POPULATION DENSITY OF RICE ROOT NEMATODE, *HIRSCHMANNIELLA ORYZAE* (Luc and Goodey, 1964) IN NAY PYI TAW UNION TERRITORY AND RESPONSE OF SOME RICE VARIETIES

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A Thesis submitted to the post-graduate committee of the Yezin Agricultural University in the partial fulfillment of the requirements for the degree of Master of Agricultural Science (Plant Pathology)

Department of Plant Pathology
Yezin Agricultural University
Yezin, Nay Pyi Taw

NOVEMBER 2018

The thesis attached hereto, entitled **"Population Density of Rice Root Nematode, *Hirschmanniella oryzae* (Luc and Goodey, 1964) in Nay Pyi Taw Union Territory and Response of Some Rice Varieties"** was prepared under the direction of the chairperson of the candidate supervisory committee and has been approved by all members of that committee and board of examiners as partial fulfillment of the requirements for the degree of **Master of Agricultural Science (Plant Pathology)**.

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DECLARATION OF ORIGINALITY

This thesis represents the original work of the author, except where otherwise stated. It has not been submitted previously for a degree at this or any other university.

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DEDICATED TO MY BELOVED PARENTS

U MYA AUNG AND DAW KYU KYU

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ABSTRACT

Soil and root samples from 44 rice fields of 5 summer rice varieties (Manawthukha, Sinthukha, Shwethweyin, Palethwe and Yet-90) were collected in 5 townships (Lewe, Tatkon, Pyinmana, Zabuthiri and Dekkhinathiri) to determine the population density of *Hirschmanniella oryzae*. It was observed that 98.89% out of 44 fields sampled were infested with the rice root nematode, *H. oryzae*. Based on the prominence value (a combination of the frequency of occurrence and abundance) of *H. oryzae*, the highest population was found in Tatkon Township and the lowest in Lewe Township. The highest population of *H. oryzae* from soil and root were observed in Sinthukha and the lowest population was found in Shwethweyin. All summer rice varieties surveyed were observed to be either susceptible or highly susceptible to *H. oryzae*. In two different cropping sequences, rice-blackgram-rice cropping sequence had the lower nematode population than that of rice-rice cropping one. Moreover, the lower nematode population was also found in direct seeding than in transplanting method. The tested 12 rice varieties were inoculated with 2000 nematodes per plant. Plant height, tiller number, fresh shoot weight, fresh root weight, dry shoot weight, filled grain percent, 1000 grain weight and final nematode population were recorded at harvest. Among tested 12 rice varieties, Yet-90 and Sinthukha were rated as highly susceptible and Pawsanmwe and Manawthukha as susceptible to *H. oryzae*. The other eight varieties; Thukhamwe, Pyitawyin, Salt tolerant Sinthwelatt, Shweasean, Myaungmyamay, Pyimyanmarsein, Submergence-tolerant 1 and Shwethweyin were found as moderately resistant varieties. Conclusively, the most infested region was Tatkon Township in Nay Pyi Taw Union Territory and the only economical way to control rice root nematode is the use of resistant rice varieties and avoidance of monocropping and transplanting practices.

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CHAPTER I
INTRODUCTION

Rice (*Oryza sativa* L.) is the most important staple food for a large part of the world's human population and also provides more than one fifth of the calories consumed worldwide by the human. It is grown in at least 114 countries with global production of 645 million tons; about 90% of the total production was occupied by Asian farmers (Sharif 2014). Rice cultivation dominates the agricultural sector of Myanmar and it is mainly grown by small scale farmers. It is important for the food security of the majority of the population as well as for a source of cash income for smallholders who sell their harvest on the local markets and in the urban areas (Maung *et al.* 2010).

Rice is currently challenged by different biotic and abiotic factors. The biotic factors such as fungi, bacteria, viruses and nematodes diseases have been reported on rice crop in the world. Diseases are considered as the major constraints in rice production and responsible for losses in quantity and quality of harvested produce (Wasihun and Flagote 2016). Of these, plant-parasitic nematodes are major biotic constraints to higher productivity (Bridge *et al*. 2005). Nematodes feed on roots and reduce their potential for uptake of water and nutrients, but this hidden pest and its associated damage are generally hidden from the growers (Mojtahedi and Lownsbery 1975).

In Myanmar, ufra, root rot, root knot, and white tip are the most destructive nematode diseases in rice production (Mya *et al*. 1983; Aung *et al.* 1993). Presently root rot caused by *Hirschmanniella oryzae* is the second most important disease among rice nematode diseases in Myanmar. It had been reported that this disease became widely distributed in Western Bago Region including, Pyi, Paukhaung, Thegone, Natalin, Zigone and Paungte Townships. It was also observed that the disease was most commonly found in Kyaukse, Pyinmana and Patheingyi Townships (Myint *et al*. 2004).

Root nematodes belonging to the genus *Hirschmanniella* infest 58% of the world's rice fields and cause 25% yield reduction. *H. oryzae* is the most frequently found and considered as the most important rice root nematode species in most of the rice growing areas in Asia such as India, Pakistan, Bangladesh, Sri Lanka, Nepal, Thailand, Vietnam, Indonesia, Philippines, China, Korea and Japan. Worldwide, *H. oryzae* is the most common plant-parasitic nematode on irrigated rice areas with a long history of cultivation and when the plants are constantly flooded (Bridge *et al*. 2005). This nematode is completely adapted for irrigated lowland rice in which the plants are constantly flooded (Fortuner and Merny 1979).

Roots invaded by *H. oryzae* show discoloration, deterioration and rotting, while heavily infested plants also exhibit above-ground symptoms like stunting and reduction in tillering might be as much as 60% (Kyndt *et al*. 2014). In South and Southeast Asia, this nematode can cause varying degree of yield losses under different environmental conditions. Yield loss due to *H. oryzae* ranged from 27 to 39% when rice seedlings were inoculated with 1200 nematodes per plant (Yamsonrat 1967), 100 nematodes per plant (Mathur and Prasad 1972) and 1-10 nematodes per g soil (Jonathan and Velayuthan 1987). Babatola and Bridge (1979) observed that 69% reduction in grain yield when the rice plants were inoculated with 1000 *H. oryzae*. *H. oryzae* also affected the number of panicles and grain weight (Mathur and Prasad 1972; Jonathan and Velayuthan 1987).

An important aspect of plant-parasitic nematode management is the testing of a particular site for the presence of nematodes prior to the planting of a crop (Davies 1997). It is needed to determine initially which problems exist by doing survey and accurately identify which nematodes are harmful or economically important by pathogenicity tests and field trials, and deciding on which treatments or methods are appropriate for the control of nematode. Therefore, knowing which nematode genera and species occurred is necessary as the first step (Luc *et al*. 1990). If there is no knowledge regarding on the occurrence and abundance of potential pathogens, yield reduction can be high. An accurate knowledge and the population density of which nematode is the first step in any nematode management strategy (Maung 2011).

Rice root nematode, *H. oryzae*, can be controlled by various practices such as particular fallow, weed control, use of resistant cultivars, rotation with non-host plants, chemical soil treatments of nurseries and fields, and chemical root dipping and seed coating (Bridge *et al*. 1990). The use of resistant varieties is nowadays considered as a promising and often effective management practice to improve crop yield in the presence of plant-parasitic nematodes population density that exceed the damage threshold level. It is also an environment-friendly approach to nematode management in contrast with the currently use of expensive and hazardous chemicals (Starr and Roberts 2004).

It was found that 90% of the rice fields were infested by *H. oryzae* in the major rice production regions (Bago, Ayeyarwaddy, Yangon and Mandalay Division) (Maung *et al.* 2010). Nay Pyi Taw Union Territory has been recently considered as one of the centres where Palethwe is introduced and intensively cultivated (MOAI 2014). Hybrid rice variety, Palethwe, is a good option to increase rice productivity, but it is susceptible to a wide range of pests and diseases (Shanti *et al.* 2010). There are also indications that *H. oryzae* is more pathogenic on the recently introduced high-yielding varieties compared with the traditionally grown cultivars (Thein 2003).

At present, detailed information on the occurrence and distribution along with the damage and yield loss caused by *H. oryzae* is not available. In Myanmar, evaluation on varietal resistance of rice against *H. oryzae* was still limited. Therefore, the present study was conducted with the following objectives:

(1) to determine the population density of *H. oryzae* in rice production areas of Nay Pyi Taw Union Territory, and

(2) to evaluate the response of some rice varieties to rice root nematode

CHAPTER II
LITERATURE REVIEW

2.1 Production of Rice in Myanmar

In Myanmar, rice is the major agricultural crop and is grown throughout the country under different agroecosystems. It is grown both under rainfed lowland (monsoon) and upland conditions during the rainy season as well as under irrigated conditions during the dry summer (Maung *et al*. 2010). According to the national planning targets, the total area of paddy was 7.60 million hectares comprising 6.42 million hectares under monsoon paddy and 1.19 million hectares under summer paddy and average yield was 4.19 metric ton per hectare in 2015-2016. Actual paddy sown area in the year 2015-2016 was 7.21 million hectares and the production was reached at 28.21 million metric ton (MOALI 2016).

Nay Pyi Taw Union Territory is one of the rice producing areas in Myanmar where ecological environment is favorable for monsoon and summer rice production. In this area, total rice sown area in 2015-2016 was 71,689 hectares of which 67,632 hectares were sown during monsoon and 4057 hectares in summer (DoA 2016). As part of efforts for boosting per hectare yield for the food security in the country and doubling income of farmers, the Myanmar government has been setting up its efforts throughout Myanmar including Nay Pyi Taw Union Territory (MOAI 2014).

Although the total sown area of Nay Pyi Taw is 1% compared with national sown area, farmers in Nay Pyi Taw grow rice intensively as monsoon and summer crops. Currently popular rice varieties in Nay Pyi Taw are Palethwe, Manawthukha, Sinthukha, Shwethweyin and Sinthwelatt in monsoon, and Manawthukha, Shwethweyin, Palethwe and Sinthukha in summer (Department of Agricultural Economics 2013).

2.1.1 Rice cropping sequences in Myanmar

Rice is one of the most important cereal crops in developing countries. It serves as staple diet and provides food for nearly half of the global population (FAO 2004). The cultivation of high yielding varieties nowadays has generated more income from rice for small-scale farmers (DAP 2009). However, the farmers’ income has not yet improved appreciably (Aye and Yin 1996). Due to the nature of the crops, and soil and weather conditions of Myanmar, pulses and oil seed crops are the major groups of crops which are being grown after rice in rice-based cropping systems. Moreover, because of the high market value and the minimal irrigation water or external input requirements in addition to the awareness of farmers of the role of legumes in enhancing soil fertility, the cultivation of pulses and oil seed crops is much preferred by farmers (Matsuno and Fujita 1986). As a result, rice-blackgram or rice-mungbean cropping sequences are being popular in both Upper and Lower Myanmar. In most regions of Central Myanmar, sesame cultivation followed by rice is commonly practiced (Naing *et al*. 2002; Okamato 2004).

2.2 Rice Root Nematode, *Hirschmanniella oryzae*

Plant-parasitic nematodes consititute one of the important groups of organisms which inhabit the soil around the roots of plants and frequently play a vital part in their growth and production (Thorne 1961). They caused severe constraint on agricultural production (Oteifa 1997). In general, not all the plant-parasitic nematode species associated with rice are considered of much importance for rice production (Prot and Rahman 1994). More than 144 different species of stylet bearing nematodes occur in association with rice culture (Gedder and Smart 1987; Singh *et al*. 2005). However, some of these plant-parasitic nematode species such as root-knot nematode (*Meloidogyne* spp.), foliar nematode (*Aphelenchoides besseyi*), stem nematode (*Ditylenchus angustus*) and root nematodes (*Hirschmanniellla* spp.) can be harmful to the crop resulting in impressive symptoms on leaves, tillers and panicles which finally in poor plant growth and yield loss (Maclean *et al*. 2002; Bridge *et al*. 2005).

Among the root parasites, the rice root nematodes (*Hirschmanniella* spp.) are a group of important migratory endoparasites with a great risk potential because of their presence in different rice agro-ecosystems in most of the rice growing regions. Most irrigated and rainfed lowland rice fields are infested with at least one species of *Hirschmanniella* which can develop high population density in the roots (Fortuner and Merny 1979; Babatola and Bridge 1980). In South and Southeast Asia, *Hirschmanniella* spp. causes a varying degree of yield losses under different field conditions (Prot *et al*. 1994).

A number of *Hirschmanniella* species, known collectively as rice root nematodes, are parasites of rice. The most commonly recorded species is *Hirschmanniella oryzae* but there was a tendency in the earlier literature for all *Hirschmanniella* spp. found in rice roots to be grouped under the name *H. oryzae*

(Taylor 1969). Seven species namely *Hirschmanniella belli*, *H. gracilis*, *H. imamuri*, *H. mexicana* (*H. caudacrena*), *H. mucronata*, *H. oryzae* and *H. spinicaudata* are reported to damage rice, whilst a further six species such as *Hirschmanniella kaverii*, *H. magna*, *H. nghetinhiensis*, *H. ornata*, *H. shamimi*, and *H. thornei* have been found in rice roots. Four species, *Hirschmanniella asteromucronata*, *H. furcata*, *H. obesa* and *H. truncata* have been recorded from weeds in rice fields. Although *Hirschmanniella* spp. attacking rice usually distributes as one or more species in all rice-growing countries and infesting some 200 million acres (Hollis and Keoboonrueng 1984), the most commonly recorded species is *H. oryzae* (Bridge *et al*. 1990).

2.2.1 Economic importance

Infection of rice roots by *H. oryzae* usually results in stunting and a decrease in number of tillers and root weight (Mathur and Prasad 1972). *H. oryzae* can cause varying degrees of yield loss (in terms of number of panicles and grain weight) depending on the nematode population density (Henje *et al*. 1994). It has been reported that *H. oryzae* caused yield losses of rice up to 10 to 30% in China (Liao *et al*. 2000). Microplot experiments in Senegal established that *H. oryzae* can cause a yield loss of 42% when fertilizers are not applied, with nematode population at harvest of 3200-6000 nematodes per dm^3 of soil, and 5-30 nematodes per g root. Even when rice is grown in the best conditions with adequate fertilizers, yield losses are 23%, with nematode population at harvest of 1500-2500 per dm^3 of soil and 90-410 nematodes per g root were reported by Fortuner (1985). Babatola and Bridge (1979) showed that the population levels of 100 to 1000 nematodes per plant could cause significant reduction in rice growth. Inoculation with 1 and 10 *H. oryzae* per g soil caused 27 to 40% yield loss (Jonathan and Velayutham 1987), and 16% reduction in number of panicles and 32% loss in grain weight were observed with the inoculation of 1200 *H. oryzae* per plant (Yamsonrat 1967).

H. oryzae population of 100 per plant reduced grain yield by 35% (Mathur and Prasad 1972). In microplots, natural population of 29-68 *H. oryzae* 500 per cm^3 of soil at transplanting reduced grain weight by 13.8 to 19.2% (Venkitesan and Charles 1979). In non-fertilized fields, the yield loss may be up to 31% as against 19% in fertilized plots (Islam *et al*. 2004). Yield losses caused by *Hirschmanniella* spp. are influenced by soil fertility (Fortuner and Merny 1979), age of plant when infected

(Panda and Rao 1971), number of crops per season and flooding (Khuong 1987), and seasonal climatic conditions (Mathur and Prasad 1972).

2.2.2 Occurrence

The rice root nematode, *Hirschmanniella oryzae* (Van Breda de Haan 1902), Luc and Goodey 1964, is the important nematode pest of the rice crop in many countries (Carmen *et al*. 2016). In a survey of the occurrence and distribution of plant-parasitic nematodes in the Gambia, 17 genera were reported. The potentially important nematodes included *Hirschmanniella oryzae* on rice, *Heterodera gambiensis*, *Tylenchorhychus* spp., *Xiphinema* spp., *Hoplolaimus pararobustus* and *Peltamigratus* spp. on millet, sorghum and maize, *Helicotylenchus pseudorobustus* on cotton and *Helicotylenchus multicintus* on banana (Bridge *et al*. 1978). In Bangladesh, *Meloidogyne graminicola*, *Hirschmanniella oryzae*, *Tylenchorhynchus*, *Longidorus* spp. and *Xiphinema* spp. occurred in 72% of the sites surveyed in 9 deepwater rice areas (Page *et al*. 1979). In Venezuela, the most important nematodes are *Hirschmanniella* spp. (*H. oryzae*, *H. spinicaudata*, *H. caudacrena*), *Tylenchorhynchus annulatus*, *Meloidogyne salasi*, *Mesocriconema onoense*, *Helicotylenchus concavus*, and *Pratylenchus zeae* (Medina *et al*. 2009).

In Myanmar, it was reported that ufra disease caused by *Ditylenchus angustus* was the first rice nematode disease found in Ayeyarwaddy Delta region in 1939 and used to be severe in lowland areas (Ou 1985). White tip disease caused by *Aphelenchoides besseyi* was also known to occur in rice fields of Myanmar (Aung *et al*. 1993). The initial survey and identification of soil and plant-parasitic nematodes associated with economic crops in Myanmar was done by Mya (1983). She reported that *Meloidogyne graminicola*, *Hirschmanniella oryzae*, *Hoplolaimus*, *Criconema* and *Macrophosthonia* were found in paddy soil of Myanmar. Presently root rot caused by *Hirschmanniella oryzae* is the second most important disease among rice nematode diseases in Myanmar. It had been reported that this disease became widely distributed in Western Bago Region including, Pyi, Paukhaung, Thegone, Natalin, Zigone and Paungte Townships. It was also observed that the disease was most commonly found in Kyaukse, Pyinmana and Patheingyi Townships (Myint *et al*. 2004).

Maung (2011) collected soil and root samples from 539 fields from 11 monsoon rice varieties in 12 regions in Myanmar. In the aboved mentioned survey, it was found that 90% of the rice fields were infested by *H. oryzae* in the major rice production regions, Bago (Pyay, Nattalin, Letpadan and Kyauktaga), Ayeyarwaddy (Myaungmya and Nyaungdone) and Yangon Division (Hmawbi, Hlegu and Hlaingtharyar) in Lower Myanmar and Mandalay Division (Kyaukse, Patheingyi and Pyinmana) in Upper Myanmar. According to disease surveys conducted in Nay Pyi Taw during 2011-2012 rice growing seasons, commonly occurred rice diseases were sheath blight, sheath rot, false smut, bacterial blight, bacterial leaf streak, narrow brown leaf spot and root rot nematode diseases (Myint *et al*. 2012).

H. oryzae is the most frequently found and considered as the most important rice root nematode species in most of the rice growing areas in Asia (Verma 2000; Bridge *et al*. 2005). Nematode damage to plants as a rule only becomes apparent several years after establishment of an infestation, by which time the nematodes have increased their numbers and widened their distribution (Kleynhans 1999).

2.2.3 History and distribution

Hirschmanniella oryzae was first recorded from Indonesia by Van Breda de Haan in connection with ‘mentek’ disease of rice in 1902 and was named ‘*Tylenchus oryzae*’. Goodey 1936 described and illustrated the nematode in detail, having renamed it ‘*Anguillulina oryzae*’ in 1932. Thorne (1949) established the genus ‘*Radopholus oryzae*’. Van der Vecht and Bergman (1952) made extensive studies on the nematode and its relation to ‘mentek’. They failed to prove that ‘mentek’ was not due to the nematodes but found that the nematode injured the roots and caused growth retardation. Luc and Goodey (1962) differentiated the genus *Hirschmannia* from *Radopholus* and called the nematode *Hirschmannia oryzae*. Later, they found that generic name *Hirschmannia* was preoccupied and a new generic name *Hirschmanniella* was used. This name is now called *Hirschmanniella oryzae* (Van Breda de Haan 1902) Luc and Goodey 1964.

Hirschmanniella oryzae is apparently widely distributed in all rice growing regions of the world (Ou 1985) such as Indonesia (Van der Vecht and Bergman 1952), Bangladesh (Timm 1956; Sher 1968), Japan (Kawashima 1963), Philippines (Timm 1965), Nepal, Sri Lanka, Egypt, West Africa, Brazil, and Iran, Ghana, Malaysia, Nigeria, Sierra, Leone, USA, Venezuela and Madagascar (Sher 1968), Korea (Park *et al*. 1970), India (Mathur and Prasad 1971), Senegal (Fortuner and Merny 1973), Myanmar (Mya 1983), China (Yin 1986), Vietnam (Khuong 1987), Costa Rica (Lopez and Salazar 1987), Niger (Sikora *et al*. 1988), Portugal (Reis 1990), South Africa, Pakistan and EL Salvador (EPPO 1999).

2.2.4 Damage symptoms

Symptoms induced by rice root nematodes in the field are never specific and therefore not easily identifiable and usually not recognized if the root system is not heavily infected (Edward *et al*. 1985; Bridge *et al*. 2005). Infected roots may first show a yellowish to brown color that eventually darkens, and heavily infected roots may decay after turning brown or black (Mathur and Prasad 1972; Ichinohe 1972). These below ground symptoms begin by the formation of small brown lesions at points where nematodes have ruptured the surface and entered. Following these early symptoms, damaged epidermal cells may become necrotic and cavities may form inside the roots as a result of damaged cortical cells (Mathur and Prasad 1972). Infected roots are yellow initially and finally they rot. The rot is caused in part by transcriptional reprogramming by the nematode that induces programmed cell death, oxidative stress and obstruction of normal root metabolic activity (Larry and Maurice 2013).

Rice root nematodes are common in rice seed-beds and seedlings with heavy infection show growth retardation, reduced survival, and decreased tiller emergence with discolored older leaves, delayed flowering and late maturation (Taylor 1969). Retardation of growth rate occurs especially in early growth, with a decrease in tillering. Yellowing of rice plants is observed occasionally, and flowering can be delayed up to 14 days. The roots invaded by the nematodes are characterized by yellowish to rusty brown discoloration of the cortical tissue with the darkest colour around the stele and the base of the root hairs where the nematodes are found most frequently (Van der Vecht and Bergman 1952).

Sivakumar and Seshadri (1969) and Mathur and Prasad (1972) observed that cell wall thickening and dissolution in rice roots around sites infected with *H. oryzae*. Babatola and Bridge (1980) and Vovlas *et al*. (1996) reported that the rice root nematode is commonly found in the aerenchyma tissues of roots which are adapted for its survival in aquatic conditions permitting the movement of oxygen from leaves to roots. Hence, the damage occurred to these tissues due to nematode infection is likely to restrict the passage of oxygen, thus placing rice roots under oxygen stress. As a result, the aerial respiration of rice root cells may be affected.

Kawashima and Fujinuma (1965) reported from laboratory tests that nematode caused changes in physiological functions of rice roots, resulting in a staining with iron oxide. They found that oxidative activity of rice roots was less in nematode-infested than in nematode-free roots. Sturgis (1936) found that the incrustation formed on and around older rice roots was composed of iron and manganese oxides. Kawashima (1964) revealed that the invasion and parasitism of roots by the nematode starts several days after rice transplantation. The invasion is rapid at high temperature; the optimum temperature is 25-30˚C and the death of cells is seen to occur after invasion. Because of the accumulation of substances stained with hematoxylin near the head of the nematode, he believed that substances secreted by the nematode caused the death of cells.When rice seedlings were grown in soil amended with healthy rice roots and the nematode was added, the stunting effect became more severe. Infestation of rice seedling roots by *H. oryzae* caused increases in the level of phenolic compounds and in the activity of polyphenol oxidase, catalase and beta-glucosidase in the roots (Sman 1936).

2.2.5 Taxonomic classification

Domain: Eukaryota

Kingdom: Metazoa

Phylum: Nematoda

Family: Pratylenchidae

Genus: *Hirschmanniella*

Species: *oryzae* (CABI 2018)

2.2.6 Morphology

Female body is straight or slightly arcuate ventrally; transverse striae are distinct, about 1.6 µm apart near middle. Lateral fields are not areolated except in neck region but occasional incomplete areolation may occur in tail region. Lip region is low, continuous, flattened with rounded edges, with 3-5 (usually 4) annules. Outer margins of head skeleton are prominent, extending posteriorly about 3 body annules. Anterior cephalids 3 annules are behind lip region, posterior ones are indistinct; hemizonid 2-3 annules long, 2-4 annules are anterior to excretory pore. Stylet knobs are rounded, about 3.2 µm across. Median oesophageal bulb is oval. Oesophageal glands are elongate, mostly ventral to intestine; dorsal gland is anterior and usually with large vacuoles and more granular cytoplasm. Vulva and vagina are prominent.

Spermatheca is oval or sometimes round, with sperms. Ovaries are paired, outstretched, mostly with a single row of oocytes. "Thorneian cells" appear to be of hypodermal origin and serve to connect the intestinal epithelial cells with the hypodermis. Intestine is not overlapping rectum. Tail is elongate-conoid, length 4.3-5.5 times anal body width, with a rounded terminus bearing a sharp ventral mucro, with 49-54 annules ventrally. Striae are extending almost to terminus without a hyaline terminal portion. Phasmids are small, pore-like, 12-17 annules from tail terminus. Male lip region, stylet and oesophagus are as in female. Bursa is crenate, arising a little anterior to head of spicules and ending near phasmids. Spicules are distinctly cephalated, slightly arcuate, ventrally flanged in distal third. Gubernaculum is simple, non-protruding. Tail terminus is rounded with a ventral mucro or a sharp projection (Siddiqi 1973; CABI 2018).

2.2.7 Biology

Hirschmanniella species are migratory endoparasites of roots (Bridge *et al.* 1990). All stages of the life cycle are infective. Eggs of *H. oryzae* are deposited in the roots a few days after invasion by adult female nematodes, and hatching occurs 4-6 days after deposition (Mathur and Prasad 1972; CABI 2018). The adults of both sexes invade young roots at some distance from the tip, and often use the opening produced by other individuals (Van der Vecht and Bergman 1952). The nematodes penetrated the roots of healthy rice plants, feeding on the parenchyma tissue, multiplying in them, the root cortex becoming discolored. The nematode moves through the air channels between the lamellae of the radial parenchyma of the root. They produce cavities and channels through the cortex which become necrotic for some distance into the root (Hollis and Keoboonrueng 1984). Large number of nematodes can be found at the base of the seedling coleoptile. Rice root nematode can penetrate anywhere along the roots of rice except at the tips or the thin lateral roots. The penetrated nematodes can either enter the root completely or simply embed their heads into the cortex. The rice root nematode makes tunnels after penetration into roots which are almost perpendicular to the root surface (Youssef and Eissa 2014). After penetration, *H. oryzae* positioned near the vascular bundle and formed big cavities in the cortex. The nematode was observed to feed on cortical cells, seldom reaching the central vascular region and stele (Mathur and Prasad 1972).

2.2.8 Life cycle and reproduction

Rice root nematode, *H. oryzae* is sexually dimorphic, i.e. sexes are separate, and reproduction is amphimictic or bisexual; both sexes are required (Mai and Mullin 1996). There are 4 molts that occur throughout the life cycle, the first of which occurs in the egg (Mehmet 2004). The hatched stage 2 juvenile, while migrating through and feeding on cells in the cortex, then completes 3 successive molts, and after the final molt, immature adult females and males emerge with gonads not yet fully developed (Thorne 1961).

The life cycle of *H. oryzae* can be completed in about 33 days at 28±2˚C under *in vitro* conditions. The female commences oviposition a few days after invasion. The minimum time for development from egg to adult is at least one month, and the multiplication factor per generation may be as high as thirteen. Eggs of *H. oryzae* are deposited in the roots a few days after invasion, and hatching occurs 4-6 days after deposition. Eggs hatched into larvae whose appearance and structure were similar to those of the adults. Larvae grew in size and each larva stage was terminated by a molt. *H. oryzae* had four larva stages, with the first molt occurred in the egg. After the final molt, the larvae differentiated into adult males and females (Mehmet 2004). Different larval stages can be distinguished by gonad length. Body and stylet lengths, and the distance from head to anus are additional characters to differentiate these stages (Mathur and Prasad 1972).

The life cycle from second stage larva to second stage larva was completed in 33 days. The L_2 to L_2 life cycle of *H. oryzae* was completed under gnotobiotic conditions at 28±2˚C in 33 days. The L_2 moved to the root tips and began feeding within 1.2 hour after inoculation. Feeding lasted for 12 to 24 hours, then the L_2 became immobile and remained positioned like a “C” or a closed circle. The second molt (M_2) started 3 days after inoculation. The most significant change during molting occurred in the oesophageal region. During the first 12 to 24 hours of molting, the stylet shaft, oesophageal lumen and median bulb became invisible. Only the stylet cone remained discernible. Twelve hours later, the new cuticle became visible inside the old one, followed by the appearance of the new stylet shaft. Then the oesophageal lumen and the median bulb emerged and gradually became more distinctive. The larva body progressively elongated until it was confined by the old cuticle. At this time the new stylet began to probe the old cuticle at the rate of once every 5 to 15 seconds, associated with contraction of the median bulb once every 4 to 6 probings. The nematode finally broke through the old cuticle and migrated out. This molting period (M_2) lasted for 2 days. The third stage larva (L_3) began feeding again. At 9 days after inoculation the L_3 entered the third molting (M_3) period, which lasted for 3 days and resulted in the emergence of the fourth stage larva (L_4). The L_4 started feeding on the roots again, followed by the fourth molting period. Larvae that developed into males started the fourth molt (M_4) 19 days after inoculation. By the end of the 6 days molting period, the male gonad, the spicules and the caudal alae had formed and the male migrated out of the old cuticle 25 days after inoculation. Larvae that developed into females started M_4 at 19 days after inoculation, which lasted for 6 days. By the end of the molting period, the female gonads and the vulva had formed. The female migrated out of the old cuticle 25 days after inoculation. The first eggs were laid 27 days after inoculation (Mehmet 2004).

All stages of the life cycle are infective. Eggs of *H. oryzae* are deposited in the roots a few days after invasion by adult female nematodes, and hatching occurs 4-6 days after deposition (Van der Vecht and Bergman 1952; Mathur and Prasad 1972). The length of life cycle is variable. In North India, there are only one generation of *H. oryzae* in a year (Mathur and Prasad 1972), two generations in Japan (Kawahara and Iyatomi 1970; Ou 1985), and three generations in Senegal (Fortuner and Merny 1979). In Java, the minimum duration of development from egg to adult is one month, with a multiplication rate of 13 per generation (Van der Vecht and Bergman 1952; CABI 2018).

2.2.9 Feeding behaviour and histopathology

According to Babatola and Bridge (1980), four distinct feeding phases were observed in *H. oryzae*: (1) probing or exploration, (2) stylet penetration, (3) salivation and (4) ingestion. Probing or exploration of roots was followed by stylet penetration, which was normally at right angles to the root surface. Rate of stylet thrust was initially low and irregular. After stylet penetration, the nematodes remained motionless. During salivation, the ampula of the dorsal oesophageal gland duct enlarged, followed by a continuous flow of secretions through the stylet into the punctured cell. The cytoplasm of the cell slightly darkened and the flow of secretion gradually stopped as the median oesophageal bulb began to twitch spasmodically. The twitching soon built up into a steady pulsation, and a reverse flow was observed of cell contents through the stylet into the nematode. When the cells collapsed, the nematodes withdrew completely from the root, or started to puncture adjacent cells.

Adults and juveniles of all stages invaded roots, often through the same opening. Juveniles and males moved in and out of the roots more frequently than did gravid females, which remained inside the roots and laid eggs along their burrowed channels. The nematode fed at all points along the root but were found more often around root tips and along lateral roots. During salivation and ingestion, *H. oryzae* became coiled like an open C posture. Damage to the tips of main roots was severe, resulting in cessation of growth and development of lateral roots around the tips. Necrosis was observed in two to four cells around the feeding and invasion sites in roots. The burrowing activities of *H. oryzae* almost severed primordia of lateral roots. Such root primordia turned brown when killed. Cortical and lacunal cells were collapsed, and cell walls were broken by nematode feeding. The nematodes moved progressively from necrotic areas to healthy cortical tissues as necrosis spread. Necrosis, together with separation and breakdown of cell walls and collapse of cells, ultimately killed roots invaded by as few as 10 nematodes. The feeding and invasion of root tissues, both inter-cellularly and intra-cellularly, caused extensive mechanical damage. Feeding of the nematodes caused cessation of growth of the main roots and a proliferation of lateral roots close to the damaged root tips, apparently from breaking of the apical dominance of the roots. The creation of large cavities in the cortical parenchyma of the roots was often accompanied by rot-inducing microorganisms and caused disruption and eventual death of the roots. The feeding activities of *H. oryzae* appears to have deleterious effects on the rice plant in terms of mechanical damage to the stele and parenchyma tissues, disruption of growth, and predisposition of the rice root to secondary infection through the feeding and invasion sites. This results in poor grain yield (Babatola and Bridge 1980).

2.2.10 Survival and dissemination

H. oryzae survives in ratooning rice roots and in undecayed roots of rice stubble, in weeds and other hosts (Ichinohe 1972; Mathur and Prasad 1973; Feng 1986). *Hirschmanniella* spp. can also survive in soil. They survive longer in roots than in soil, but survival of root population is shorter in flooded soil due to the more rapid decay of roots. *H. oryzae* can survive in the soil for at least 5 months in the absence of a host plant and up to 12 months, by means of quiescence, in soil that is not continually wet. *H. oryzae* survives poorly in dried fields but may overwinter in dead roots as eggs if kept moist (Fortuner and Merny 1979).

Two factors are especially important for the survival of *Hirschmanniella* spp. in the interval between two rice crops: soil moisture and presence or absence of host roots. As these nematodes are endoparasites, after rice has been harvested high population is still inside the roots and a variable number are found in the soil. In the laboratory, Van der Vecht and Bergman (1952) observed that, in every condition a high proportion of the initial population of *H. oryzae* was still alive after ten weeks. They noted that the death rate in a drying soil was about the same as in a wet soil or sand and that it is generally higher in soil or sand without roots than with roots. Thorne (1961) found numerous quiescent *H. oryzae* individuals in the dried up clay of rice fields in Thailand and Philippines during the dry season. In Japan, Kawashima (1962) stated that nematodes survive in winter as juveniles or adults in dead rice roots; in very wet fields they often hibernate in the egg stage but they seldom survive in well drained fields due to drying of soil in spring. In Korea, *H. oryzae* can survive for up to seven months in the water of flooded rice fields (Park *et al*. 1970). In India, Mathur and Prasad (1973) have shown that, in aerated water, 50 to 60% of the nematodes were still alive after eight weeks but only 0 to 20% survived after sixteen weeks: in a soil sample allowed to dry for ten months and containing rice roots, 240 nematodes per liter of soil were still alive while a soil without roots contained no more living nematodes after the same period.

In the Senegal River Delta, *H. oryzae* can withstand very severe dry seasons between January and August. Nematodes leave the roots in August when the water comes. In a microplot trial with *H. oryzae* if soil remains wet after the harvest, roots decay and the nematodes in the roots are released into the soil; population decrease slowly and become eradicated after one year. In dry soil, both root and soil population decrease slowly until, after flooding, population are released from the roots into the soil (Fortuner 1977). This nematode is perfectively adapted to the constant flooded conditions in which irrigated rice is often being grown in the lowlands (Fortuner and Merny 1979). It is one of the few plant-parasitic nematode species that can survive under anaerobic conditions (Babatola 1981).

H. oryzae survived very well at pH’s ranging from 5 to 9. Soil temperature is one of the most important environmental factors influencing the development of plant-parasitic nematode species (Babatola 1981). Youssef (1998) observed that there was a positive correlation between the root population density of *H. oryzae* and soil temperature. A soil temperature ranging from 21 to 28˚C is optimal for multiplication. In fallow field soil, population of *H. oryzae* can survive high temperature of 34-45˚C and low temperature of 8-12˚C (Mathur and Prasad 1973). *H. oryzae* adapts better to clay soil than to sandy soil. In India, the highest population of this nematode species was found in heavy clay soil (Fortuner and Merny 1979). Numerous quiescent *H. oryzae* were found in dried up clay in rice fields in Thailand and the Philippines during the dry season (Thorne 1961).

Population in the soil decrease at flowering stage and increase after harvest as the nematode leaves the roots when the panicles appear (Youssef and El-Hamawi 1996). However, high population density can still be found in the roots while variable population numbers are found in the soil after harvest. Rice root nematodes cannot survive when the moisture content in the root is less than 13% and the survival of second-stage juveniles (J_2) of *H. oryzae* failed when the relative humidity was 50% (Babatola 1980; Wang *et al*. 1992).

The nematodes move along with mud water and rice seedlings during transplanting and canal-irrigation (Pokharel and Regmi 2000). *H. oryzae* is spread in irrigation and flood water, and in soil adhering to implements and field workers. Where there is a long history of rice cultivation, the nematodes are likely to be widespread. The nematodes are also disseminated to the field in roots of rice seedlings from nurseries (Fortuner and Merny 1979).

2.3 Host Range of Rice Root Nematode, *Hirschmanniella oryzae*

The rice root nematodes, *Hirschmanniella* spp. have a large host range (Bridge *et al*. 1990). Although rice is the preferred host of *H. oryzae*, other crops such as okra, cotton, maize, wheat and sugarcane (Babatola 1979), and weeds of the families Cyperaceae and Gramineae are also known to be hosts of *H. oryzae* (Van der Vecht and Bergman 1952; Mathur and Prasad 1973). However, cowpea, pigeon pea, soybean, groundnut, tomato, sweet potato, bulrush millet, finger millet, sorghum and onion did not support *H. oryzae* population build up (Edward *et al*. 1985; Prot 1992; Korayem 1993; Bridge *et al*. 2005).

2.4 Economic Threshold Level of *Hirschmanniella oryzae*

Ying *et al.* (1996) proposed the injury level of *H. oryzae* to rice as 15 nematodes per g root at the end of maximum tillering stage and that could be applied as an economic threshold level. In Vietnam, economic damage by *Hirschmanniella* spp. occurs when 40 or more nematodes are present in a rice hill 3 weeks after transplanting, equivalent after multiplication to 800 nematodes per hill at heading (Khuong 1987). Prasad *et al.* (1987) observed that at low levels of nematode infection (below 50 per g root) there was no appreciable reduction in important biochemical components like chlorophyll, starch and proteins in plants indicating no apparent change in metabolic processes. Park *et al.* (1970) rated the varieties, which had more than 1000 nematodes per 10 g roots as susceptible to *H. oryzae.* Safdar *et al*. (2011) found that among ten rice cultivars, only Basmati Pak supported nematode population below the damage threshold level (5-30 nematodes per g root).

2.5 Control Measures of Rice Root Nematode, *Hirschmanniella oryzae*

Although Van der Vecht and Bergman (1952) stated that cleaning the fields, keeping a good fertility and proper agronomic practices should allow rice to withstand attacks by *H. oryzae* without noticeable losses, control of rice nematodes is difficult and several kinds of control measures should be considered including agricultural practices, varietal resistance, chemical control and biological control.

2.5.1 Cultural practices

Numerous weed species are common in rice fields and can reduce yield and quality of rice by competing for light, nutrients and space. In addition, their seeds can be a contaminant of harvested grain. Although competition is the most important effect, weeds have on crop production; they are also alternative hosts for plant-parasitic nematodes and have long been recognized for their ability to maintain nematode population (Anwar *et al*. 2011). Therefore, the management of weeds, which are generally good hosts of *H. oryzae*, will reduce nematode population both in the absence of rice and during growing season of the crop. In Japan, early planting and direct sowing, which both reduce initial infection of *Hirschmanniella* spp. (Sato *et al*. 1970). Higher population of *H. oryzae* was reported intermediate or dry nurseries or from directly sown fields (Miura and Shoji 1964). Higher population was found in transplanted rice roots than in directly sown ones (Nakazato *et al*. 1964). Time of transplanting is also important, for instant in Punjab, less build-up of *H. oryzae* occurred when basmati rice is transplanted later in mid-July compared with mid-June (Randhawa *et al*. 1991).

It is well documented that the population of plant-parasitic nematodes in a field can increase considerably when the same host plant is grown in the same field year after year. The build-up of this population can be prevented or limited by crop rotation with non-host plants, less susceptible or resistant rice varieties of the same crop or a completely different crop. In fact, crop rotation is considered one of the most effective and acceptable alternatives to chemical control of plant-parasitic nematodes for the small-scale farmers of low-input agricultural systems in the tropics (Bridge 1996). This management practice is also favoured because of the economic constraints in these tropical low-input agricultural systems (Luc *et al*. 2005). Rotation of crops is not possible in continuous rice cropping, but is often normal practice where a single wet season rice crop is followed by dry season crops. Rotation with non-host dry season crops such as cotton, cowpea, groundnut, millet, onion, pigeon pea, sorghum, soybean, sweet potato, tobacco and wheat can reduce the population density of *H. oryzae* in the field (Mathur and Prasad 1973; Babatola 1979; Gao *et al*. 1998).

2.5.2 Use of resistant cultivars

The use of resistant varieties is considered a promising and often effective management practice to improve crop yields in the presence of plant-parasitic nematode population density that exceed the damage threshold level. It is also an environment-friendly approach to nematode management in contrast with the currently much disputed use of expensive and hazardous chemicals (Sasser 1989; Reversat *et al*. 2003; Starr and Roberts 2004).

Rice is the good host of *Hirschmanniella* spp. and cultivars (cv.) supporting relatively low nematode numbers have been rated as ‘resistant’ (Arayarungsarit *et al*. 1986; Rao *et al*. 1986). *H. oryzae* population density was consistently influenced by cultivar (Coyne *et al*. 1999). Six cultivars belonging to Japonica (Sakha 101, Sakha 102, Giza 176 and Giza 177), Indica-Japonica (Giza 178) and Indica (Yamini) were evaluated against *H. oryzae* infection, and the rice varieties belonging to Japonica group were more susceptible than the other two groups (Youssef 1998; 1999). The plants of fine grained rice varieties were found to escape the attack of the rice-root nematode, when grown with coarse grained varieties (Birat 1965). Selections from crosses Taichung Native 1 x T 141, Peta x DgWg and Taichung Native 1 x CO 29 appeared resistant to the rice-root nematode (Anon 1970).

Out of the rice varieties, IR 20, IR 50, Paiyur 1, TKM 6 and TKM 9 evaluated against *H. oryzae,* only TKM 9, the deep rooted rice cultivar was found to be resistant to the nematode at all growth stages (Ramakrishnan *et al.* 1984*).* Rice varieties, Kao

Paung Klang, Kao Paung, Kao Tah Jue, Kao Yaun and Kao Klang Pee with Rf values between 1.54 to 1.98 after 60 days of transplanting were classified as resistant to *Hirschmanniella* spp. (Arayarungsarit *et al.* 1986*).* Rice varieties and breeding lines, viz., Annapurna, CR 44-140-2-1051, CR 130-203, CR 141-6058-1-35, CR 142-3-2, CR 294-548, CR 320, RP 1155-128-1, MTU 8 and PTB 27 were found to be resistant or tolerant to the rice-root nematode (Rao *et al.* 1986*).* Higher population build-up of *H. oryzae* was observed in Pak Basmati and Basmati 370, while Basmati 385 supported lowest population among 7 basmati rice cultivars evaluated (Randhawa *et al.* 1991; 1992). All the eleven rice cultivars viz., PR 103, PR 106, PR 108, PR 109, Jaya, PAU 1126, PAU 1628, PUSA 44, PUSA 429, HKR 120 and HKR 126 were found susceptible to *H. oryzae,* while PR 108 and PAU 1126 having the greatest nematode build-up and PR 103 the least (Singh *et al.* 1992*).*

It had been reported that *H. oryzae* is more pathogenic on the recently introduced high-yielding varieties compared with the local varieties in Bago Division which is one of the major rice producing regions (Thein 2003). Than (2003) observed that the rice varieties, Sinekari-3, Yezin-3, Shwethweyin and Theedatyin were resistant to *H. oryzae*. Yatanartoe and Shwemanaw varieties were resistant to *H. oryzae* (Soe 2011).

2.5.3 Fertilizer application

Nitrogenous fertilizers favour the development of nematode population and increase the decay and browning of roots. The height and tillering of plants are improved (Ishikawa 1965). Inversely, potassium fertilizers and compost keep nematode population at a low level. Potassium also has a beneficial effect on the quality of a wide range of crops, especially in terms of improved protein quantity and quality. Potassium can decrease the incidence of plant diseases and reduce abiotic stresses, particularly cold stress. The element may have a direct action on pathogen penetration, lesion size and on inoculum density (Mishra and Das 1959). Pokharel *et al*. (2004) observed that significantly lower nematode population was observed in plots fertilized with 100 kg of potassium per hectare. The population of *Hirschmanniella* spp. reduced with application of potassium (Ichinohe 1972). The application of higher doses of potassium might help to reduce *H. oryzae* population and the damage they cause to rice (Pokharel 1995). Soe (2011) reported that the application of potassium (muriate of potash) at the rate of 45 kg per hectare could be applied for management of rice root nematode, *H. oryzae.* Tomonaga and Kurokawa (1964) have reported that calcium silicate and compost reduce *Hirschmanniella* spp. population and increase yield. Fertilizers containing 64.3% and 30% of silicic acid and various micro-elements reduced *Hirschmanniella* spp. 15% and increased yield up to 5% (Yokoo and Morimitsu 1969).

CHAPTER III
MATERIALS AND METHODS

3.1 Determination of Population Density of *Hirschmanniella oryzae* in Nay Pyi Taw Union Territory

3.1.1 Survey site and samples selection

The survey was carried out during 2017 summer rice growing season (March-June) in five townships (Lewe, Tatkon, Pyinmana, Zabuthiri and Dekkhinathiri) of Nay Pyi Taw Union Territory (Figure 3.1). The selected townships and villages were based on the rice growing areas of Nay Pyi Taw Union Territory (DoA 2017). Three villages in each township and three fields in each village were selected for survey. A total of 15 villages and 44 rice fields were visited, and soil and root samples of 5 summer rice varieties (Manawthukha, Sinthukha, Shwethweyin, Palethwe and Yet-90) were collected from the fields in different townships (Plate 3.1). The varieties selection was done from the most commonly grown ones in each village. All the sampling was done at the maximum tillering stage of the rice plants. Numbers of samples collected from each township were shown in Table 3.1.

3.1.2 Selection of farmers

Three contact farmers in each village were selected. A total of 44 farmers - 9 in Lewe, 9 in Tatkon, 8 in Pyinmana, 9 in Zabuthiri and 9 in Dekkhinathiri were interviewed. The questionnaires were addressed using structured questionnaires with the cropping sequences, sowing methods, variety selection and soil type (Appendix 2).

3.1.3 Sampling procedure

In each rice field, a survey area of 20 × 20 m was marked off with a measuring tape in the centre of the field and then soil and root samples were collected from each rice hill at nine sampling points roughly in equal distance from one another on the two cross diagonal lines. The selected plant was uprooted and the soil from rhizosphere (about 100 g soil from each sampling point) was collected up to a depth of 15 cm (Plate 3.2). The composite soil and root samples from each rice field were placed in different plastic bags, labelled and kept at room temperature for one night before extraction for determination of nematode population density.

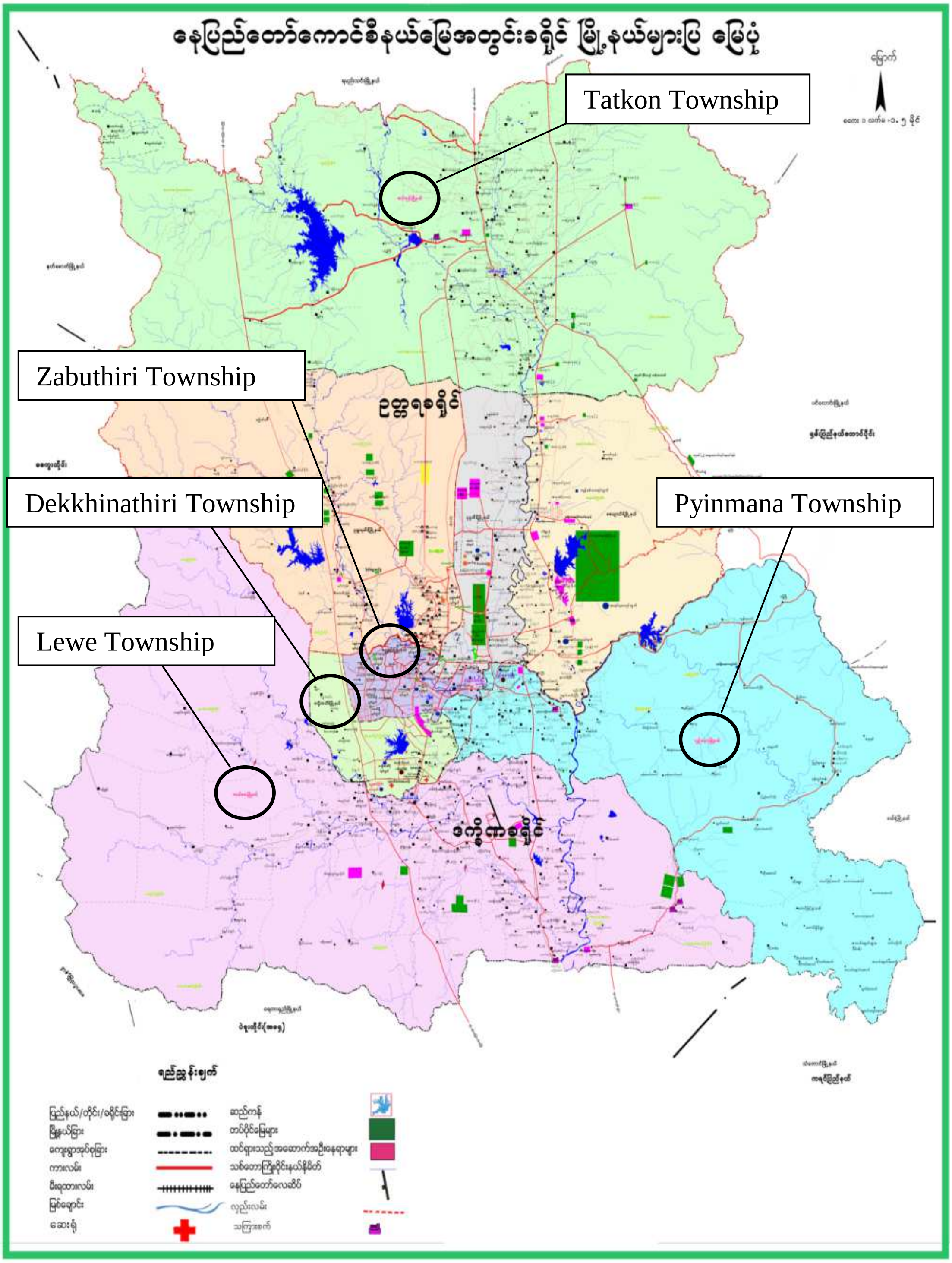
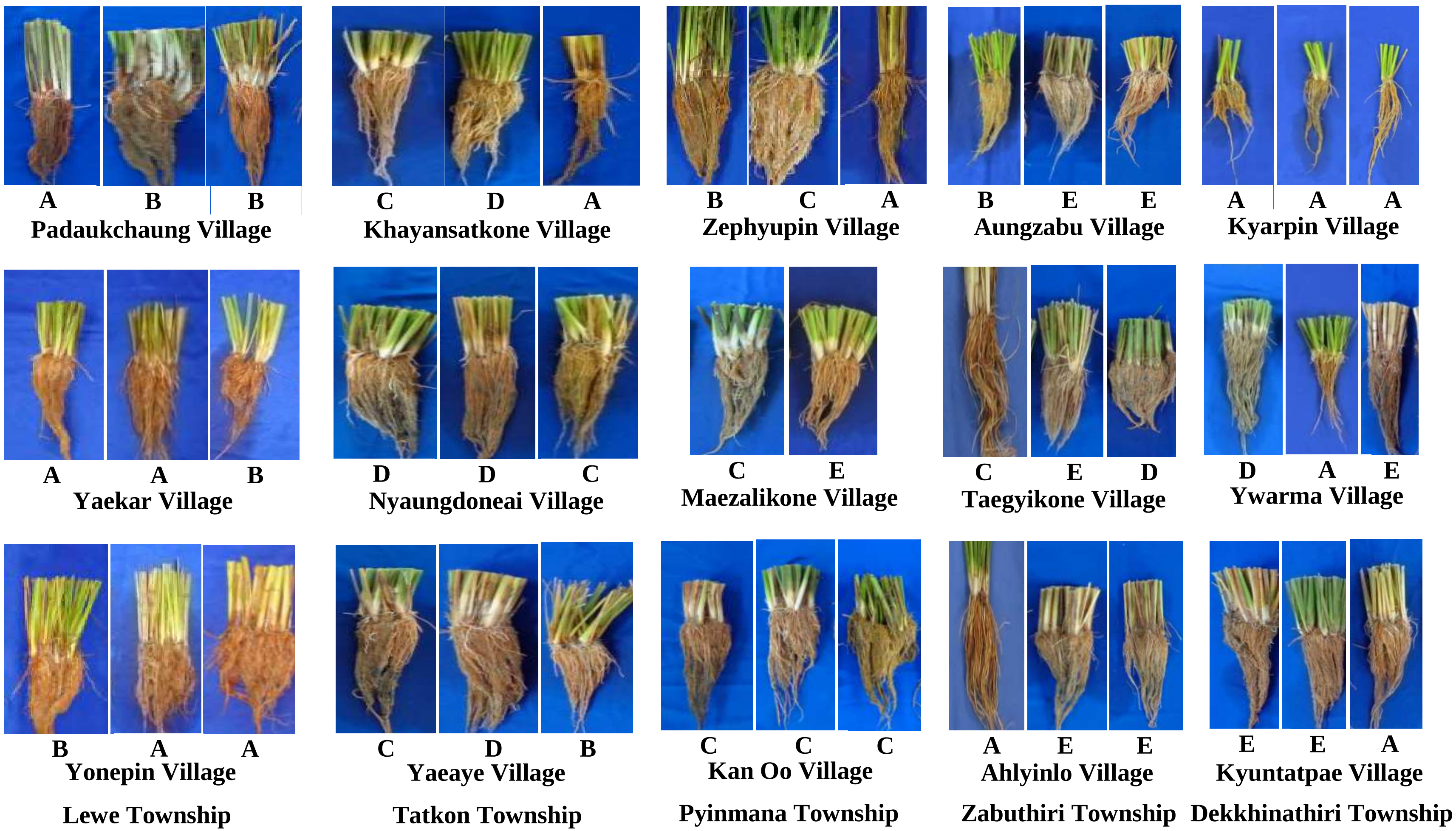


Figure 3.1 Survey areas in Nay Pyi Taw Union Territory for the occurrence of *Hirschmanniella oryzae* on summer rice varieties



A = Yet-90, B = Shwethweyin, C = Palethwe, D = Sinthukha, E = Manawthukha

Plate 3.1 Root samples from five townships in Nay Pyi Taw Union Territory

Table 3.1 Number of samples collected from each township of Nay Pyi Taw Union Territory

Sr. no.	Township	Area sown (summer) (ha) [x]	No. of samples
1	Lewe	2730	9
2	Tatkon	1552	9
3	Pyinmana	802	8*
4	Dekkhinathiri	694	9
5	Zabuthiri	325	9
	Total	**6103**	**44**

[x] Rice growing areas of 2016-2017 in each township (DoA 2017).

* One sample in Pyinmana Township was discarded because it had only one variety which was not grown in other townships.

(A)

(B)

(C)

(D)

Plate 3.2 Sampling procedure in field survey:

(A) Marking off survey area (20×20 m) with a measuring tape in the centre of the field

(B) Uprooting the selected plant

(C) Collection of rhizosphere soil (about 100 g)

(D) Rinsing the rice plants roughly in the field

3.1.4 Determination of nematode population density

In the laboratory of Plant Pathology Department, Yezin Agricultural University, the soil samples from each rice field were pooled and the nematodes were extracted from a 150 g sub-sample using Whitehead's tray method (Whitehead and Hemming 1965). As the procedure, the soil was spread over a muslin cloth placed in a plastic sieve (8×10 inches). And then, the sieve was placed onto a plastic tray (9×12 inches). About 300 ml tap water was carefully poured from the edge of the tray until the soil layer looked wet. The root samples were washed again with tap water, pooled, chopped into approximately 1 cm pieces and thoroughly mixed. A 50 g sub-sample of chopped root pieces was taken from each pooled sample and the nematodes were extracted by using Whitehead's tray method (Plate 3.3). After 24 hours, the nematodes which had moved through the sieve into the water were collected in a beaker. And then the suspension was allowed to settle for 1 hour and concentrated into a 200 ml suspension. 1 ml nematode suspension was taken by 5 ml syringe and one syringe was used for one sample. The juvenile and adult *H. oryzae* were counted under the compound microscope (OPTIMA, Model: G-206) with 5 ml counting dish for 10 times and calculated the average value for each rice variety and for each field.

3.1.5 Data recording

Agricultural practices and field records were collected from the farmers' interview. Nematode population density of soil and root samples, frequency of occurrence, prominence value of *H. oryzae*, cropping sequences and sowing methods were recorded.

Frequency of occurrence and prominence value for each township and variety were calculated according to the formula as follows (Orisajo 2013).

$$\text{Frequency of occurrence} = \frac{\text{Number of samples containing a species}}{\text{Number of samples collected}} \times 100$$

$$\text{Prominence value(PV)} = \text{Nematode population density} \times \sqrt{\text{frequency of occurrence}/10}$$

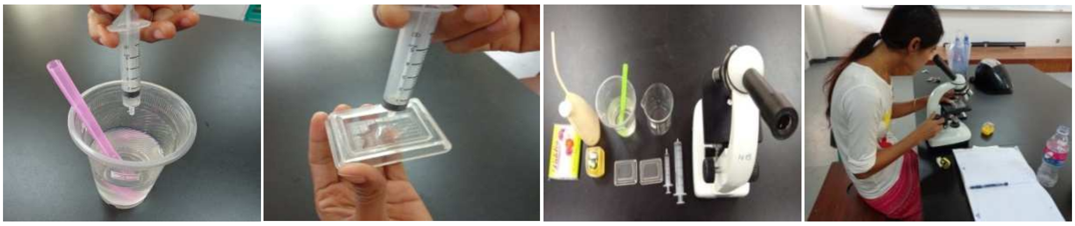
Disease reactions of the summer rice varieties to *H. oryzae* were categorized according to the disease rating scale by de Man (1880) (Table 3.2).

3.1.6 Statistical analysis

Survey data were analyzed by using Microsoft Excel. Descriptive statistics was employed for the analysis of frequency of occurrence, prominence value and population of *H. oryzae* related to different rice varieties among five townships.

(A)

(B)



(C)

Plate 3.3 Procedure for nematodes extraction from root samples:

(A) Rinsing, cutting, weighing and spreading root sample

(B) Pouring water and collection of nematode suspension in the beaker and settling for 1 hr

(C) Transferring 1 ml nematode suspension into 5 ml counting dish with syringe and counting nematodes under compound microscope (OPTIMA, Model:G-206)

Table 3.2 Rating index of rice varieties at maximum tillering stage for resistance against the infection of the rice-root nematode, *Hirschmanniella* spp.

Scale	**Nematode population per g root**	**Host reaction**
1	0	Immune (I)
2	1-10	Highly resistant (HR)
3	11-50	Moderately resistant (MR)
4	51-100	Susceptible (S)
5	>100	Highly susceptible (HS)

(de Man 1880)

3.2 Evaluation of the Response of Tested Rice Varieties to Rice Root Nematode

3.2.1 Test varieties

Twelve rice varieties namely (1) Thukhamwe, (2) Pawsanmwe, (3) Yet-90, (4) Pyitawyin, (5) Salt tolerant Sinthwelatt, (6) Shweasean, (7) Myaungmyamay, (8) Pyimyanmarsein, (9) Submergence-tolerant 1, (10) Shwethweyin (resistant check), (11) Manawthukha (susceptible check) and (12) Sinthukha which were obtained from the Rice Section, Department of Agricultural Research (DAR) were used for this experiment (Appendix 1).

3.2.2 Preparation of test plants

Seeds of tested rice varieties were surface sterilized with 10% sodium hypochloride for 10 minutes and thoroughly washed with sterilized water for 3 times. The surface sterilized seeds were pre-germinated under wet tissue for 24 hours. The sprouting seeds were sown in plastic pot containing sterilized soil. Sterilization of soil was done by autoclaving at 121˚C for 30 minutes. Ten days old seedlings were transplanted individually to a polythene bag (5×9 inches) containing 3 kg of sterilized lowland (sandy loam) soil. Compound fertilizer at the rate of 0.41 g per bag (126 kg per hectare) was applied at basal and mixed with the sterilized soil in polythene bag. Urea (46% N) at the rate of 0.136 g per bag (42 kg per hectare) was applied at 10 days after transplanting, 0.136 g per bag (42 kg per hectare) at 30 days after transplanting and 0.136 g per bag (42 kg per hectare) at panicle initiation stage, respectively.

3.2.3 Inoculum preparation and inoculation

The nematode inoculum collected from Manawthukha rice fields was multiplied on Manawthukha rice variety. The nematodes freshly extracted by Whitehead's tray method from 45 days old rice plants were used as inoculum. The aliquot solution was adjusted to obtain the required inoculum levels. Ten days after transplanting, each variety was inoculated with 2000 *H. oryzae* per plant by syringe into four holes of soil around the rice plant. For each variety, non-inoculated plants were included as control plants. The water level in each bag was kept about 3 cm above the soil layer to maintain its flooded conditions.

3.2.4 Experimental design

This experiment was conducted at the Department of Plant Pathology, Yezin Agricultural University during monsoon rice growing season from June to November, 2017. Factorial experiment was laid out in a completely randomized design (CRD) with 12 treatments and 5 replications (Plate 3.4). There were two factors involving inoculated and non-inoculated controls as the first factor, and twelve rice varieties as the second factor.

3.2.5 Assessment of plant growth parameters, yield components and nematode population density

Starting from 10 days after inoculation (10 DAI), plant height and number of tillers were measured every 10 days to assess the damage caused by the nematodes. The experiment was terminated when the panicles of each variety were ripened and ready to harvest. Extraction of nematodes from soil and roots were similar as the method described in section 3.1.4. The nematode suspensions were collected and allowed settling for 1 hour. The suspension was adjusted to 100 ml concentration. 1 ml nematode suspension was taken by 5 ml syringe and one syringe was used for one sample. The juvenile and adult *H. oryzae* were counted under the microscope with 5 ml counting dish for 10 times and calculated for the average value.

Plant height (cm), tiller number (no.), fresh shoot weight (g), fresh root weight (g), dry shoot weight (g), filled grain percentage (%), 1000 grain weight (g) and final nematode population (no.) were recorded.

Yield loss percentage was calculated with the following equation (Saharan *et al*. 2005).

$$\text{Yield loss (\%)} = \frac{\text{Mean yield of non-inoculated plants-Mean yield of inoculated plants}}{\text{Mean yield of non-inoculated plants}} \times 100$$

The varietal response was categorized according to the scale adopted by de Man (1880).

3.2.6 Statistical analysis

The data were analyzed by using the statistix software (version 8.0). Means were compared by using least significant difference (LSD) test at 5% level.

Plate 3.4 Experimental layout for evaluation for the response of tested rice varieties to rice root nematode, *Hirschmanniella oryzae*

CHAPTER IV
RESULTS AND DISCUSSION

4.1 Population Density of *Hirschmanniella oryzae* in Nay Pyi Taw Union Territory

4.1.1 Population density of *Hirschmanniella oryzae* in different townships and summer rice varieties

In the present study, 98.89% out of 44 fields sampled were infested with the rice root nematode, *H. oryzae* (Plate 4.1). The highest frequency of occurrence (100%) was observed in Tatkon and Pyinmana Townships but the lowest (77.78%) occurred in Lewe Township. The number of *H. oryzae* found in the soil averaged 115 per 150 g soil. The lowest nematode population (40 per 150 g soil) was found in Kyarpin Village, Dekkhinathiri Township and Yonepin Village, Lewe Township and the highest (273 per 150 g soil) in Khayansatkone Village, Tatkon Township. The number of *H. oryzae* found in the roots was 7128 per 50 g roots in average. The lowest average root nematode population (3687 per 50 g roots) occurred in Kyarpin Village, Dekkhinathiri Township and the highest (12,913 per 50 g roots) in Nyaungdoneai Village, Tatkon Township. In Pyinmana region, 98.2% of fields sampled were infested by *H. oryzae* (Maung *et al*. 2010). The same author also found that the average nematode population in this region was 14 per 100 g soil and 208 per 20 g roots.

The highest prominence value (863) of soil was found in Khayansatkone Village, Tatkon Township and the lowest value (103) in Yonepin Village, Lewe Township.The highest prominence value (40,834) of root was found in Nyaungdoneai Village, Tatkon and the lowest (11,659) in Kyarpin Village, Dekkhinathiri Township (Table 4.1). Based on the prominence value (a combination of the frequency of occurrence and abundance) of *H. oryzae*, the highest population was found in Tatkon Township and the lowest in Lewe Township (Figure 4.1). In Tatkon Township, the rice-rice cropping sequence was the most common one and the farmers used the same rice variety for the monsoon and summer rice growing seasons. These factors as well as other condition such as a long history of rice cultivation might have contributed to the highest prominence value.

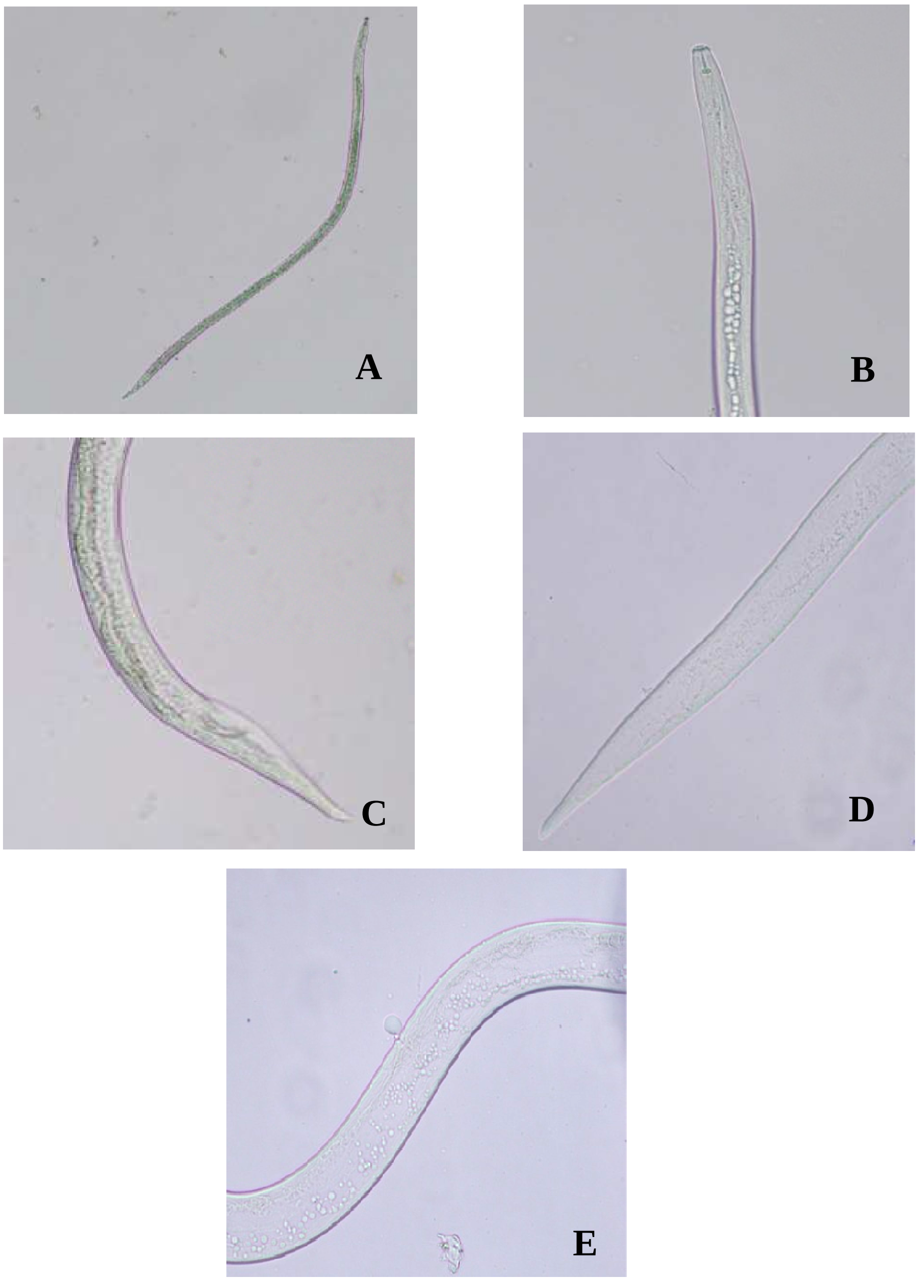


Plate 4.1 Rice root nematode, *Hirschmanniella oryzae*:

(A) Whole body (female) (10x)

(B) Head region (40x)

(C) Male tail (40x)

(D) Female tail (40x)

(E) Vulva (medium) (40x)

Table 4.1 Population density of *Hirschmanniella oryzae* in different townships of Nay Pyi Taw Union Territory

Township	Village	No. of fields (n)	Frequency of occurrence (%)		Population density				Prominence value (PV)	
					No. of *H. oryzae* per 150 g soil		No. of *H. oryzae* per 50 g roots			
			Soil	Root	Min-max	Mean±SE	Min-max	Mean±SE	Soil	Roots
Zabuthiri	Aungzabu	3	100	100	60-160	107±29	2560-8740	5873±1798	338	18572
	Taegyikone	3	100	100	20-180	80±50	3800-10420	6733±1948	253	21292
	Ahlyinlo	3	66.67	100	0-200	120±61	4600-9560	6707±1480	310	21209
Dekkhinathiri	Kyarpin	3	100	100	20-60	40±12	3380-4020	3687±185	126	11659
	Ywarma	3	66.67	100	0-260	120±76	4160-16320	8947±3741	310	28293
	Kyuntatpae	3	100	100	40-200	100±50	5120-6860	5900±510	316	18657
Pyinmana	Zephyupin	3	100	100	40-140	87±29	4620-6140	5493±453	275	17370
	Maezalikone	2	100	100	60-280	170±110	9060-10800	9930±870	538	31401
	Kan Oo	3	100	100	80-280	160±61	8500-12840	10687±1253	506	33795
Tatkon	Khayansatkone	3	100	100	40-600	273±168	2620-6000	4680±1043	863	14799
	Nyaungdoneai	3	100	100	20-220	100±61	10400-15120	12913±1371	316	40834
	Yaeaye	3	100	100	40-160	107±35	4580-13360	9093±2537	338	28755
Lewe	Padaukchaung	3	100	100	80-240	180±50	3700-8840	5673±1599	569	17940
	Yaekar	3	66.67	100	0-100	47±29	1940-11660	5733±3002	121	18129
	Yonepin	3	66.67	100	0-60	40±20	4120-5720	4867±465	103	15391
Total (range)	**15**	**44**			**0-600**		**1940-16320**			
Mean			**97.78**	**100**		**115 ± 16**		**7128±670**	**352**	**22540**

Frequency of occurrence: (Number of samples containing a species / Number of samples collected) ×100

PV: nematode population density × $\sqrt{\text{frequency of occurrence}/10}$

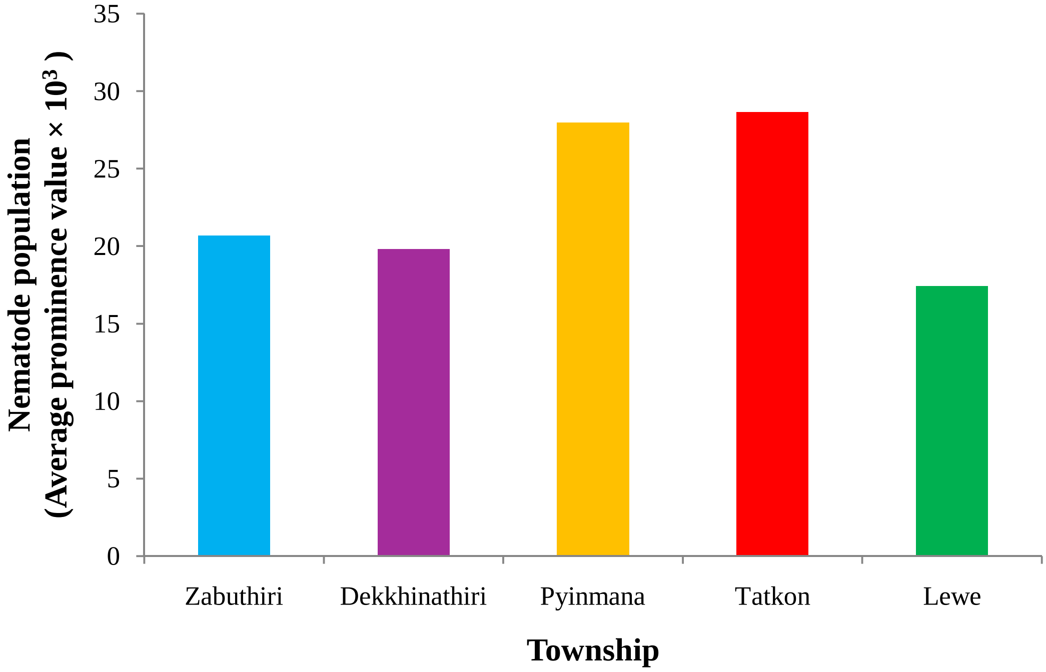


Figure 4.1 Nematode population in different townships of Nay Pyi Taw Union Territory

Bridge (1996) mentioned that the continuous growing of a highly susceptible host as a monocrop will greatly increase the population density and cause damage to the crop. Bridge *et al*. (2005) remarked that where there is a long history of rice cultivation, *H. oryzae* is likely to be widespread and abundant. The rice varieties namely Sinthukha and Manawthukha were the most common in Tatkon Township. In the present study, Sinthukha had the highest average population in both soil and root. This variety can be considered as good host for *H. oryzae*. The soil texture of the rice fields in Tatkon Township is clay soil, which may be one of the factors for the detection of the highest population density of *H. oryzae*. Fortuner (1976) noted that the soil texture could explain the occurrence of *H. oryzae*. He found that the development of *H. oryzae* was better in clay than in sandy soil in Senegal. Mathur and Prasad (1971) also described that the highest *H. oryzae* population was found in heavy clay soil in India. The fields sampled in Tatkon Township were mostly established with transplanted rice plants. Sato *et al*. (1970) described that there were more adult and juvenile population of *H. oryzae* in the roots of transplanted rice.

In the present study, rice-blackgram-rice cropping sequence was practiced in all rice fields surveyed in Lewe Township. Bridge *et al*. (2005) stated that due to the combination of dry soil and non-host dry season crops, the population density of *H. oryzae* is always low in this cropping system. Maung (2011) also found that rotation with non-host crops can reduce the population density of *H. oryzae* and result in better rice plant growth and yield. Among 5 summer rice varieties surveyed, the lowest soil population was observed in Yet-90 and the lowest root population in Shwethweyin. The rice varieties sampled in Lewe Township were Shwethweyin and Yet-90. Therefore, the rice variety might be one of the factors contributing to the lowest population occurred in this township. All rice fields sampled in this township were established with direct seeding method. Sato *et al*. (1970) revealed that direct seeding reduce initial infection of *Hirschmanniella* spp.

It was found that 96.58% out of 5 summer rice varieties were infested with the rice root nematode, *H. oryzae*. The frequency of occurrence of *H. oryzae* in different summer rice varieties ranged from 88.46% (Yet-90) to 100% (Sinthukha, Shwethweyin and Palethwe). The average number of *H. oryzae* in soil was 127 per 150 g soil. The highest average soil nematode population density (240 per 150 g soil) was observed in Sinthukha and the lowest (72 per 150 g soil) in Yet-90. The average number of *H. oryzae* in roots was 7240 per 50 g roots. The highest average root nematode population density (10,070 per 50 g roots) was observed in Sinthukha and the lowest (4260 per 50 g roots) in Shwethweyin. Nant (2016) found that *H. oryzae* population in roots was the highest in Shwewarsan and Sinthukha and the lowest in Manawbaykyar. The present study agreed with the findings of Myat *et al*. (2004) and Hlaing *et al*. (2005) who observed that Shwethweyin had the lowest number of *H. oryzae* in the roots. Maung (2011) also found that Shwethweyin had the lowest soil and the second-lowest root numbers of *H. oryzae*.

Among the different summer rice varieties, the highest prominence value (759) of *H. oryzae* in the rhizosphere soil was found in Sinthukha and the lowest (200) in Yet-90. In the roots, the highest prominence value (31,844) of *H. oryzae* was found in Sinthukha and the lowest (13,471) in Shwethweyin (Table 4.2). Among five rice varieties, the highest nematode population was found in Sinthukha and the lowest in Shwethweyin based on average prominence value in both soil and roots population (Figure 4.2).

4.1.2 Reaction of rice varieties to *Hirschmanniella oryzae*

It was observed that four summer rice varieties; Manawthukha, Sinthukha, Palethwe and Yet-90 were highly susceptible and Shwethweyin was susceptible to *H. oryzae*. Of these varieties, the highest nematode population (201 *H. oryzae* per g root) was observed in Sinthukha and the lowest population (85 *H. oryzae* per g root) in Shwethweyin. In this study, the average root population density of *H. oryzae* was over 100 *H. oryzae* per g root in four rice varieties except Shwethweyin. Park *et al.* (1970) rated the varieties, which had more than 100 nematodes per g root as susceptible to *H. oryzae.* In all varieties surveyed, the average root population density of *H. oryzae* exceeded 50 *H. oryzae* per g root (Table 4.3). In China, 15 *H. oryzae* per g root at the end of the maximum tillering stage was considered as an acceptable economic threshold level to control this nematode (Ying *et al*. 1996). Prasad *et al.* (1987) observed that at low levels of nematode infection (below 50 *H. oryzae* per g root) there was no appreciable reduction in chlorophyll, starch and proteins.

Table 4.2 Population density of *Hirschmanniella oryzae* in different summer rice varieties in Nay Pyi Taw Union Territory

Variety	No. of fields (n)	Frequency of occurrence (%)		Population density				Prominence value (PV)	
				No. of *H. oryzae* per 150 g soil		No. of *H. oryzae* per 50 g roots			
		Soil	Root	Min-max	Mean±SE	Min-max	Mean±SE	Soil	Roots
Manawthukha	9	88.89	100	0-200	84±21	4160-10420	6678±732	250	21118
Sinthukha	6	100	100	20-600	240±79	2620-16320	10070±2379	759	31844
Shwethweyin	7	100	100	40-240	126±32	2560-6140	4260±425	398	13471
Palethwe	9	100	100	20-280	111±33	4620-13360	9247±1047	351	29242
Yet-90	13	76.92	100	0-200	72±19	1940-11660	5943±761	200	18793
Total (range)	**44**			**0-600**		**1940-16320**			
Mean		**93.16**	**100**		**127±30**		**7240±1071**	**392**	**22894**

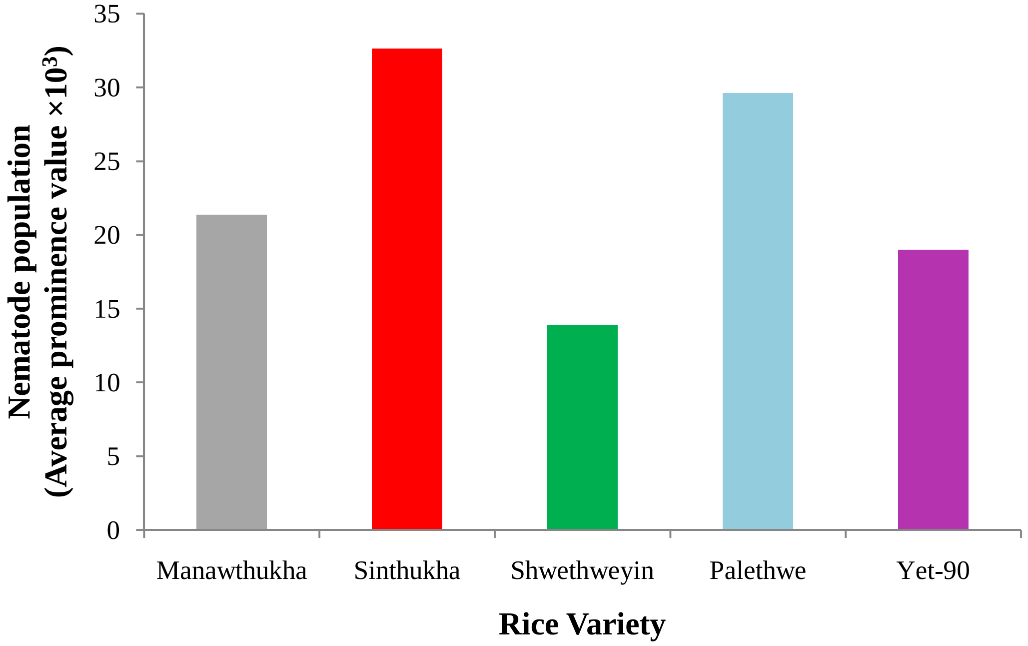


Figure 4.2 Nematode population in different summer rice varieties in Nay Pyi Taw Union Territory

Table 4.3 Reaction of rice varieties to *Hirschmanniella oryzae*

No.	Rice varieties	Nematode population per g root	Reaction[x]
1.	Manawthukha	134	Highly susceptible (HS)
2.	Sinthukha	201	Highly susceptible (HS)
3.	Shwethweyin	85	Susceptible (S)
4.	Palethwe	185	Highly susceptible (HS)
5.	Yet-90	119	Highly susceptible (HS)

[x] Based on nematode population per g root at maximum tillering stage is 0 = Immune (I), 1-10 = Highly resistant (HR), 11-50 = Moderately resistant (MR), 51-100 = Susceptible (S), > 100 = Highly susceptible (HS), (de Man 1880)

4.1.3 Population density of *Hirschmanniella oryzae* in different cropping sequences and sowing methods

The rice-rice cropping sequence was the most common one among the townships. Rice-blackgram-rice cropping sequence was practiced only in Lewe Township and Zephyupin Village in Pyinmana Township. In two different cropping sequences, the frequency of occurrence of *H. oryzae* observed in rice-rice cropping sequence (96.88%) was higher than in rice-blackgram-rice cropping one (91.67%). The highest average soil population density (124 per 150 g soil) was found in rice-rice cropping sequence and the lowest (88 per 150 g soil) in rice-blackgram-rice cropping one. Moreover, the highest average root population density (7673 per 50 g roots) was also observed in rice-rice cropping sequence and the lowest (5442 per 50 g roots) in rice-blackgram-rice cropping one. Based on these results, the average nematode population density in both soil and roots was higher in rice-rice cropping sequence than in rice-blackgram-rice cropping one. This might be due to blackgram is non-host for *H. oryzae*. *H. oryzae* cannot reproduce on the most common varieties of sesame, mungbean and blackgram cultivated in Myanmar. These crops are non hosts for *H. oryzae*. Moreover, rotation with these dry season crop can reduce the population density of *H. oryzae* and result in better rice plant growth and yield (Maung 2011). Rotation with non-host dry season crops such as cotton, cowpea, groundnut, millet, onion, pigeon pea, sorghum, soybean, sweet potato, tobacco and wheat can reduce the population density of *H. oryzae* in the field (Mathur and Prasad 1973; Babatola 1979; Gao *et al*. 1998). Prot and Rahman (1994) mentioned that crop rotation can significantly increase rice yield in *H. oryzae* infested fields.

In two different cropping sequences, the highest average prominence value (380) of soil was found in rice-rice cropping sequence while the lowest value (254) occurred in rice-blackgram-rice cropping one. In the roots, the highest prominence value (24,264) was found in rice-rice cropping sequence and the lowest value (17,209) in rice-blackgram-rice cropping one. The higher prominence value in both soil and roots was found in rice-rice cropping sequence than in rice-blackgram-rice cropping one (Table 4.4).

Table 4.4 Population density of *Hirschmanniella oryzae* in different cropping sequences

Cropping sequence	No. of fields (n)	Frequency of occurrence (%)		Population density				Prominence value (PV)	
				No. of *H. oryzae* per 150 g soil		No. of *H. oryzae* per 50 g roots			
		Soil	Root	Min-max	Mean±SE	Min-max	Mean±SE	Soil	Roots
Rice-Rice	32	93.75	100	0-600	124±21	2560-16320	7673±665	380	24264
Rice-Blackgram-Rice	12	83.33	100	0-240	88±22	1940-11660	5442±745	254	17209

It is well recognized that the population of plant-parasitic nematodes in a field can drastically increase when the same host plant is grown in the same field year after year. The build-up of this population can be prevented or restricted by crop rotation with non-host plants, less susceptible or resistant varieties of the same crop or a completely different crop (Bridge 1996; Maung 2011). In the microplot experiment, it was clearly observed that growing of rice in *H. oryzae* infested field can increase the population density of *H. oryzae* in the soil but the rate of increase in populaiton was different and depended upon the crop used in the cropping sequence (Maung 2011).

It was observed that the higher frequency of occurrence of *H. oryzae* occurred in transplanting (100%) than in direct seeding method (92.86%). The soil population density of *H. oryzae* in transplanting method averaged 131 per 150 g soil and 104 per 150 g soil in direct seeding. The average root nematode population density in transplanting and direct seeding was 8860 and 6038 per 50 g roots respectively. On the other hand, the average nematode population density per g root was 177 in transplanting and 121 in direct seeding. The injury level of *H. oryzae* to rice was 15 nematodes per g root at the end of maximum tillering stage and that could be applied as an economic threshold level (Ying *et al*. 1996). Park *et al.* (1970) also rated the varieties, which had more than 100 nematodes per g root as susceptible to *H. oryzae.* Therefore, management action should be taken to prevent an increasing nematode population in both sowing methods. On average, *H. oryzae* population density in both soil and roots was higher in transplanting method than in direct seeding. Nakazato *et al*. (1964) stated that the nematode population was higher in transplanted rice roots than in directly sown ones. Sato *et al*. (1970) also found that direct seeding reduced initial infection of *Hirschmanniella* spp.

The highest average prominence value (414) of soil was observed in transplanting method and the lowest value (304) in direct seeding. In the roots, the highest prominence value (28,018) occurred in transplanting and the lowest value (19,094) in direct seeding. There were more prominence value in both soil and roots in transplanting method than in direct seeding (Table 4.5). It could be suggested that the transplanted rice roots suffered damage when they were uprooted and they need more time to recover from transplanting shock. This condition can increase the probability of soilborne pathogen, *H. oryzae* invasion and also favour substantial damage from *H. oryzae*.

Table 4.5 Population density of *Hirschmanniella oryzae* in different sowing methods

Sowing method	No. of fields (n)	Frequency of occurrence (%)		Population density				Prominence value (PV)	
				No. of *H. oryzae* per 150 g soil		No. of *H. oryzae* per 50 g roots			
		Soil	Root	Min-max	Mean±SE	Min-max	Mean±SE	Soil	Roots
Direct Seeding	28	85.71	100	0-280	104±15	1940-12840	6038±540	304	19094
Transplanting	16	100	100	20-600	131±38	2620-16320	8860±1028	414	28018

4.2 Response of Tested Rice Varieties to Rice Root Nematode, *Hirschmanniella oryzae*

In the evaluation of host response of twelve different rice varieties, the plant height, tiller number, fresh shoot weight, fresh root weight, dry shoot weight, filled grain percent, 1000 grain weight and grain yield of the plants inoculated with 2000 *H. oryzae* per 3 kg soil were significantly reduced when compared with those of non-inoculated plants. This finding was similar to the observation of Edward *et al.* (1985) who stated that the initial population density of 1000 and 2000 *H. oryzae* per seedling affected the rice plant growth. A reduction in plant growth parameters such as plant height, tiller number, root length and fresh root weight, were observed in *H. oryzae* inoculated plants compared with non-inoculated plants (Buangsuwon *et al*. 1971; Mathur and Prasad 1973; Myat *et al*. 2004; Hlaing *et al*. 2005; Maung 2011). Gaur *et al.* 1993 stated that *Hirschmanniella* spp., migratory endoparasites of roots, causes poor root growth and stunting of plants. Infection of rice roots by *H. oryzae* usually results in stunting and a decrease in number of tillers and root weight (Mathur and Prasad 1972). Growth retardation, reduced survival and decreased tiller emergence with discolored older leaves, delayed flowering and late maturation occurred in *H. oryzae* heavily infected rice plants (Taylor 1969).

4.2.1 Plant growth parameters

4.2.1.1 Plant height and tiller number

According to the results, it was observed that significant reduction in plant growth parameters (Plate 4.2) such as plant height and tiller number was observed in *H. oryzae* inoculated plants compared with non-inoculated ones. However, there was no interaction between inoculation, non-inoculation (factor A) and rice varieties (factor B) in plant height and tiller number (Table 4.6). Therefore, it can be determined that there was no effect *H. oryzae* on these plant growth parameters. There was no significant difference in plant height between inoculated and non-inoculated plants of Thukhamwe, Yet-90, Pyitawyin, Shweasean, Shwethweyin and Sinthukha whereas those of inoculated plants of Pawsanmwe, Salt tolerant Sinthwelatt, Myaungmyamay, Pyimyanmarsein, Submergence-tolerant 1 and Manawthukha were significantly lower than non-inoculated ones (Figure 4.3).

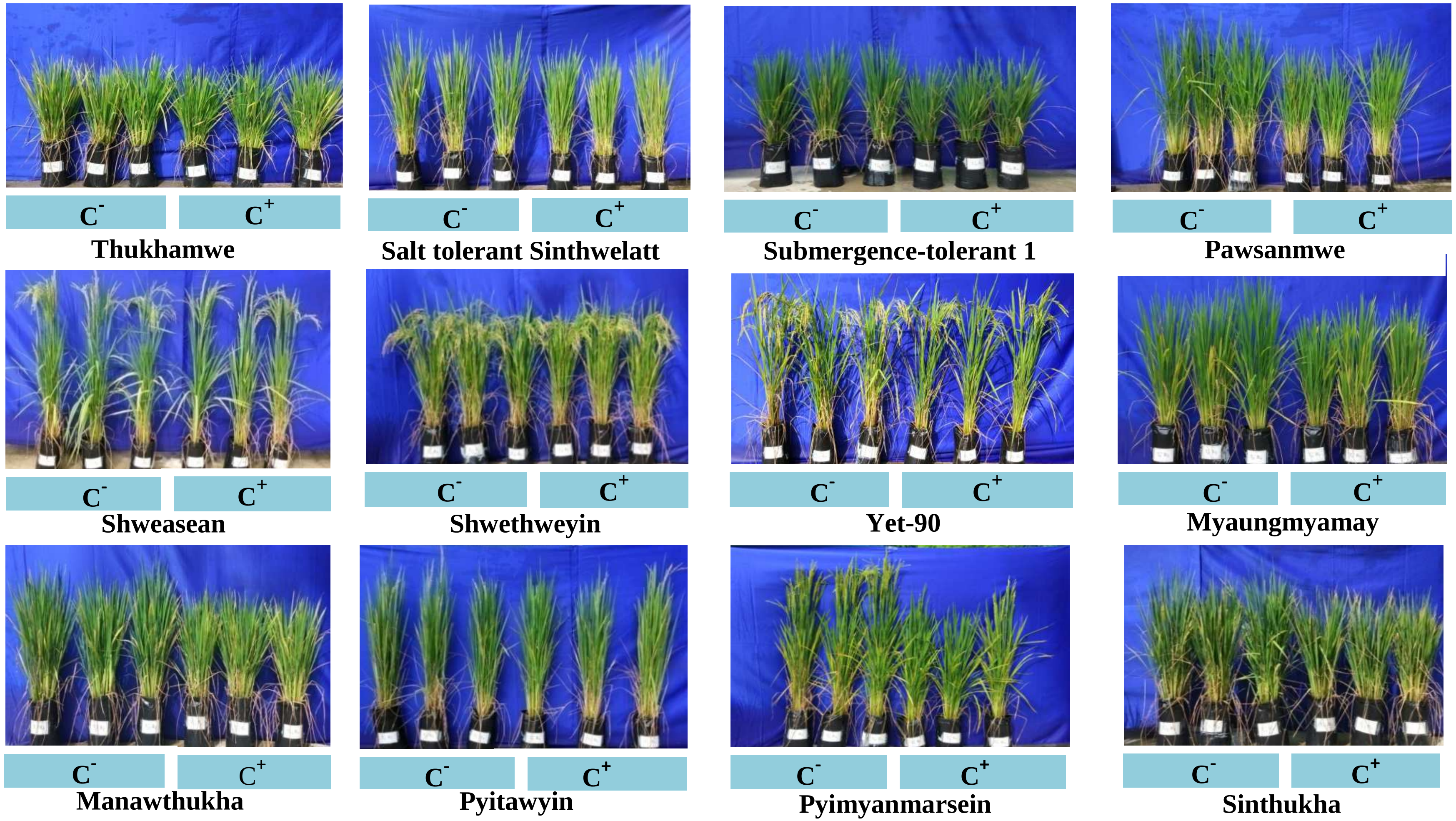


Plate 4.2 Plant growth of twelve rice varieties at 60 days after inoculation (C^- = non-inoculated, C^+ = inoculated with *H. oryzae*)

Table 4.6 Effect of *Hirschmanniella oryzae* on plant height and tiller number of twelve rice varieties

Treatment	**Plant height (cm)**[x]	**Tiller (no.)** [x]
Factor (A)		
Inoculation	54.00 b[y]	23.00 b
Non-inoculation	56.83 a	26.00 a
LSD $_{0.05}$	**0.84**	**1.17**
Factor (B)		
Thukhamwe	67.87 b	27.00 ab
Pawsanmwe	98.19 a	20.00 cd
Yet-90	42.72 f	15.00 e
Pyitawyin	43.96 f	21.00 c
Salt tolerant Sinthwelatt	66.28 bc	27.00 ab
Shweasean	47.61 e	17.00 de
Myaungmyamay	65.67 c	28.00 ab
Pyimyanmarsein	43.72 f	25.00 b
Submergence-tolerant 1	51.46 d	27.00 ab
Shwethweyin	37.75 g	29.00 a
Manawthukha	48.56 e	29.00 a
Sinthukha	51.21 d	28.00 ab
LSD $_{0.05}$	**2.07**	**2.87**
Pr>F		
A	<0.0001	<0.0001
B	<0.0001	<0.0001
A×B	0.33	0.40
CV%	**3.74**	**11.82**

[x] Means of four replications

[y] Means followed by the same letter in the same column were not significantly different at 5% level

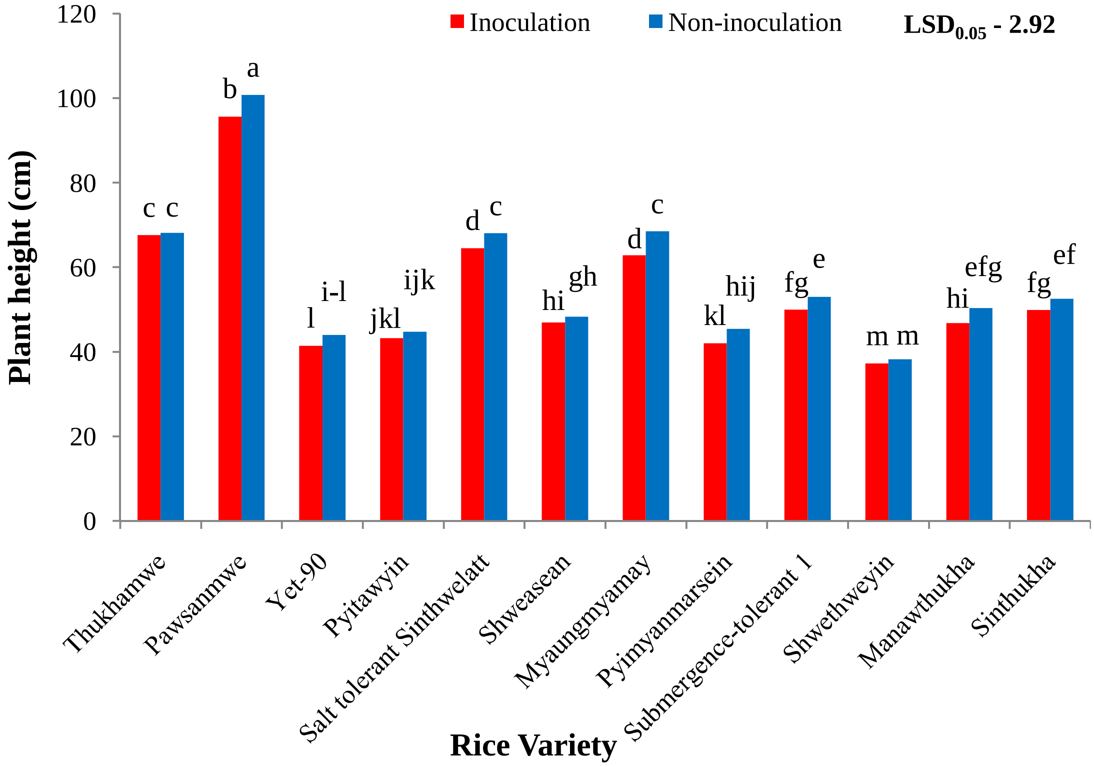


Figure 4.3 Effect of *Hirschmanniella oryzae* on plant height of twelve rice varieties

Maung (2011) also found that inoculation with 2000 *H. oryzae* per plant reduced the plant height of Manawthukha and Sinthwelatt with 9.2% and 8.1% respectively. Jonathan and Velayutham (1987) found that inoculation with 1 and 10 *H. oryzae* per g soil reduced the plant height of IR 20 at harvest with 11% and 13% respectively. In tiller number, no significant differences were observed between inoculated and non-inoculated plants of Thukhamwe, Pawsanmwe, Pyitawyin, Shweasean and Shwethweyin while inoculated plants of Yet-90, Salt tolerant Sinthwelatt, Myaungmyamay, Pyimyanmarsein, Submergence-tolerant 1, Manawthukha and Sinthukha were significantly lower than non-inoculated ones (Figure 4.4). Mathur and Prasad (1972) indicated that inoculation of rice plants with *H. oryzae* could reduce the number of tillers. Many authors had also pointed out that *H. oryzae* caused the reduction in tiller number of rice (Mathur and Prasad 1972; Babatola and Bridge 1979; Venkitesan and Charles 1979). The root nematodes (*Hirschmanniella* spp.) were found to be responsible for reduction of tillers and stunting of plants (Edward *et al*. 1985).

4.2.1.2 Fresh shoot weight, dry shoot weight and fresh root weight

Fresh shoot weight, dry shoot weight and fresh root weight of inoculated plants were also significantly lower than those of non-inoculated plants. Interaction effect was observed between inoculation, non-inoculation (factor A) and rice varieties (factor B) in dry shoot weight whereas there was no interaction between inoculation, non-inoculation (factor A) and rice varieties (factor B) on fresh shoot and root weight (Table 4.7). The result showed that there was infection effect of *H. oryzae* on dry shoot weight although no effect of *H. oryzae* was found on fresh shoot and root weight. In this study, fresh shoot weight of inoculated plants of Thukhamwe, Pyitawyin, Salt tolerant Sinthwelatt, Shweasean, Pyimyanmarsein and Shwethweyin was not significantly different with those of non-inoculated plants. However, fresh shoot weight of inoculated plants of Pawsanmwe, Yet-90, Myaungmyamay, Submergence-tolerant 1, Manawthukha and Sinthukha was significantly lower than those of non-inoculated plants (Figure 4.5). Siddiqi (1973) observed that *H. oryzae* retarded the growth of rice causing delayed tillering, fewer shoots, discoloration of older leaves and root-rot. Dry shoot weight of the inoculated plants did not differ significantly with those of non-inoculated plants of Thukhamwe, Pyitawyin, Shweasean, Pyimyanmarsein, Submergence-tolerant 1 and Shwethweyin whereas those of inoculated plants of Pawsanmwe, Yet-90, Salt tolerant Sinthwelatt, Myaungmyamay, Manawthukha and Sinthukha were significantly lower than those of non-inoculated ones (Figure 4.6).

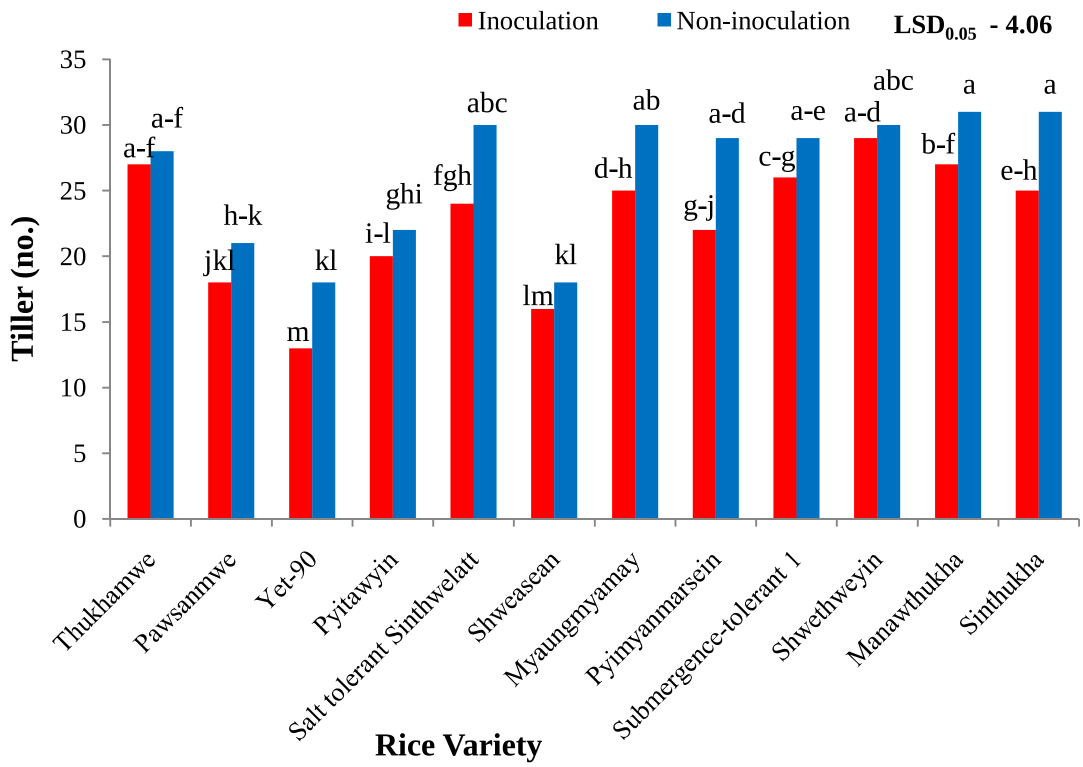


Figure 4.4 Effect of *Hirschmanniella oryzae* on tiller number of twelve rice varieties

Table 4.7 Effect of *Hirschmanniella oryzae* on fresh shoot weight, dry shoot weight and fresh root weight of twelve rice varieties

Treatment	**Fresh shoot weight (g)** [x]	**Dry shoot weight (g)** [x]	**Fresh root weight (g)** [x]
Factor (A)			
Inoculation	118.52 b [y]	44.93 b	93.65 b
Non-inoculation	132.35 a	51.19 a	100.37 a
LSD $_{0.05}$	**3.65**	**1.70**	**3.57**
Factor (B)			
Thukhamwe	130.47 cd	57.53 b	120.97 c
Pawsanmwe	142.52 ab	64.49 a	130.99 ab
Yet-90	102.70 e	42.76 d	62.95 f
Pyitawyin	146.24 a	42.30 d	82.15 e
Salt tolerant Sinthwelatt	130.51 cd	51.68 c	139.56 a
Shweasean	126.83 cd	40.92 d	71.55 f
Myaungmyamay	135.72 bc	54.22 bc	138.52 a
Pyimyanmarsein	131.24 cd	43.37 d	43.01 g
Submergence-tolerant 1	106.84 e	41.12 d	85.17 e
Shwethweyin	104.93 e	44.40 d	65.90 f
Manawthukha	123.63 d	43.71 d	126.57 bc
Sinthukha	123.62 d	50.17 c	96.81 d
LSD $_{0.05}$	**8.95**	**4.21**	**8.75**
Pr>F			
A	<0.0001	<0.0001	<0.0004
B	<0.0001	<0.0001	<0.0001
A×B	0.31	0.05	0.80
CV%	**7.15**	**8.79**	**9.04**

[x] Means of four replications

[y] Means followed by the same letter in the same column were not significantly different at 5% level

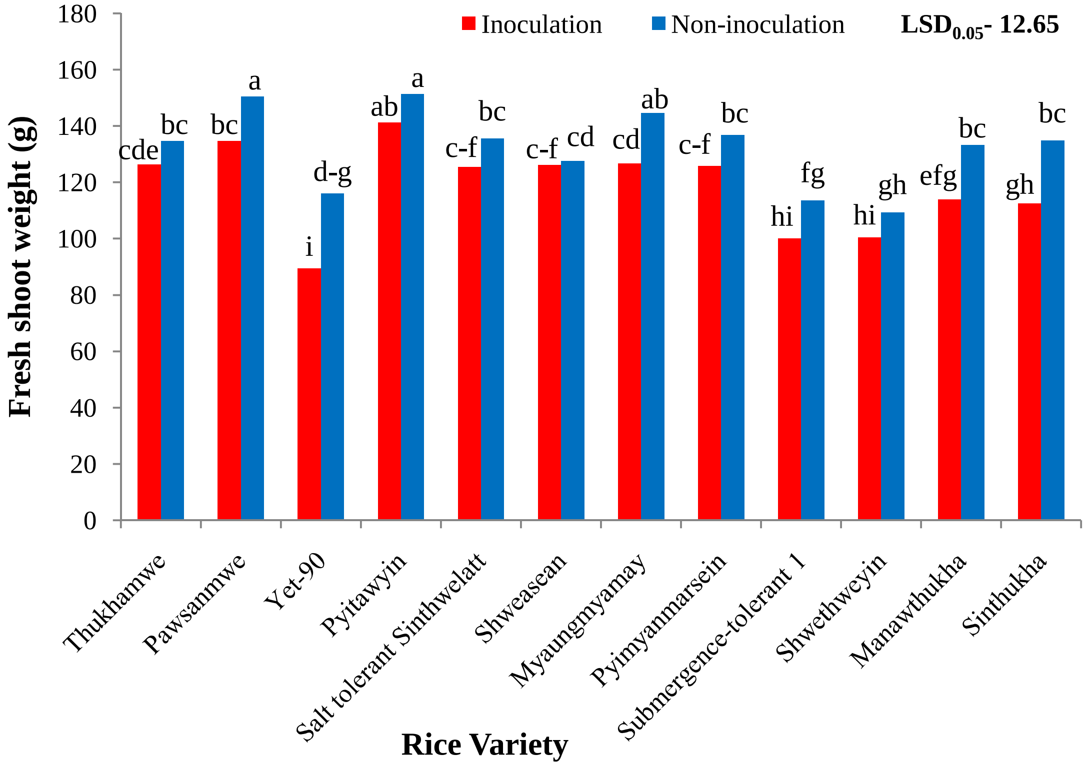


Figure 4.5 Effect of *Hirschmanniella oryzae* on fresh shoot weight of twelve rice varieties

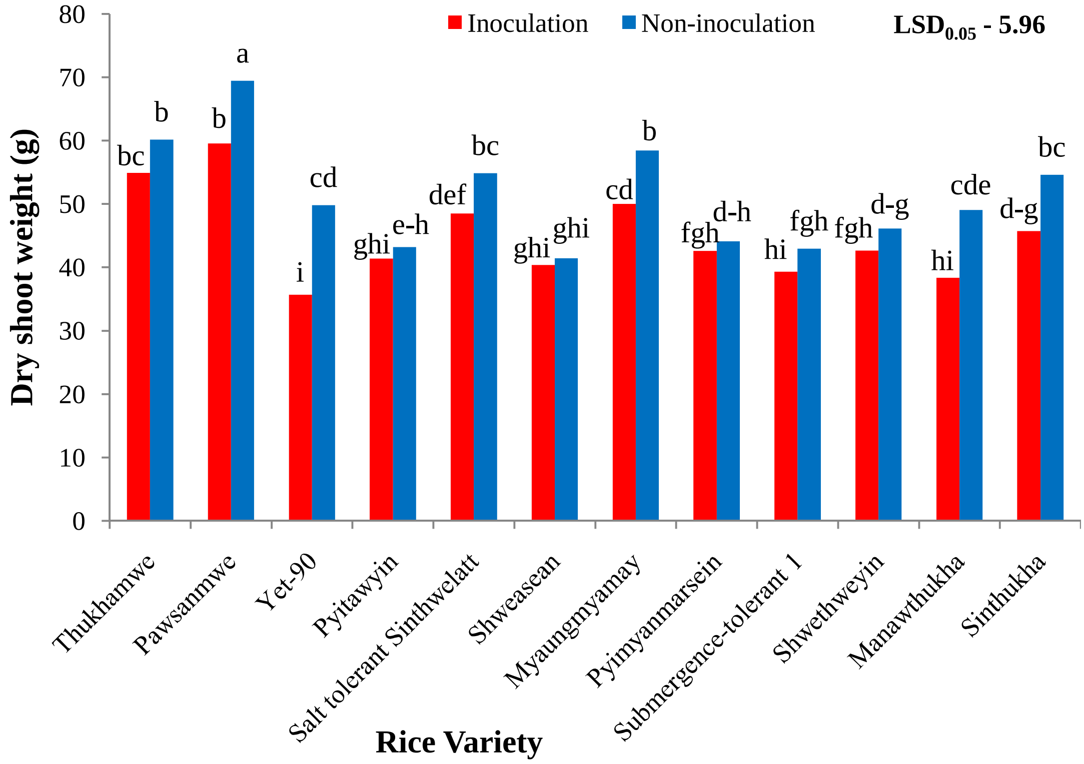


Figure 4.6 Effect of *Hirschmanniella oryzae* on dry shoot weight of twelve rice varieties

There was no significant difference in fresh root weight (Plate 4.3) between inoculated and non-inoculated plants of Thukhamwe, Yet-90, Pyitawyin, Salt tolerant Sinthwelatt, Myaungmyamay, Submergence-tolerant 1, Shwethweyin, Manawthukha and Sinthukha while those of inoculated plants of Pawsanmwe, Shweasean and Pyimyanmarsein were significantly lower than those of non inoculated ones (Figure 4.7). Thorne (1961) and Edward *et al*. (1985) noticed that the injury caused by *H. oryzae* is less evident due to the rapid regeneration of new roots under favourable environmental conditions. They also stated that the root system of *H. oryzae*-infected rice plants is poor. Mathur and Prasad (1972) and Babatola (1979) showed that rice plants inoculated with *H. oryzae* may have lower fresh root weights. Maung (2011) observed that 38.6% reduction in root weight in 2000 *H. oryzae* inoculated plant.

4.2.2 Yield components and grain yield

In the present study, 1000 grain weight, filled grain percent and grain yield of inoculated plants were significantly lower than those of non-inoculated ones. There was no interaction between inoculation, non-inoculation (factor A) and rice varieties (factor B) in 1000 grain weight while highly significant interaction effect was observed between inoculation, non-inoculation (factor A) and rice varieties (factor B) in filled grain percent and grain yield (Table 4.8). The result indicated that no effect of *H. oryzae* was observed on 1000 grain weight. This result was similar with the finding of Maung (2011) who observed that no effect of inoculation with 2000 *H. oryzae* on 1000 grain weight in Immayebaw, Manawthukha and Sinthwelatt at harvest. In the present study, it was observed that inoculation with 2000 *H. oryzae* can significantly reduce filled grain percent and grain yield. No significant difference in 1000 grain weight was observed between inoculated and non-inoculated plants of Thukhamwe, Pyitawyin, Salt tolerant Sinthwelatt, Shweasean, Submergence-tolerant 1 and Shwethweyin whereas those of inoculated plants of Pawsanmwe, Yet-90, Myaungmyamay, Pyimyanmarsein, Manawthukha and Sinthukha were significantly lower than those of non-inoculated ones (Figure 4.8). Significant reductions in filled grain percent of inoculated plants compared with non-inoculated ones were observed in Pawsanmwe, Yet-90, Pyitawyin, Salt tolerant Sinthwelatt, Myaungmyamay, Pyimyanmarsein, Manawthukha and Sinthukha.

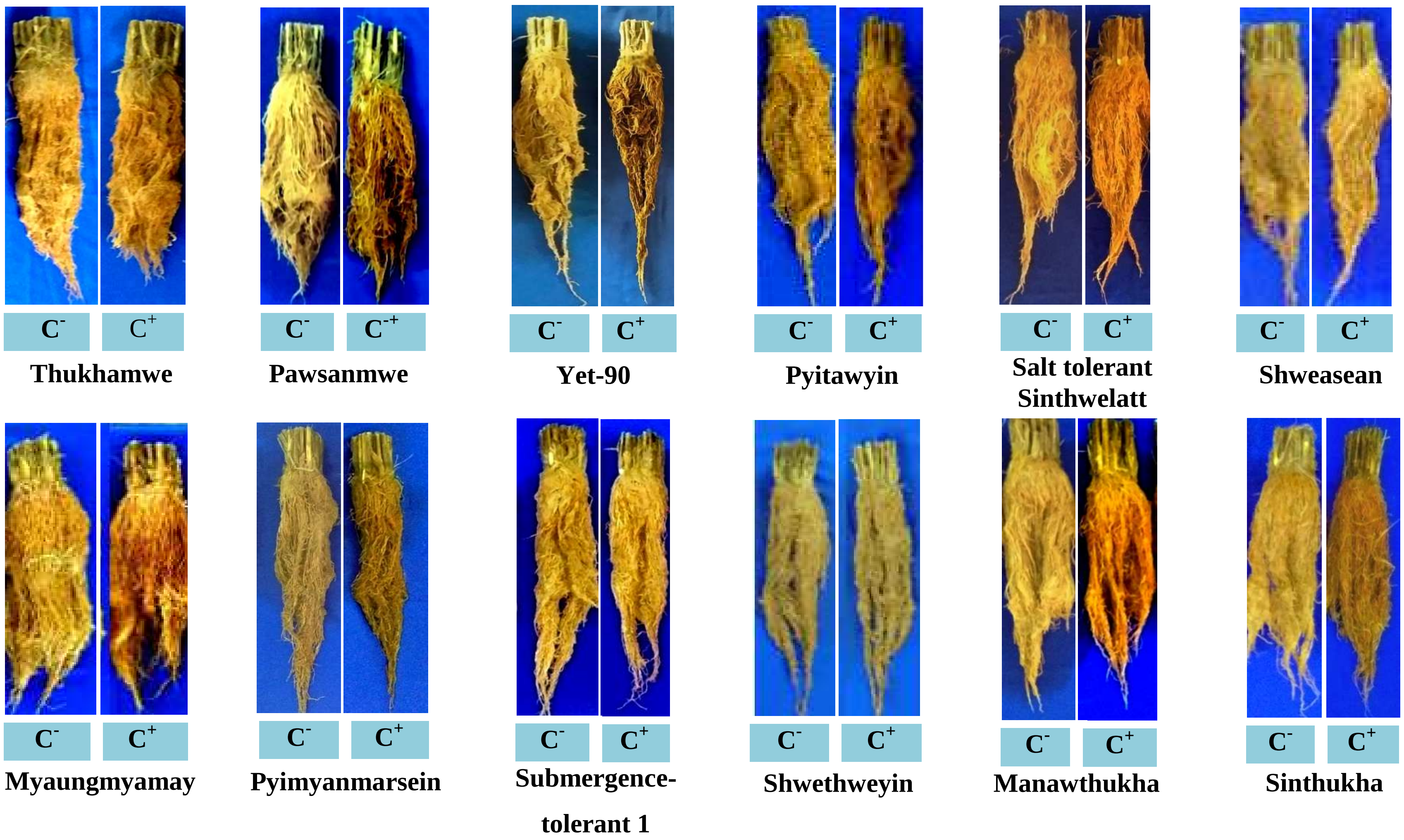


Plate 4.3 Root symptoms of twelve rice varieties (C^- = non-inoculated, C^+ = inoculated with *H. oryzae*)

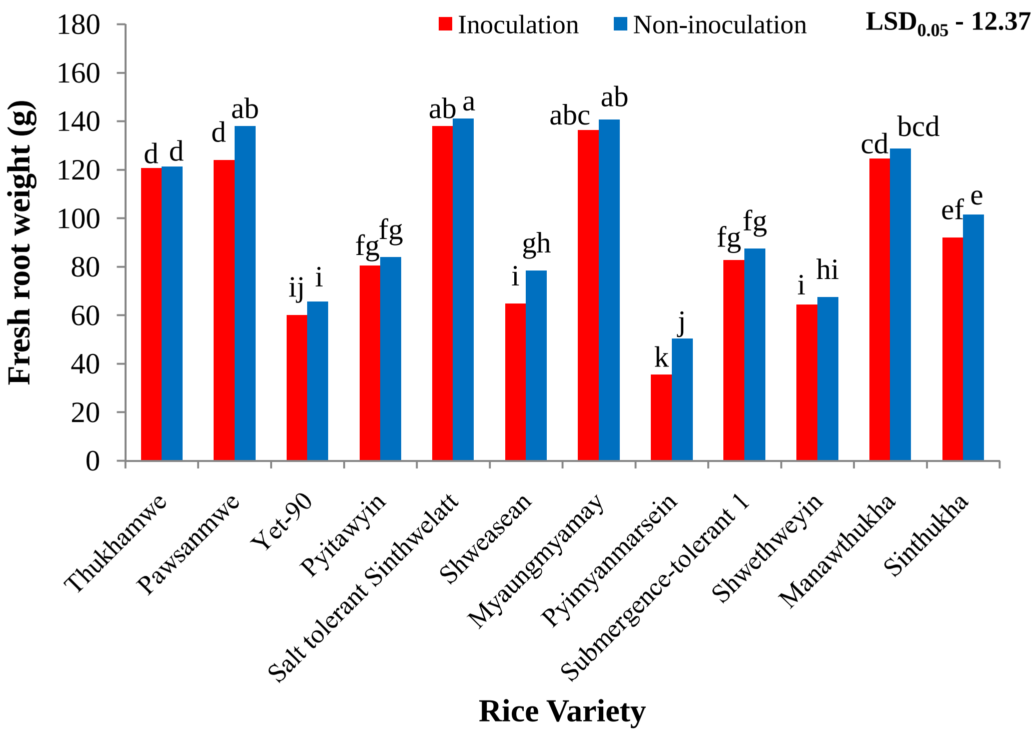


Figure 4.7 Effect of *Hirschmanniella oryzae* on fresh root weight of twelve rice varieties

Table 4.8 Effect of *Hirschmanniella oryzae* on 1000 grain weight, filled grain percent and grain yield of twelve rice varieties

Treatment	1000 grain weight (g) [x]	Filled grain percent (%)[x]	Grain yield per plant (g) [x]
Factor (A)			
Inoculation	20.83 b [y]	72.03 b	20.78 b
Non-inoculation	22.19 a	79.60 a	24.08 a
LSD $_{0.05}$	**0.46**	**1.37**	**0.74**
Factor (B)			
Thukhamwe	17.20 g	74.35 d	18.37 d
Pawsanmwe	23.77 c	70.80 e	16.87 de
Yet-90	30.96 a	88.88 b	27.79 b
Pyitawyin	26.78 b	69.16 e	21.80 c
Salt tolerant Sinthwelatt	22.03 e	59.80 g	17.72 d
Shweasean	23.74 c	87.18 b	31.89 a
Myaungmyamay	23.35 de	65.73 f	17.16 de
Pyimyanmarsein	23.28 cd	83.74 c	27.52 b
Submergence-tolerant 1	15.88 h	71.90 de	20.56 c
Shwethweyin	18.76 f	92.68 a	33.63 a
Manawthukha	16.31 gh	80.81 c	20.21 c
Sinthukha	17.04 g	64.80 f	15.67 e
LSD $_{0.05}$	**1.14**	**3.36**	**1.81**
Pr>F			
A	<0.0001	<0.0001	<0.0001
B	<0.0001	<0.0001	<0.0001
A×B	0.23	0.002	0.004
CV%	**5.29**	**4.44**	**8.10**

[x] Means of four replications

[y] Means followed by the same letter in the same column were not significantly different at 5% level

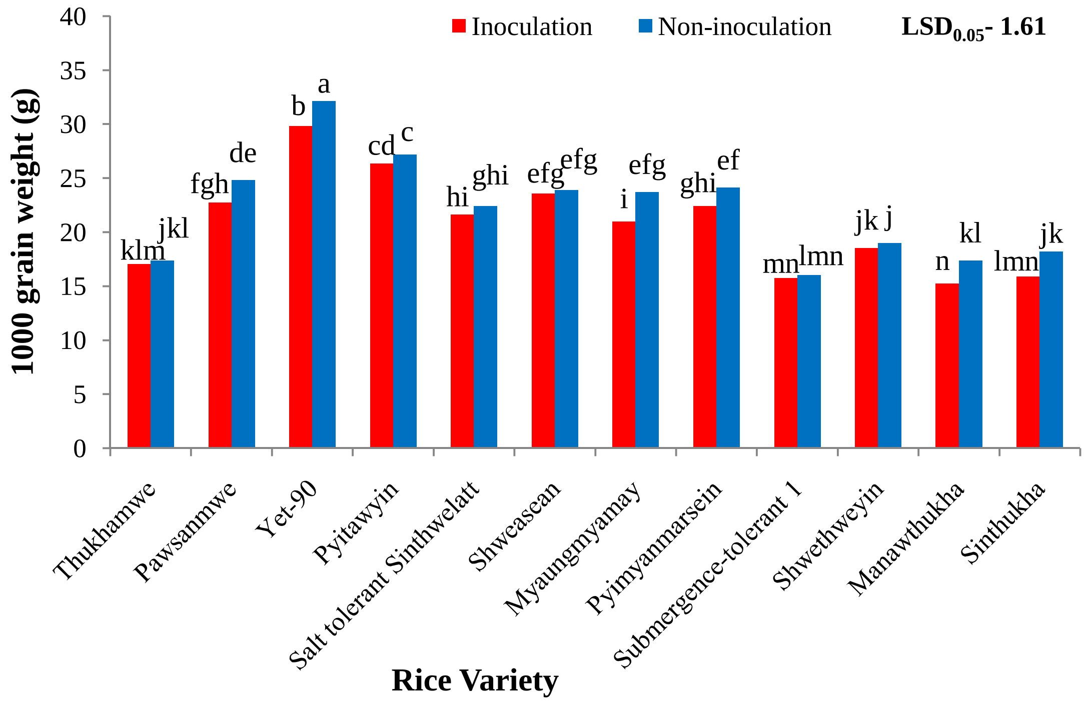


Figure 4.8 **Effect of *Hirschmanniella oryzae* on 1000 grain weight of twelve rice varieties**

However, there were no significant differences in filled grain percent of Thukhamwe, Shweasean, Submergence-tolerant 1 and Shwethweyin (Figure 4.9). In grain yield, significant reductions of inoculated plants were found in Pawsanmwe, Yet-90, Myaungmyamay, Pyimyanmarsein, Manawthukha and Sinthukha whereas grain yields of the other varieties, Thukhamwe, Salt tolerant Sinthwelatt, Shweasean and Shwethweyin were not significantly different between inoculated and non-inoculated plants (Figure 4.10). Henje *et al.* (1994) noted that losses in grain yield were different among different rice varieties even when the same number of *H. oryzae* was inoculated.

4.2.3 Yield loss percent

Yield loss percent of rice varieties inoculated with *H. oryzae* ranged from 1.45 to 12.68% in 1000 grain weight, 1.76 to 17.47% in filled grain percent and 2.42 to 26.44% in grain yield. In 1000 grain weight, the maximum yield loss percent was observed in Sinthukha (12.68%) followed by Manawthukha (12.29%) whereas the minimum yield loss percent was observed in Shweasean (1.45%) followed by Submergence-tolerant 1 (1.99%), Thukhamwe (2.10%) and Shwethweyin (2.28%). Studies with different rice varieties also showed that *H. oryzae* can cause varying degrees of yield loss (in terms of number of panicles and grain weight) depending on the nematode population density (Yamsonrat 1967; Mathur and Prasad 1972; Jonathan and Velayutham 1987; Babatola and Bridge 1979; Venkitesan *et al*. 1979; Jairaipuri and Baqri 1991; Prot *et al*. 1992; Henje *et al*. 1994). The maximum yield loss percent of (17.47%) in filled grain percent was found in Sinthukha followed by Manawthukha (16.46%) while the minimum yield loss percent (1.76%) in Shweasean followed by Shwethweyin (3.53%) and Thukhamwe (5.22%). The maximum yield loss percent in grain yield was observed in Manawthukha (26.68%) which was not significantly different from Sinthukha (26.44%) and Yet-90 (24.96%). However, the minimum yield loss percent was observed in Thukhamwe (2.42%) which was not statistical different as compared to Submergence-tolerant 1 (5.32%), Shwethweyin (5.37%) and Shweasean (5.52%) (Table 4.9). Population of 100 *H. oryzae* per plant reduced grain yield by 35% (Mathur and Prasad 1972). When the variety IR 8 was inoculated with 1000 and 5000 *H. oryzae*, a 31 to 37% reduction in grain yield was observed (Babatola and Bridge 1979). Inoculation with 1 and 10 *H. oryzae* per g soil caused 27 to 39.4% yield loss (Jonathan and Velayutham 1987). The reduction in yield parameters in susceptible varieties are attributable to root injury due to penetration and feeding by the nematodes, leading to impairement of the efficiency of root systems to absorb water.

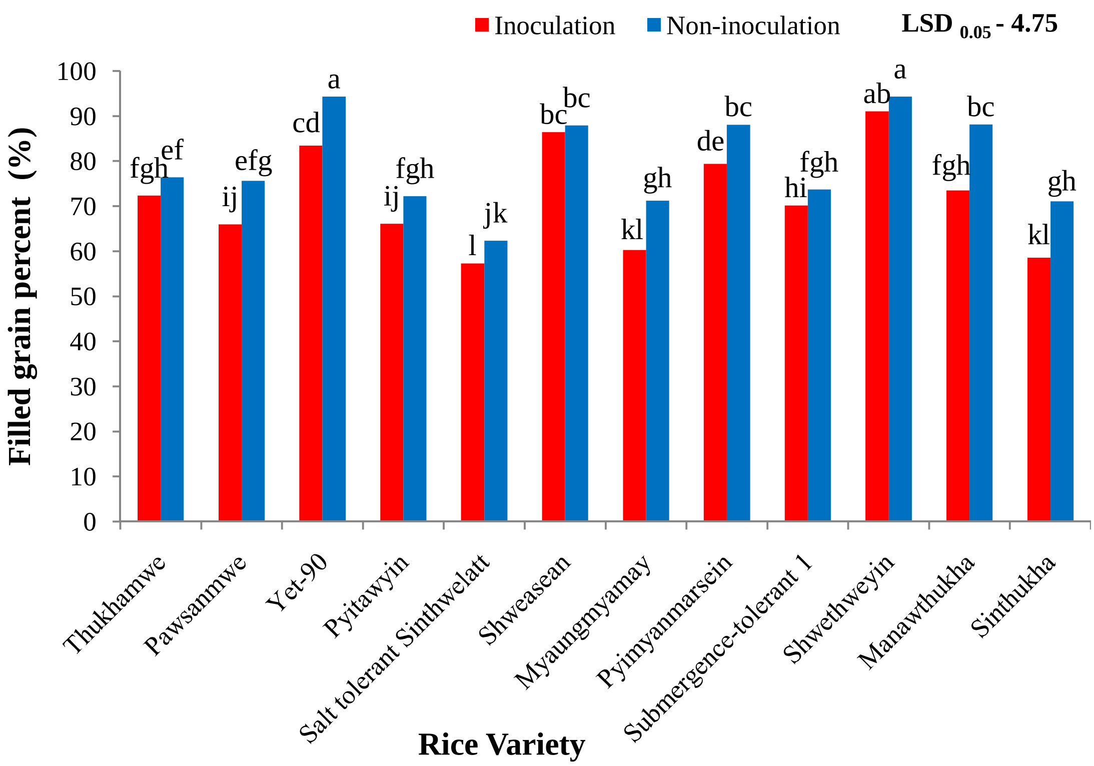


Figure 4.9 **Effect of *Hirschmanniella oryzae* on filled grain percent of twelve rice varieties**

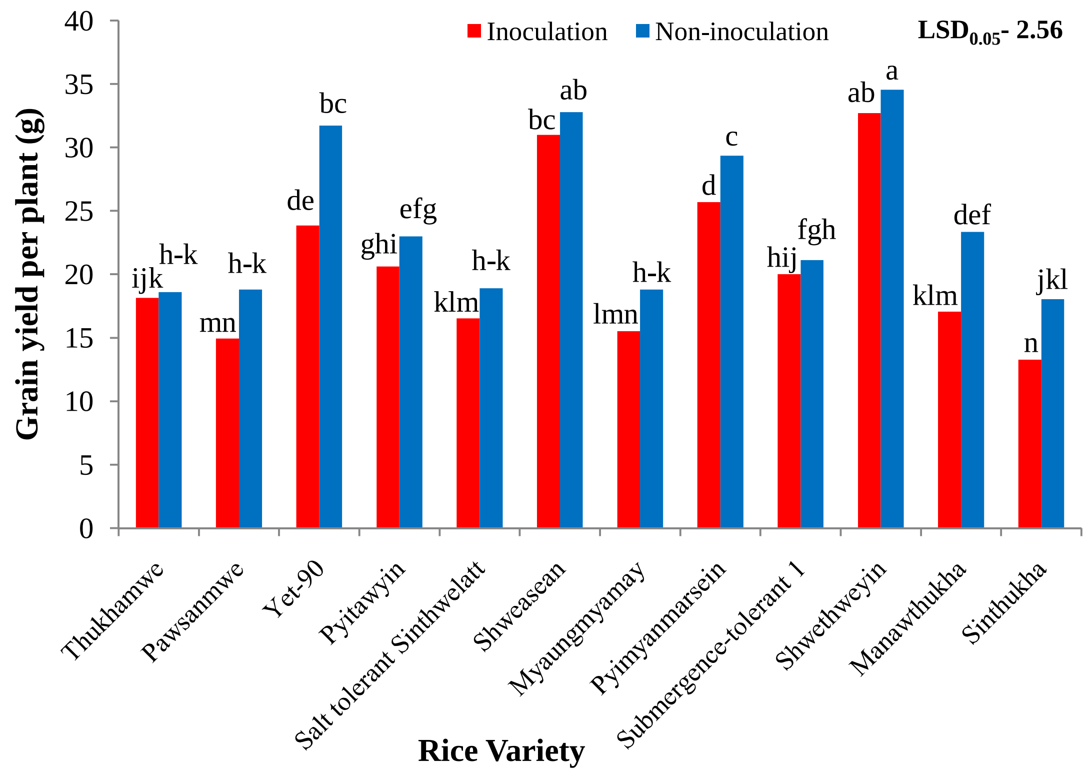


Figure 4.10 **Effect of *Hirschmanniella oryzae* on grain yield of twelve rice varieties**

Table 4.9 Effect of *Hirschmanniella oryzae* on reduction in yield components of twelve rice varieties

No.	Variety	Yield loss percent (%)[x]		
		1000 grain weight	**Filled grain percent**	**Grain yield per plant**
1	Thukhamwe	2.10 de[y]	5.22 efg	2.42 e
2	Pawsanmwe	8.35 abc	12.86 abcd	20.55 ab
3	Yet-90	7.21 abcd	11.50 bcd	24.96 a
4	Pyitawyin	3.00 cde	8.39 def	10.46 cd
5	Salt tolerant Sinthwelatt	3.23 cde	8.06 def	12.51 cd
6	Shweasean	1.45 e	1.76 g	5.52 de
7	Myaungmyamay	11.65 ab	15.48 abc	16.84 bc
8	Pyimyanmarsein	6.91 bcde	9.86 cde	12.47 cd
9	Submergence-tolerant 1	1.99 de	4.86 efg	5.32 de
10	Shwethweyin	2.28 de	3.53 fg	5.37 de
11	Manawthukha	12.29 ab	16.46 ab	26.68 a
12	Sinthukha	12.68 a	17.47 a	26.44 a
	CV %	**65.33**	**41.58**	**37.50**
	Pr ≥ F	**0.0002**	**0.0001**	**0.0001**
	$LSD_{0.05}$	**5.71**	**5.74**	**7.60**

[x] Means of four replications

[y] Means followed by the same letter in the same column were not significantly different at 5% level

Nematode feeding extensively disrupts xylem tissues and greatly retards absorption and upward movement of water and nutrients. The infection also greatly reduces permeability of roots to water (Wyss 2002; Di Vito *et al*. 2004). Due to the inadequate supply of water, nutrients, photosynthates and energy, growth and developments of leaf tissue and its constituents, especially chlorophyll pigments, are adversely affected (Khan and Khan 1997). The poor growth of foliage subsequently leads to decreased production (Hussain *et al*. 2016; Kayani *et al.* 2017).

4.2.4 Host response of twelve rice varieties to rice root nematode, *Hirschmanniella oryzae* at harvest

In the present study, the lowest nematode population per g root was observed in the inoculated plants of Thukhamwe (11 nematodes) followed by Salt tolerant Sinthwelatt (14 nematodes), Submergence-tolerant 1 (17 nematodes), Shweasean (19 nematodes), Shwethweyin (20 nematodes) and Pyitawyin (20 nematodes). In contrast, the nematode population per g root was the highest in Sinthukha (131 nematodes) and Yet-90 (117 nematodes) followed by Manawthukha (72 nematodes). Shwethweyin supported the lowest population density under screenhouse conditions and was rated as a resistant variety while Manawthukha and Immayebaw were rated as susceptible varieties (Than 2003; Myat *et al*. 2004; Hlaing *et al*. 2005). The lowest soil and second-lowest root population density of *H. oryzae* was observed from Shwethweyin (Maung *et al*. 2010). Under field conditions, the lowest nematode population density was recovered from Shwethweyin (Maung 2011).

According to de Man (1880) rating scale, eight varieties; Thukhamwe, Pyitawyin, Salt tolerant Sinthwelatt, Shweasean, Myaungmyamay, Pyimyanmarsein, Submergence-tolerant 1 and Shwethweyin were rated as moderately resistant to *H. oryzae*. Two varieties; Pawsanmwe and Manawthukha were susceptible and another two varieties, Yet-90 and Sinthukha were highly susceptible to *H. oryzae* (Table 4.10). Coyne *et al.* (1999) reported that *H. oryzae* population density was consistently influenced by cultivars. However, Ramakrishnan *et al*. (1984) and Dash *et al*. (2008) mentioned that the host response to *H. oryzae* infection of the same rice variety can be different under different environmental conditions.

Table 4.10 Host response of twelve rice varieties to rice root nematode, *Hirschmanniella oryzae*

No.	Rice varieties	Nematode population per g root [x]	Host response [y]
1	Thukhamwe	11 e [z]	Moderately resistant (MR)
2	Pawsanmwe	61 bc	Susceptible (S)
3	Yet-90	117 a	Highly susceptible (HS)
4	Pyitawyin	20 e	Moderately resistant (MR)
5	Salt tolerant Sinthwelatt	14 e	Moderately resistant (MR)
6	Shweasean	19 e	Moderately resistant (MR)
7	Myaungmyamay	37 d	Moderately resistant (MR)
8	Pyimyanmarsein	50 cd	Moderately resistant (MR)
9	Submergence-tolerant 1	17 e	Moderately resistant (MR)
10	Shwethweyin	20 e	Moderately resistant (MR)
11	Manawthukha	72 b	Susceptible (S)
12	Sinthukha	131 a	Highly susceptible (HS)
	CV %	**21.78**	
	Pr ≥ F	**0.0001**	
	$LSD_{0.05}$	**14.75**	

[x] Means of four replications

[y] Based on de Man (1880) rating scale, nematode population per g root is 0 = Immune (I), 1-10 = Highly resistant (HR), 11-50 = Moderately resistant (MR), 51-100 = Susceptible (S), > 100 = Highly susceptible (HS)

[z] Means followed by the same letter in the same column were not significantly different at 5% level

CHAPTER V
CONCLUSION

It was observed that 98.89% of fields sampled were infested with the rice root nematode, *H. oryzae*. Based on the prominence value (a combination of the frequency of occurrence and abundance) of *H. oryzae*, the highest nematode population was observed in Tatkon Township and the lowest in Lewe Township. The highest population of *H. oryzae* from soil and root were observed in Sinthukha and the lowest in Shwethweyin. All summer rice varieties surveyed were observed to be either susceptible or highly susceptible to *H. oryzae*. In two different cropping sequences, rice-blackgram-rice cropping sequence had the lower nematode population than that of rice-rice cropping one. Moreover, the lower nematode population was also found in direct seeding than in transplanting method.

Among the twelve rice varieties, Yet-90 and Sinthukha were rated as highly susceptible and Pawsanmwe and Manawthukha as susceptible to *H. oryzae*. The other eight varieties; Thukhamwe, Pyitawyin, Salt tolerant Sinthwelatt, Shweasean, Myaungmyamay, Pyimyanmarsein, Submergence-tolerant 1 and Shwethweyin were found as moderately resistant varieties. Based on this finding, susceptible varieties; Yet-90, Sinthukha, Pawsanmwe and Manawthukha are not probably suitable to grow where there is a potential for natural infection of root rot disease. Moderately resistant varieties namely Thukhamwe, Pyitawyin, Salt tolerant Sinthwelatt, Shweasean, Myaungmyamay, Pyimyanmarsein, Submergence-tolerant 1 and Shwethweyin could be recommended to grow in disease prevalent areas. However, integrated with other control measures should be attempted for effective disease control.

When comparing the host response of the rice varieties in the field survey and screen house conditions, three varieties; Sinthukha, Yet-90 and Manawthukha showed similar response but not identical results for Shwethweyin. The host response to *H. oryzae* infection of the same rice variety can be different under different environmental conditions (Ramakrishnan *et al*. 1984 and Dash *et al*. 2008). Although Shwethweyin showed susceptible response under field condition, the lowest nematode population was recovered from this variety. The fifth lowest population of *H. oryzae* was also observed in this variety in screen house condition and showed the moderately resistant response. All these observations may indicate that Shwethweyin has some level of resistance to *H. oryzae*. However, further studies should be carried out to obtain rice root nematode resistant variety in the field under different environmental conditions.

Conclusively, the most infested region was Tatkon Township in Nay Pyi Taw Union Territory and rice production in this township may be potential at risk to undergo important yield losses due to *H. oryzae*. Therefore, it is necessary to firstly investigate yield losses of rice due to *H. oryzae* in Tatkon Township. Popular variety namely Sinthukha with the highest nematode population was the most susceptible variety among surveyed rice varieties. In rice-based cropping sequences, the use of a resistant rice variety or dry season crops can reduce the soil population density of *H. oryzae* compared with continuous rice cropping sequences with susceptible varieties. The only economical way to control rice root nematode is the use of resistant rice varieties and avoidance of monocropping and transplanting practices.

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APPENDICES

Appendix 1 Characteristics of twelve rice varieties included in the host response of tested rice varieties to rice root nematode, *Hirschmanniella oryzae*

No.	Variety name	Thukhamwe	Pawsanmwe	Yet-90	Pyitawyin	Salt-tolerant Sinthwelatt	Shweasean
1.	Origin	Myanmar	Myanmar	Thailand	IRRI	Myanmar	India
2.	Parent	Manawthukha/ Nantharmwe	Local variety (pure line selection)	Yet-90	IRBB 60/IR 71730-51-2	IR 53596/ Pokkali	CSR 13/ Panvel 2/ IR 36
3.	Breeding number	Yn 3248-BC_4F_2-112	D44-8	-	IR 77542-90-1-1-1-5	Yn 3220-MAS-62	CSR 36
4.	Released year	2013	1944	2009	2015	2013	2015
5.	High quality/ high yielding variety	HQV	HQV	HYV	Irrigated lowland rice	Salt-tolerant rice	Salt-tolerant rice
6.	Grain type	Let Ywe Zin	Meedon	Emahta	Emahta	Emahta	Emahta
7.	Day to maturity	140	Nov (Flowering)	95	130-137	140-145	116-120
8.	Plant height (cm)	112	165	75.3	127	84	119
9.	Panicles per hill	9-12	6-8	13	9-10	7-8	10-15
10.	1000 grain weight (g)	20.7	30.0	23.8	22.3	30.2	27.6
11.	Grain appearance	Translucent	Not translucent	Translucent	Translucent / Trace white belly present	Translucent	Translucent
12.	Grain no. per panicle	170	180	145	142	125	150
13.	Amylose content (%)	24.4	21.0	29.55	24.1	25.9	23.6
14.	Yield (t/ha)	4.1-5.2	2.1-3.1	5.2-6.2	4.6-5.2	3.9-5.2	5.2-5.7

Rice Division, Department of Agricultural Research (2017)

Appendix 1 Characteristics of twelve rice varieties included in the host response of tested rice varieties to rice root nematode, *Hirschmanniella oryzae* (Continued)

No.	Variety name	Myaungmyamay	Pyimyanmarsein	Submergence-tolerant 1	Shwethweyin	Manawthukha	Sinthukha
1.	Origin	Cambodia	IRRI (Philippine)	India	IRRI (Philippine)	India	Myanmar
2.	Parent	PREMBEIKOUR (IR 41581/ IR 26460// Lonethwemwe)	IRRI 126/ IRRI 135	Swarna/ IR 49830-7-12 (FR 13A)	IR2153/ IR28/ IR36	Taichung 65/*2 Mayang Ebos 80	Manawthukha/ IRBB21
3.	Breeding number	PREMBEIKOUR	IR 10T107	Swarna-sub-1	IR 50	Mashuri mutant 3628	IR Yn 1068-7-1
4.	Released year	2013	2015	2011	1985	1977	2009
5.	High quality/ high yielding variety	Rainfed lowland rice	Salt-tolerant rice	Submergence-tolerant rice	HYV	HYV	Rainfed lowland rice
6.	Grain type	Emahta	Emahta	Let Ywe Zin	Emahta (long)	Let Ywe Zin (medium)	Emahta
7.	Day to maturity	130-135	116-120	155	105-110	135 -140	138-140
8.	Plant height (cm)	127	124	100	100	105	107
9.	Panicles per hill	9-10	10-15	7-9	10-12	10 - 12	10-12
10.	1000 grain weight (g)	25.4	24.5	21.6	19.5	19.0	20.6
11.	Grain appearance	Translucent	Translucent	Translucent	Translucent	Translucent	Translucent
12.	Grain no. per panicle	149	107	163	130	188	220
13.	Amylose content (%)	23.4	21.6	26.6	30.6	26.5	23.7
14.	Yield (t/ha)	4.1-5.2	5.2-5.7	4.1-5.2	5.2-6.2	5.2-6.2	4.6-6.7

Rice Division, Department of Agricultural Research (2017)

Appendix 2 Agricultural practices and some field records in five townships

No.	Township	Village	Cropping sequence	Sowing method	Variety	Soil type
1.	Zabuthiri	Aungzabu	Rice-rice	Direct seeding	Shwethweyin	Sandy loam
			Rice-rice	Direct seeding	Manawthukha	Clay
			Rice-rice	Direct seeding	Manawthukha	Clay
		Taegyikone	Rice-rice	Transplant	Palethwe	Sand
			Rice-rice	Transplant	Manawthukha	Sandy loam
			Rice-rice	Direct seeding	Sinthukha	Sandy loam
		Ahlyinlo	Rice-rice	Transplant	Yet-90	Silt
			Rice-rice	Direct seeding	Manawthukha	Clay
			Rice-rice	Direct seeding	Manawthukha	Clay
2.	Dekkhinathiri	Kyarpin	Rice-rice	Direct seeding	Yet-90	Sand
			Rice-rice	Direct seeding	Yet-90	Sand
			Rice-rice	Direct seeding	Yet-90	Sand
		Ywarma	Rice-rice	Transplant	Sinthukha	Silt
			Rice-rice	Direct seeding	Yet-90	Silt
			Rice-rice	Direct seeding	Manawthukha	Silt
		Kyuntatpae	Rice-rice	Transplant	Manawthukha	Clay
			Rice-rice	Direct seeding	Manawthukha	Clay

Appendix 2 Agricultural practices and some field records in five townships (Continued)

No.	Township	Village	Cropping sequence	Sowing method	Variety	Soil type
			Rice-rice	Direct seeding	Yet-90	Clay
3.	Pyinmana	Zephyupin	Rice-blackgram-rice	Transplant	Shwethweyin	Clay
			Rice-blackgram-rice	Transplant	Palethwe	Clay
			Rice-blackgram-rice	Direct seeding	Yet-90	Clay
		Maezalikone	Rice-rice	Direct seeding	Palethwe	Clay
			Rice-rice	Direct seeding	Pyitawyin	Sand
			Rice-rice	Transplant	Manawthukha	Clay
		Kan Oo	Rice-rice	Direct seeding	Palethwe	Clay
			Rice-rice	Direct seeding	Palethwe	Sandy loam
			Rice-rice	Direct seeding	Palethwe	Clay
4.	Tatkon	Khayansatkone	Rice-rice	Transplant	Palethwe	Sandy loam
			Rice-rice	Transplant	Sinthukha	Clay
			Rice-rice	Direct seeding	Yet-90	Clay
		Nyaungdoneai	Rice-rice	Transplant	Sinthukha	Clay
			Rice-rice	Transplant	Sinthukha	Clay
			Rice-rice	Transplant	Palethwe	Clay

Appendix 2 Agricultural practices and some field records in five townships (Continued)

No.	Township	Village	Cropping sequence	Sowing method	Variety	Soil type
		Yaeaye	Rice-rice	Transplant	Palethwe	Clay
			Rice-rice	Transplant	Sinthukha	Clay
			Rice-rice	Direct seeding	Shwethweyin	Clay
5.	Lewe	Padaukchaung	Rice-blackgram-rice	Direct seeding	Yet-90	Clay
			Rice-blackgram-rice	Direct seeding	Shwethweyin	Silt
			Rice-blackgram-rice	Transplant	Shwethweyin	Sandy loam
		Yaekar	Rice-blackgram-rice	Direct seeding	Yet-90	Sandy loam
			Rice-blackgram-rice	Direct seeding	Yet-90	Sandy loam
			Rice-blackgram-rice	Direct seeding	Shwethweyin	Clay
		Yonepin	Rice-blackgram-rice	Direct seeding	Shwethweyin	Clay
			Rice-blackgram-rice	Direct seeding	Yet-90	Sandy loam
			Rice-blackgram-rice	Direct seeding	Yet-90	Clay