total number of stink bugs in given area. Ten 180° sweeps of rice canopy were

performed in each plot. Densities of stink bug (number of stink bug per plot) were estimated by counting the total number of adults and nymphs from ten sweeps in each plot.

3.2.5. Plant analysis

Plant samples were collected at internode formation (four weeks after permanent flood) and grain maturity (two to three weeks before harvest) for plant silicon analysis. Silicon content in whole plant samples was determined by Oven-Induced Digestion (Kraska and Breitenbeck 2010) followed by the Molybdenum Blue Colorimetric procedure (Hallmark et al. 1982). For digestion, dried plant tissue was ground into powder and 100 mg of ground plant tissue was weighed into unused 50-mL polyethylene centrifuge tubes. The ground plant tissue was oven-dried for 15 minutes at 60° C to get rid of any moisture. Five drops of octyl alcohol (C₈H₁₈O) and two mL of hydrogen peroxide (H₂O₂) were added to the tubes and the tubes were placed back into the oven at 95° C for 30 minutes. Then, four mL of 50% sodium hydroxide (NaOH) were added and tubes were loosely capped and placed back into the oven for four hours. Tubes were taken out every 15 minutes and were mixed using a vortex mixer. After 4 hours, one mL of ammonium fluoride (NH₄F) was added to the digested samples. Tubes were gently mixed and diluted to 50 mL with deionized water. Reference samples with known silicon content from sugarcane and soybean as well as blanks were also included in the digestion.

For the colorimetric procedure, two mL aliquots of plant-digested solution were placed in 50-mL polyethylene centrifuge tubes. Then, 10 mL of 20% acetic acid solution (CH₃COOH) and two mL of 0.26 M ammonium molybdate [(NH₄)₆Mo₇O₂] were added. Tubes were allowed to stand for five minutes before two mL of 20% tartaric acid (C₄H₆O₆) were added. Solutions were mixed and allowed to stand for two minutes before adding two mL of reducing agent (ANSA: 0.5 mg of 1-amino-2-naphthol-4-sulphonic acid, 1.0 g sodium sulfite, and 30.0 g of sodium

bisulfite). The samples were diluted to a final volume of 30 mL with 20% acetic acid and absorbance readings were measured at 630 nm using UV-Visible Spectrophotometer (Hach DR 500). Standard series at rates of 0, 0.4, 0.8, 1.6, 3.2, 4.8, and 6.4 ug mL⁻¹ of Si were also included in the colorimetric procedure.

3.2.6. Data analysis

Rice water weevil larval densities, whitehead incidences, rice stink bug densities, plant silicon contents, and yields in 2016 and 2017 were analyzed separately as factorial randomized block design experiments with block as random effect and rice cultivar, soil silicon amendment, and their interaction as fixed effects using mixed model analysis of variance in PROC MIXED of SAS (SAS 2013). Rice water weevil densities from two sampling dates (Table 3.2) in each year were analyzed separately. Estimated means were separated using Tukey's HSD test (Tukey 1953). Residuals were analyzed for normality using PROC UNIVARIATE (SAS 2013) and data transformations were performed as necessary to satisfy normality assumption, but untransformed data are presented. Transformations were performed on the following data sets using either square or cube root: whitehead incidence in 2016; and weevil densities (first core sampling), whitehead incidences, and rice stink bug densities in 2017. The Kenward-Rogers adjustment for degrees of freedom in mixed models was applied in all analyses (Littell et al. 2002).

3.3. Results

3.3.1. Rice water weevil densities

Data collected from each year were analyzed separately by core sampling date. In 2016, rice water weevil larval densities differed among varieties in the second core sampling (F = 2.84; df = 9, 59; P = 0.0076) but not in the first core sampling (F = 1.39; df = 9, 59; P = 0.2155). In 2017, the effect of rice cultivar was significant in both the first (F = 3.93; df = 9, 59; P = 0.0006)

and second (F = 2.13; df = 9, 59; P = 0.0413) core samplings. For each year, levels of weevil infestations varied among rice cultivars (Figure 3.1); however, there appears to be no clear trend in what specific cultivar can support higher levels of weevil infestations.

The effect of soil silicon amendment on rice water weevil densities was significant in the second core sampling (F = 6.05; df = 1, 59; P = 0.0168) and marginally significant in the first core sampling (F = 3.75; df = 1, 59; P = 0.0576) in 2016. In contrast, soil silicon amendment did not affect weevil larval densities in the first (F = 0.38; df = 1, 59; P = 0.5417) and second (F = 1.36; df = 1, 59; P = 0.2482) core samplings in 2017. Weevil larval densities were observed to be lower in silicon treated plots compared to untreated plots in the second core sampling in 2016 (Figure 3.2). Although lower larval densities were observed in plots treated with silicon compared to untreated plots in the first core sampling in 2016, post-hoc analysis (Tukey 1953) revealed no significant differences among silicon treatments (Figure 3.2). Moreover, there were no significant interactions between rice cultivar and soil silicon amendment (data not shown). 3.3.2. Whitehead incidence

Densities of whiteheads (whiteheads per plot) were assessed to estimate stem borer infestations in the field. Mexican rice borer was found to be the predominant cause of whiteheads in 2016 and 2017. In this study, the effects of rice cultivar on whitehead densities was significant in 2016 (F = 6.11; df = 9, 59; P < 0.0001) and 2017 (F = 8.18; df = 9, 60; P < 0.0001). Consistently, lower whitehead incidence was observed in plots of the specialty long-grain rice cultivar 'Jazzman' and higher incidence was observed in the medium-grain Clearfield rice cultivar 'CL261' (Table 3.3). In contrast, soil silicon amendment did not affect whitehead incidence in 2016 (F = 0.03; df = 1, 59; P = 0.8608) or 2017 (F = 0.07; df = 1, 60; P = 0.7938) (Figure 3.2). There were no significant interactions between rice cultivar and soil silicon amendment in 2016 (F = 0.53; df = 9, 59; P = 0.8470) or 2017 (F = 0.48; df = 9, 60; P = 0.8831).

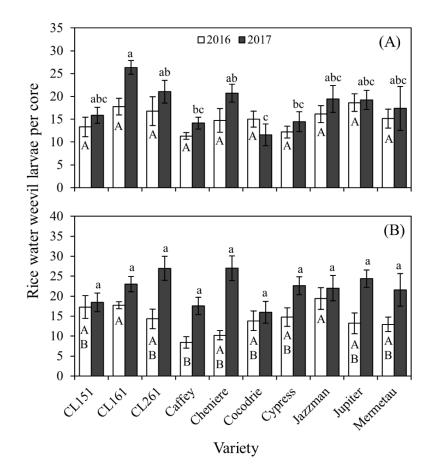


Figure 3.1. Main effect of variety on rice water weevil larval densities in 2016 and 2017, Crowley, LA. Core 1 (A) and Core 2 (B) samplings were performed three and four weeks, respectively, after permanent flooding. For each year, bars accompanied by the same letter represent means that are not significantly different (P>0.05; Tukey's HSD). Error bars represent standard error of means (SEM).

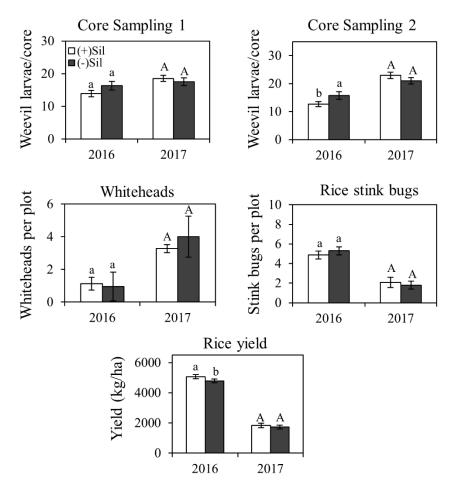


Figure 3.2. Main effects of soil silicon amendment on rice water weevil larval densities (Core 1 and Core 2), whitehead incidences, rice stink bug densities, and rice yields in 2016 and 2017, Crowley, LA. For each year, bars accompanied by the same letter represent means that are not significantly different (P>0.05; Tukey's HSD). Error bars represent standard error of means (SEM).

Variety	2016		2017	
	WHs ^a /plot	RSB ^b /plot	WHs/plot	RSB /plot
CL151	3.00±1.20 ab	4.50±0.82 abc	3.75±2.26 b	2.75±1.06 ab
CL161	0.38±0.38 c	4.87±1.06 abc	1.37±0.50 bc	1.00±0.50 ab
CL261	3.88±1.02 a	7.50±1.15 ab	20.00±3.69 a	2.12±0.55 ab
Caffey	0.38±0.26 bc	7.88±0.79 a	3.12±1.61 bc	4.50±1.50 a
Cheniere	0.12±0.12 c	4.25±0.77 abc	1.75±0.37 bc	2.00±0.82 ab
Cocodrie	0.38±0.26 bc	3.88±1.04 abc	1.25±0.41 bc	1.62±0.10 ab
Cypress	0.00±0.00 c	4.62±0.62 abc	0.25±0.16 bc	1.75±0.96 ab
Jazzman	0.00±0.00 c	3.50±0.87 bc	0.00±0.00 c	1.87±0.51 ab
Jupiter	0.62±0.32 bc	6.50±1.36 abc	1.62±0.10 bc	1.5±0.82 ab
Mermentau	1.63±0.68 abc	3.37±0.78 c	3.25±1.26 bc	0.25±0.16 b

Table 3.3. Main effect of variety on whiteheads (\pm SEM) and rice stink bug densities (\pm SEM) in 2016 and 2017, Crowley, LA. Means in a column followed by the same letter are not significantly different (P>0.05; Tukey's HSD).

^a Whiteheads

^b Rice stink bug

3.3.3. Rice stink bug densities

Densities of rice stinkbugs (total number of stink bug adults and nymphs per plot) were assessed in 2016 and 2017 by using sweepnets. For data analysis, numbers of adults and nymphs were combined since both stages feed on developing rice grains. Effects of rice cultivar on stink bug densities were significant in 2016 (F = 3.38; df = 9, 59; P = 0.0021) and 2017 (F = 2.12; df = 9, 60; P = 0.0416). Consistently, higher stink bug densities were observed in the plots of medium-grain rice cultivar 'Caffey' and lower densities were observed in the long-grain rice cultivar 'Mermentau' (Table 3.3). In contrast, soil silicon amendment did not affect stink bug densities in 2016 (F = 0.58; df = 1, 59; P = 0.4504) or 2017 (F = 0.44; df = 1, 60; P = 0.5083) (Figure 3.2). There were no interactions observed between rice cultivar and soil silicon amendment in 2016 (F = 1.96; df = 9, 59; P = 0.0603) or 2017 (F = 0.70; df = 9, 60; P = 0.7068). 3.3.4. Silicon analysis

Shoots and roots were collected for silicon analysis in both years but only rice samples collected at internode formation in 2016 have been analyzed to date. For these samples, silicon

accumulation in shoots was found to differ significantly among rice cultivars (F = 7.76; df = 9, 60; P < 0.0001). Highest silicon contents were observed in the medium-grain cultivars 'CL261' and 'Jupiter' and lowest contents were observed in the long-grain cultivars 'CL161' and 'Cypress' (Figure 3.3). Unexpectedly, silicon treatment did not affect silicon accumulation in either rice shoots (F = 0.33; df = 1, 60; P = 0.5698) or roots (F = 1.39; F = 1, 59; P = 0.2438). Silicon accumulation in rice roots did not vary among rice cultivars (F = 1.06; df = 9, 59; P = 0.4018). Moreover, higher silicon accumulation was observed in shoots compared to roots (F = 232.59; df = 1, 119; P < 0.0001). There were no interactions observed between rice cultivar and soil silicon amendment in both shoots and roots (data not shown).

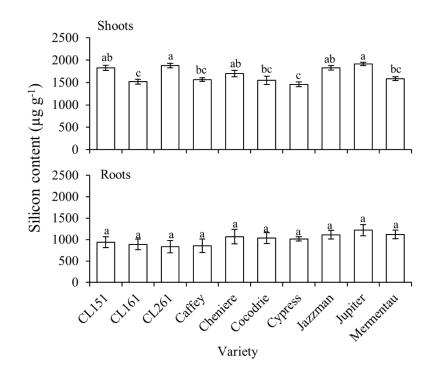


Figure 3.3. Main effect of variety on silicon accumulation ($\mu g g^{-1}$) in plant shoots and roots in 2016. Bars accompanied by the same letter represent means that are not significantly different (P>0.05; Tukey's HSD). Error bars represent standard error of means (SEM).

3.3.5. Yield

Effects of rice cultivars on yields were significant in 2016 (F = 10.52; df = 9, 60; P < 0.0001) and 2017 (F = 2.12; df = 9, 60; P = 0.0416). The variation in yields among cultivars between years were inconsistent (Table 3.4). For instance, in 2016, 'Cypress' produced the lowest yield, while in 2017, it produced a moderately high yield. The effects of soil silicon amendment on yields were significant in 2016 (F = 4.01; df = 1, 60; P = 0.0497) but not in 2017 (F = 0.95; df = 1, 59; P = 0.3338). In 2016, higher yields were observed on plots ammended with silicon slag compared to untreated plots (Figure 3.2). There were no significant interactions between rice cultivars and soil silicon amendment in 2016 (F = 1.28; df = 9, 60; P = 0.2646) and 2017 (F = 0.41; df = 9, 59; P = 0.9239).

Table 3.4. Main effect of variety on rice yields (kg/ha±SEM) in 2016 and 2017, Crowley, LA. Means in a column followed by the same letter are not significantly different (P>0.05; Tukey's HSD).

Cultivar	Year	
	2016	2017
CL151	6116.30±257.57 a	2054.69±187.15 abcd
CL161	4305.05±205.13 ef	2876.91±62.00 a
CL261	5406.50±230.50 abc	2606.18±216.65 ab
Caffey	4424.96±183.74 cdef	1421.12±194.81 cdef
Cheniere	5445.65±192.96 ab	2266.48±61.89 abc
Cocodrie	5004.14±182.79 bcde	1081.73±202.40 f
Cypress	3732.51±193.94 f	1927.33±217.80 bcde
Jazzman	4376.54±185.40 def	1405.96±174.33 def
Jupiter	5342.89±306.15 abcd	1161.00±199.43 ef
Mermentau	5142.49±286.40 abcde	985.32±226.81 f

3.4. Discussion

There is a need to develop pest management strategies that incorporate a variety of tactics to ensure a more sustainable crop production. The major rice insect pests in Louisiana are the rice water weevil (major early-season pest), rice stink bug (major late-season pest), sugarcane borer and rice stalk borer (long-established but sporadic stem boring pests), and Mexican rice borer (invasive stem boring pest). This study was conducted to investigate the effects of rice cultivars and soil silicon amendment on rice insect pest complex in field conditions. This study was also among the first to investigate the effects of soil silicon amendment on root-feeding insect pest of rice. Moreover, this study is one of the few that examine effects of silicon on multiple insect pest of rice under natural infestations.

In this study, silicon accumulation in shoots was found to differ significantly among rice cultivars (Figure 3.3). Higher concentration of silicon was also observed in shoots compared to roots. However, silicon concentration in both the shoots and roots was not increased by soil silicon amendment. The amounts of plant-available silicon in the soil might have been high enough to begin with, thus, silicon amendment did not affect the concentration in plants. Silicon content in the remaining plant samples need to be analyzed in order to shed more light on this.

It has been proposed that amendment of soils with silicon-based fertilizers can augment plant resistance via two mechanisms (Reynolds et al. 2009). One mechanism involves the uptake of silicon by plants that lead to deposition of amorphous silica in plant tissues that contributes to the thickening of the epidermal cell layer, forming a mechanical barrier. This barrier results in increased rigidity and abrasiveness of plant tissues, thus reducing the digestibility and palatability of plants to herbivorous pests (Ma 2004, Massey et al. 2006, Massey and Hartley 2009). A second mechanism involves the ability of silicon to stimulate biochemical pathways related to expression of specific resistance related traits against biotic stresses. For instance, silicon treated rice plants show increased activity of the enzymes pheny-lalanine-ammonia lyase, polyphenoloxidase, and β - 1, 3 glucanase, in response to hopper infestations (Yang et al. 2017).

In this study, densities of the root-feeding rice water weevil larvae was negatively affected by soil silicon amendment in one core sampling in 2016 (Figure 3.2). It has been

reported recently that soil silicon amendment reduced the density of rice water weevil larvae in a single core sampling under field condition but the effects of silicon on weevil larvae were weaker that the effects of insecticide treatment and variety (Villegas et al. 2017). The weak effect of soil silicon amendment on weevil larval densities might be attributed to several factors. Silicon accumulation in rice plants might not have reached sufficient levels to influence an effect on weevil densities. Typically, older plants have higher silicon content compared to younger plants due to continual uptake and immobility of silicon once deposited in plant tissues (Ishizuka 1964, Epstein 1994). Moreover, monosilicic acid released from calcium silicate slag is highly influenced by soil properties such as pH, organic matter, and clay content (Tubana et al. 2016). To our knowledge, the only other study to report reductions on performance of a root-feeding herbivore in response to silicon was conducted in a glass house experiment on sugarcane using soil-dwelling 'canegrubs' (Frew et al. 2016).

Soil silicon amendment also did not affect whitehead incidence in 2016 and 2017 (Figure 3.2) which was unexpected because there is a wealth of studies that support the role of silicon in augmenting resistance to stem-boring lepidopteran pests. In fact, the first study to report the role of silicon in plant resistance was associated with resistance against the rice stem borer, *Chilo simplex* (Sasamoto 1953). Since that time, several studies have documented the role of silicon in enhancing plant resistance to several stem boring pests such as yellow rice borer (Panda et al. 1975), African striped borer (Ukwungwu and Odebiyi 1985), Asiatic rice borer (Hou and Han 2010), and sugarcane borer (Sidhu et al. 2013). The lack of effect of soil silicon amendment on whitehead incidence might be caused by relatively low population and uneven distribution of stem borers in the field. Other types of stem borer injury such as deadhearts, partial whiteheads, and unemerged whiteheads may have been present in the field but was not evaluated. Mexican

rice borer was also found to be the predominant cause of whiteheads in the field in 2016 and 2017, which may potentially be less sensitive to silicon-induced changes.

Rice stinkbug population was also examined in the field. Results showed no significant differences on stink bug population between silicon treated and untreated plots (Figure 3.2). Although soil silicon amendment did not affect the densities of stink bugs, this study is the first attempt to investigate the effect of silicon on stink bug population.

Despite the lack or weak effects of soil silicon amendment on rice insect pest densities, rice yields was significantly affected by silicon treatment (Figure 3.2). In 2016, higher yields were observed on silicon treated plots compared to untreated plots. Effects of silicon fertilization on crop yields have been previously documented. Increased rice yields due to silicon fertilization was observed in field experiments conducted in Japan (Ma and Takahashi 2002) and Texas (Villegas et al. 2017). Application of silicon-containing materials also increased grain yield in wheat (Liang et al. 1994).

The development and intentional use of crop varieties possessing genetically-based traits that reduce the amount of damage from herbivores is the basic tenet of host-plant resistance (Smith 2005, Stout 2014). The use of varieties with enhance resistance can be easily integrated with other tactics and is cost-effective. In this study, ten rice cultivars that are commonly grown Louisiana were selected. Results showed some differences on the densities of rice water weevil larvae among the rice cultivars (Figure 3.1). However, the differences on weevil infestations among the rice cultivars were not very high. Previous study has reported that some of the cultivars commonly grown in Louisiana do differ in their susceptibility to infestation by rice water weevil larvae and their tolerance of rice water weevil feeding; however, none possess high levels of resistance (Stout et al. 2001). Recently, a study conducted in Texas also evaluated

varietal resistance to rice water weevil and found no significant difference on larval densities between selected commercial varieties that were inbred tropical japonica subspecies and commercially grown in the southern United States and germplasm lines that were indica subspecies adapted to tropical climates of Asia (Vyavhare et al. 2016).

Whitehead incidence was also found to differ among rice cultivars evaluated in this study (Table 3.3). Consistently, lowest whiteheads were observed in 'Jazzman' and the highest were observed in 'CL261'. There have been some studies conducted on stem borer resistance in Louisiana rice cultivars. It was reported that the oviposition preference and larval performance of sugarcane borers differed among commonly grown rice cultivars in Louisiana (Hamm et al. 2011, Sidhu et al. 2013). Douglas and Ingram (1942) also observed that sugarcane borer and rice stalk borer were more abundant in rice plants with larger culms. A recent study in Texas indicate that some conventional rice cultivars are more susceptible to sugarcane borer and Mexican rice borer injury, while hybrid cultivars were less injured and yielded more than conventional cultivars (Way et al. 2006). In Asia, rice cultivars are routinely screened for resistance to stem bores and resistant cultivars are widely used as control tactic (Chaudhary et al. 1984).

Furthermore, densities of rice stinkbug significantly differed among rice cultivars (Table 3.3). Consistently, higher stink bug densities were observed in 'Caffey' and lower densities were observed in 'Mermentau' (Table 3.3). Although feeding of stink bug on developing rice grains does not cause yield loss, it affects the quality of rice (Tindall et al. 2005, Espino 2007). It has been reported that some level of resistance to stink bug exist among rice cultivars but correlation between resistance and yield appear to be negative (Bernhardt et al. 2004). Studies indicated that rice resistance to stinkbug increases with grain length and rice cultivars least affected by stink bug feeding were not the highest yielding (Bernhardt et al. 2004).

It is important to understand the role of soil silicon amendment as a potential component of control strategies against rice pests in the southern United States. This study is among the first to investigate the role of silicon in augmenting the resistance of rice against multiple insect pests in field conditions, and also one of the first to document effects of silicon on a root-feeding pest in rice. Rice cultivars can play a big role to influence the potential effect of soil silicon amendment on insect pest densities. In order to maximize the benefits of silicon fertilization, a rice cultivar should be able to accumulate silicon at levels where the impact on insect pests will be apparent. Despite the weak effect of silicon on insect pests in this study, silicon could still play an important role in rice production considering the positive effects on yield and documented effects on disease suppression.

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Chapter 4. Summary and Conclusions

Rice is one of the major crops in Louisiana. Rice plants are subjected to various pressures by pests and diseases throughout the growing season. The major rice insect pests in Louisiana are the rice water weevil, rice stink bug, sugarcane borer, rice stalk borer, Mexican rice borer, and fall armyworm.

Currently, management of these insect pests relies heavily on chemical insecticides (Way 1990, Johnson et al. 2003, Reay-Jones et al. 2007, Blackman et al. 2014). However, the use of insecticides has several negative aspects such as pest resurgence, hazards to users, environmental contamination, and costs associated with multiple applications (Chelliah and Bharathi 1994). Cultural methods that involve weed control, field draining, delayed flooding, and early planting have been utilized to manage rice water weevil, stem borers and rice stink bugs (Hesler et al. 1992, Kendig et al. 2003, Tindall et al. 2005, Stout et al. 2009). Some levels of resistance and tolerance also exist among rice cultivars against rice water weevil, stem borers, and stink bugs (Stout et al. 2001, Bernhardt et al. 2004, Way et al. 2006, Mohammad Saad 2017). Although resistant varieties have been an integral part of pest management in rice in other parts of the world, it can potentially lose its effectiveness in insect pest management if target pests acquired virulence (Fujita et al. 2009).

This study investigated the effects of soil amendment, in combination with nitrogen fertilization and rice cultivars, on the rice insect pest complex in Louisiana. It has been proposed that amendment of soils with silicon-based fertilizers can augment plant resistance against herbivorous pests (Reynolds et al. 2009). In fact, the first study to report the role of silicon in plant resistance involved silicon-induced resistance against the rice stem borer, *Chilo simplex*, in Japan (Sasamoto 1953).

Field experiments were conducted from 2015 to 2017 to investigate the effects of soil silicon amendment and nitrogen fertilization on the rice insect pest complex. In this study, we found that high nitrogen fertilization levels significantly increased the infestations of the rice water weevil larvae in 2015, 2016, and 2017. Effects of nitrogen rates on whitehead densities was marginally significant in 2017 but not in 2015 and 2016. Higher rice yields were also observed on plots with higher levels of nitrogen. In contrast, soil silicon amendment did not affect weevil densities, whitehead incidences, stink bug population, or yield.

Furthermore, studies conducted in the greenhouse in 2015 and 2016 revealed that the force required to penetrate rice stem was higher on silicon-treated rice plants compared to untreated plants. However, soil silicon amendment did not affect growth rates of fall armyworm and sugarcane borer. On the hand, higher rates of nitrogen applied to rice plants increased the relative growth rates of sugarcane borer and fall armyworm. Entry holes caused by sugarcane borer were significantly higher on rice plants when nitrogen was applied.

Separate field experiments were also conducted in 2016 and 2017 to investigate the effects of silicon fertilization and rice cultivars on the rice insect pests. Ten commonly grown rice cultivars that differ in their susceptibility to rice water weevil and stem borer infestations were selected in this study. Previous studies have reported significant variations on silicon concentration among rice cultivars (Deren 2001, Ma et al. 2007). The ability of the selected rice cultivars to take up silicon has not been previously investigated. In this study, reductions in weevil larval densities were observed in silicon treated plots compared to untreated plots in one core sampling in 2016. Similarly, higher yields were observed in silicon-treated plots compared to untreated plots compared to untreated plots in 2016. Silicon accumulation in shoots was also found to differ significantly among rice cultivars and higher concentration of silicon was observed in shoots compared to

roots. However, silicon concentration in both the shoots and roots was not increased by soil silicon amendment. Silicon amendment also did not affect whitehead incidences or rice stink bug densities in both years. It was observed that the levels of infestations of rice water weevil, stem borers, and rice stink bugs varied among the rice cultivars evaluated in the study.

Overall, results from this study suggest that the effects of soil silicon amendment can vary depending on the environmental condition. Effects of silicon may depend on the rice cultivars and soil properties. For future studies, rice cultivars that can accumulate silicon at a higher rate should be selected and planted at soils where available silicon is limited. This will allow to elucidate the potential for silicon to enhance plant resistance against multiple insect pests.

Despite the weak effect of silicon on insect pests in this study, it still important to understand the effects of silicon fertilization and its implications for plant resistance. Silicon could still play a significant role in rice production considering the positive effects on yield and documented effects on disease suppression. Moreover, silicon amendment may be a valuable tool in areas where natural silicon levels in soil are limiting.

In future studies, it will be beneficial to investigate the effects of silicon on the expression of resistance related traits in response to infestations by insect pests. In particular, enzymes related to plant defense such as pheny-lalanine-ammonia lyase, polyphenoloxidase, β - 1, 3 glucanase, and chitinase can be quantified in silicon treated rice plants in the presence of herbivorous pests.

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Vita

James was born in Bacolod City, Philippines in 1989. He received his B.S. in Life Sciences with specialization in molecular biology and biotechnology from the Ateneo de Manila University in 2012. His undergraduate research was focused on the molecular characterization and phylogenetic analyses of RNA segments 5 and 6 of rice ragged stunt virus transmitted by brown planthopper in Southeast Asia. His career and passion in entomology began when he joined the Entomology group at the International Rice Research Institute. He was involved in developing ecological engineering approaches to restore and conserve ecosystem services for pest management in rice production. He also worked on rodent-rice interaction in response to crop management. In 2015, he began his M.S. in Entomology with a minor in applied statistics at the Louisiana State University. His research focuses on the independent and interactive effects of nitrogen fertilization and soil silicon amendment on the rice insect pest complex. He recently published the first study to report the effects of soil silicon amendment on rice water weevil (a below-ground herbivore) in rice under field conditions. After he finish his Masters, he plan to continue studying to get a PhD at LSU.