

**NETWORK ANALYSIS OF SURVEY DATA FOR
CHARACTERIZATION OF YIELD REDUCING FACTORS AND
YIELD LIMITING FACTORS OF TROPICAL RICE
ECOSYSTEMS IN SOUTH AND SOUTHEAST ASIA**

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CHAPTER I

INTRODUCTION

Introduction

It is estimated that the human population will increase to nine billion people by 2050 ?. This means that the food production needs to grow accordingly. With rice as the most important crop for regions with highest estimated population growth, rice will be the most important crop. Ray et al. (2013) claimed that future rice production will need to grow by 2.4 percent per year in order to meet the demands of a growing population. However, pests and diseases are significant yield reducing factors in global rice production. Oerke (2005) estimated that rice pests potentially caused losses of nearly 37 percent of global rice production. To maintain food security, we need to increase yield productivity, and decrease yield losses. Addressing these yield-reducing factors is essential for food security not only in rice consuming societies, but also for other societies globally.

Rice is predominantly grown in Asia, where thirty-one percent of global rice production comes from Southeast Asia alone (OECD/FAO, 2012). The most intensive and productive areas are irrigated, where farmers can grow at least two rice crops per year. Approximately 45 percent of the rice growing area in Southeast Asia is irrigated, with the largest irrigated areas being found in Indonesia, Vietnam, Philippines, Thailand (Mutert and Fairhurst, 2002). In South Asia, the two major rice-growing countries are

India and Bangladesh. India has the largest rice growing area globally, approximately 43 million hectares, and contributes 25 percent of global rice production alone. Combined, rice production in South and Southeast Asia contributes nearly half of global rice production. If rice production in South and Southeast Asia is threatened, it will significantly affect global rice production.

Strategies of pest and disease management take sustainability, production efficiency, and environmental protection into account (Mew et al., 2004). To achieve this, interactions between pests and human activities must be studied. A crop health survey may provide the necessary data which, together with adequate methods for analyzing this survey data, can produce preliminary information on their behaviors including major interactions (Savary et al., 1995). Savary et al. (2000b) concluded that the observed injury profiles (*i.e.*, the combination of disease and pest injury that may occur in a given farmer's field) were strongly dependent on production situation. The authors discussed that pest management strategies should be developed according to the patterns of cropping practices, and production situations. However, interactions among pests and environments under different locations or through time are difficult to elucidate, but are essential to design effective strategies of pest management.

To help visualize and understand these interactions, network analysis a promising tool for revealing the interactions among entities within a complex system. Network analysis have been applied in many scientific branches ranging from social sciences to computer sciences to biology (cite). A network model is an abstract model composed of a set of nodes or vertices and a set of edges, links or ties connected to the nodes. Nodes usually represent entities and the edges represent their relations. For example, an ecological network of a food web presents nodes as a species (Krause et al., 2003) and

edges as ecological relationships, or consider a social network of students (nodes) in the school, where the edges are friendships (Moody, 2001).

In order to develop effective strategies of rice pest management, agronomists focus on holistic points related to crop health, but they do not focus on a particular aspects of plant protection. Savary et al. (1995, 2000a) and Savary et al. (2005) discussed that (i) cropping practices strongly associate the pattern of disease syndromes and damage caused by animal pests, (ii) production situations are strongly related to the occurrence of individual disease, and (iii) production situations (a combination of physical and socioeconomic factors that influence agricultural production) represent very large risk factors (positive or negative) for the occurrences of disease syndromes.

These studies contributed to the body of knowledge needed to address crop health management. I have a good idea of which the relation of pests and diseases affect and rice, production situation and their effect to rice yields, but we do not have a clear picture of where individual or groups of pests and diseases occur and how much effect they have on rice yield. One approach to gain more insight into the relationships of rice pests and disease in rice agroecosystems is to develop networks based on a wide range of pest and disease injuries and corresponding yield losses under different rice production situations across Asia.

I.0.1 Objectives of the study

This study aimed to develop network approaches and apply them to analyze crop health survey data conducted in countries in South and Southeast Asia (India, Indonesia, Thailand, and Vietnam).

Specifically, this research aims to:

1. develop a network model based on crop health survey data and characterize relationships among rice injuries under rice agroecosystems in South and Southeast Asia, specifically in Tamil Nadu, and Odisha; India, West Java; Indonesia, Red river delta; Vietnam, and central plain; Thailand;
2. compare the differential relationships of network models under different seasons and/or locations; and
3. apply network analysis to compare differential relationships of rice injuries in the network model under successive levels (low, medium and high) of crop cut data from different farmers' fields in South and Southeast Asia.

I.0.2 Significance of the study

This research will be a significant contribution to rice pest management and will be an innovative approach to study rice crop health survey data. By understanding the relationships of cropping practices and rice injury profiles, over different locations on an international level, this study will be beneficial to the extensionists, agronomists and researchers for employing effective pest control management. Once network models based on crop health survey data are constructed, they will be beneficial to plant health authorities and the people who related to crop protection especially for rice. The network will support them to design specific strategies for rice pest and disease management and to limit the impact of these yield reducing factors.

Moreover, the resulting network model will reveal the relationships between farmer's practices (cropping practices) with the relationships of co-occurrence patterns of rice pest injuries. The approaches help us to understand the characteristics of rice pests and diseases, which will provide the recommendations on how to design rice pest

management. Importantly, it also serves as a new approach for researchers on the subjects of crop losses and epidemiology.

I.0.3 Scope and limitation of the study

I determine co-occurrence relationships of rice injuries caused by animal pests and diseases. Data were from surveys at the farmer's rice fields in South and Southeast Asia (India, Indonesia, Thailand, Vietnam) from 2013 to 2016. The survey data were conducted by following "survey portfolio" (Savary and Castilla, 2009).

CHAPTER II

REVIEW OF LITERATURE

The world's population is growing rapidly. It reached 6 billion people in 1999 and is anticipated to reach 8.1 billion in 2025 and 9.6 billion in 2050 (Alexandratos et al., 2012). Our long-term ability to meet growing needs for food seems uncertain. Thus, one of the greatest challenges is increasing food production in a sustainable way so that everyone can have adequate food and proper nutrition without over-exploiting the Earth's ecosystems.

Rice is predominantly produced in Asia, so much so that thirty-one percent of the rice harvested globally comes from Southeast Asia alone (OECD/FAO, 2012). The highest levels of productivity are found in irrigated areas, the most intensified rice production systems. Farmers can grow more than one rice crop per year here. Approximately 45 percent of the rice growing country in Southeast Asia is irrigated, with the largest irrigated areas been found in Indonesia, Vietnam, Philippines and Thailand (Mutert and Fairhurst, 2002). In South Asia, the two major rice-growing countries are India and Bangladesh. India has the largest rice growing area globally, about 43 million hectares, and contributes 25 percent of global rice production. Combined, rice production in South and Southeast Asia contributes around half of global rice production. If rice production in South and Southeast Asia is threatened, it will significantly affect global rice production.

Pests in rice production are significant yield reducing factors globally. Oerke (2005) estimated that weeds, animal pests, and disease caused losses around 10.2, 15.1 and 12.2 percent of global rice production, respectively. In most Asian countries, rice yields average 3-5 t/ha. One recent survey estimated that between 120 and 200 million tons of grain are lost yearly to insects, diseases, and weeds in rice fields in tropical Asia (Willcoquet et al., 2004). The mean region-wide rice yield loss due to pests was estimated at 37 percent (Savary et al., 2000a).

In crop fields, pests or so-called biotic constraints can be defined as organisms that cause plant injuries and lead potentially to economic losses. Among the pests that attack rice are microorganisms (*e.g.*, viruses, mycoplasmas, phytoplasmas, bacteria, oomycetes, and fungi) that can cause diseases, parasitic plants, weeds, invertebrates (insects, mollusks), and even vertebrates such as rats and birds can cause serious damages.

Rice Pests

Rice fields are human-managed ecosystems, which harbor diverse communities of plant, animal, and microbial species. Many of them indeed benefit with rice plants, such as predators, parasitoids, flowering plants, and soil bacteria (Norton et al., 2010). However, there are some species threatening the rice plant's health and can cause the quantity and quality losses. I briefly review some pests including weeds, animal pests, and diseases that are concerned as the important pests and disease in the rice production.

Weeds are plants considered as unwanted plants in the crop growing area and compete for nutrition, water, and light with crops. Weeds can be the cause of severe yield reduction problems. In Asia, they are estimated to cause yield losses up to 80 per-

cent, depending on rice field conditions (crop establishment, water management, field management). Weeds also reduce grain quality, and increase the production costs such as labor cost and input costs (Moody, 1991; Savary et al., 2005). The field management and environment are the big factors, which are related weed distribution and degree of infestation in rice fields. For example, in direct seeded rice fields are likely to have high weed pressure than transplanted rice fields Juraimi et al. (2013).

Rats cause serious yield reduction problems in many countries in Asia such as Bangladesh, Cambodia, People's Republic of China, India, Indonesia, Lao People's Democratic Republic, Malaysia, Myanmar, Philippines, Thailand, and Vietnam. *Rattus argentiventer* is the major rat pest species in Southeast Asia. Other species are *R. exulans* Peale, *R. rattus* spp., *R. tanezumi*. Crop losses due to *R. argentiventer* are typically about 10 to 20 percent. Rat damage in the rice fields is easily observed when a large number of the tillers are cut. They damage to rice plants at all stages from sowing to storage. At the seedling stage, they chop down the young seedlings and also feed on the endosperm. Rat damages can be observed in both the wet and dry seasons, but they increase in rain-fed rice crops in the wet season. Wide bunds in the fields are favorable habitats. They burrow and build the tunnels and nests, which can cause bunds to collapse (Singleton, 2003).

Golden apple snail (*Pomacea canaliculata* (Lamarck)) is an exotic species in Asia. Its origin is South America. They were introduced to farmers in countries in Asia to increase their income and as a source of protein in their diet, and also as an aquarium pet (Joshi, 2007). Now they are spreading around Asia and have become a major rice pest in the areas where they have founded. Snails attack younger seedlings at deep water levels. (Basilio, 1991) estimated that snail density at 8 snails/square meter could damage

the rice seedlings up to 93 percent. However, seedlings become tolerant to snail attacks at the age of 40 days old (Sin, 2003).

Insect pests are serious threats to rice production in nearly all regions, where rice is grown. Yield losses due to insect pests based on surveys from 11 countries were estimated at 18.5 percent (Pathak and Khan, 1994). The introduction of high yielding technology in the 1960s involving rice varieties with high tillering ability, denser plant spacing, high fertilizer application and irrigation where farmers planted two or three crops per year provided abundant habitat and food sources where the insect pests can reproduce continuously throughout the year. Moreover, the use of nitrogen fertilizers increased the insects' reproductive potential (Bottrell and Schoenly, 2012). In South and South East Asia, rice is grown in warm, humid environments conducive to the survival and proliferation of key insect pests: stem borers, rice loaders, brown planthoppers, whitebacked planthoppers and green leafhoppers.

Stem borers are ubiquitous throughout rice fields in Asia and cause some damage in every rice field every year. There are six species, but rice yellow stem borer, *Scirpophaga incertulas* (Walker), is the most destructive species in Asia, which may cause 20 percent yield loss in early planted rice crops, and 80 percent in late-planted crops Ooi and Shepard (1994). The larvae bore into the stems and eat their way down to the base of the plant hollowing out the stem. If the damage occurred during the vegetative stage the central leafs do not unfold, turn browns and die off, this is called “deadheart”. If this damage occurs during the reproductive stage it results in the drying of the panicles, which may not emerge or do not produce grains. This is called “whitehead”. Yield losses from stem borer damage can reach up to 95 percent in severe infestations Pathak and Khan (1994).

Asian rice gall midges, *Orseolia oryzae* (Wood-Mason) damaged tillers are characterized by pale green color and the stems elongated and converted into hollow tub called “onion leaf” or “silver shoot”. The Asian gall midge could cause significant yield losses of US \$ 80 million annually in India (Bennett, 2004). They are found widely in most rice ecosystems; irrigated, rain-fed wetland, upland and deepwater rice area when rice plant is growing at the tillering stage (Pathak and Khan, 1994).

Rice leaf feeders are diverse such as rice leaffolder (*Cnaphalocrocis medinalis*), rice whorl maggot (*Hydrellia philippina* Ferinoand), rice hispa (*Dicladispa viridicyanea*) (Kraatz), and rice thrips *Stenchaetothrips biformis* (Bagnall). They often attack rice in the early rice stages, causing visible injuries on a leaf. **Rice leaffolder** at the larvae stage fold the leaves by stitching the leaf margins and feed by scraping green leaf tissue Leaves damaged by rice hispa appear irregular translucent white patches paralleling to the leaf veins. **Rice whorl maggot** at the larvae stage feed on the inner margin of unopened leaves. Damaged leaves show white or transparent patches, pinholes, and are easily broken by the wind. They are commonly found in irrigated fields, but not in upland rice. **Rice hispa** feeds green tissue on the leaf. The leaves show white streaks parallel to the midrib. The rice hispa is common in rainfed and irrigated wetland environments and is more abundant during the rainy season. **Rice thrips** are an important pest to rice at young stage (during the seedling stage or two weeks after early sowing). The damage caused by rice thrips can be observed in the dry season than wet season. Leaves damaged by thrips are curl and discolored. However, the injuries caused by these insect pests are not considered important as often does not translate into a yield loss because of plant compensation (Pathak and Khan, 1994; Shepard et al., 1995).

Rice sucking insects cause enormously economic damages directly by feed-

ing, and indirectly by transmitting many virus diseases. Two species of planthopper, the **brown planthopper** (BPH), *Nilaparvata lugens* (Stal); and the **whitebacked planthopper** (WBPH), *Sogatella furcifera* (Horvath), infest rice. Nymphs and adults feed at the base of the tillers, then plants turn yellow and dry up, turn brown and die. At early stages of heavy infestation round and yellow, and brownish patches appear due to the drying up of the plants. This condition is called “hopperburn”. It can cover large patches in rice fields. Moreover, the BPHs can transmit rice ragged stunt and rice grassy stunt diseases. Another important planthopper pest of rice is the **smaller brown planthopper** (SBPH) *Laodelphax striatellus* Fall én, which is mainly found in temperate rice areas. can transmits rice virus diseases such as rice stripe virus disease and rice black-streaked dwarf virus disease Zhang et al. (2014). Two species of **green leafhoppers** (GLH), *Nephotettix malayanus* and *Nephotettix virescens*, infest rice. They feed on the surface of the leaf blades rather than the leaf sheaths and the middle leaves. The rice plants growing in the nitrogen rich fields are vulnerable to GLH. Plants severely damaged by GHs showed withering, and complete drying. They can transmit many Rice tungro disease (Ling et al., 1972). They also can spread rice tungro disease. **Rice black bugs** belong to several species, but Malaysian black bug *Scotinophara coarctata* is the most common in Asia. They feed on the rice plant from seedling to maturity growth stages. Their damage can cause reddish brown or yellowing of plants. Leaves also have chlorotic lesions. Severe damage may show the symptom called “bugburn” showing wilting of tillers with no visible honeydew deposits or sooty molds. Plants are also stunted; and can develop stunted panicles, no panicles, incompletely exerted panicles, and unfilled spikelets or whiteheads at booting stage (Shepard et al., 1995).

Grain sucking insects causes serious yield losses. **Rice bugs** or slander rice

bugs were found commonly two species; *Leptocorisa oratorius*, and *Leptocorisa acuta* Thunber. They are found commonly in the flowering to milk stages in all rice environments. Nymphs and adults feed on developing grain causing them deformed, spotty and empty grains. They are reported that can cause yield loss up to 30 percent. **Rice stink bugs** were reported that they damage rice grains, but the most common stink bug is *Nezara viridula* (Linnaeus). They penetrate the grain hull with their strong mouthparts to feed on the endosperm (Shepard et al., 1995).

Rice diseases result in yield reductions of 10 - 15 percent in tropical Asia (Willocquet et al., 2004). Sheath blight and brown spot are important diseases in rice in Asia, each responsible for 6 percent of yield losses, whereas rice blast, bacterial leaf blight account for 1 - 3 percent and 0.1 percent of yield losses, respectively. Sheath rot, stem rot, and those known as sheath rot complex and grain discoloration are responsible for rice yield losses ranging from 0.1 - 0.5 percent, respectively. All other diseases alone or in combination do not cause more than 0.5 - 1 percent of yield losses based on estimates (Savary et al., 2000a; Mew and Gonzales, 2002). I briefly review some rice diseases that are commonly found in South and South East Asia.

Foliar diseases include brown spot, rice blast, narrow brown spot, bacterial leaf blight, bacterial leaf streak, *etc.*

Brown spot, caused by *Cochliobolus miyabeanus* (Ito & Kuribayashi) Drechs. ex Dastur (anamorph: *Bipolaris oryzae* (Breda de Haan) Shoemaker) is one of the most important disease of rice in Asia, which is reported across South and South-East Asian countries. The disease symptoms can be observed small oval or circular, dark brown spots on leaves or glumes. The disease is seed-transmitted and infected seed is the

primary inoculum source. The disease develops quickly in fields with scarce water supply and nutritional imbalance, particularly low nitrogen, phosphorus and potassium. Yield loss caused by this disease have been reported many rice production area (Barnwal et al., 2013). On average, the lowland rice of tropical and subtropical, Savary et al. (2006) claimed that 10 % of attainable yield was damaged by this disease.

Rice blast, caused by *Magnaporthe grisea* (Hebert) Barr) (Rossman et al., 1990) (anamorph: *Pyricularia grisea* Sacc., synonym *P. oryzae* Cavara. This fungal pathogen can overwinter in rice straw and stubble and spreads rapidly by airborne spores. The symptoms can be found on all part of the plant including leaves, leaf collars, necks, panicles, pedicels, roots, and seeds. Leaf blast symptoms are characterized by elliptical or spindle shaped lesion with whitish-gray or greenish center and brown or purple margins with yellow halo. Neck or panicle blast, which is more damaging, appear as a dark necrotic lesion covering partially or complete around the panicle base or secondary branches. It may lead to breaking of panicles resulting in few or no grain setting. Disease is favored by high nitrogen. Soils with poor silica availability are blast conducive. Blast is highly destructive in lowland rice in temperate and subtropical Asia, and upland rice in tropical Asia, Latin America and Africa (Skamnioti and Gurr, 2009; Ou, 1985).

Narrow Brown Spot, caused by the fungus *Cercospora janseana* (Racib.) O. Const., synonym: *C. oryzae* Miyake (teleomorph: *Sphaerulina oryzina* K. Hara), ^{To do (??)} characterized by small, narrow, elongated dark brown spots spreading uniformly over the leaf. Symptoms are usually extensive during the later stages of growth, with lesions appearing just prior to anthesis. Severe damage decreases the grain quality and reduces the milling recovery, but has no significant effect on yield losses Ou (1985).

Bacterial leaf blight or bacterial blight, caused by *Xanthomonas oryzae* pv. *oryzae* (Ishiyama) Swings *et al.* (synonym: *X. campestris* pv. *oryzae* (Ishiyama) Dye is characterized by yellowing and dry leaves. The disease developed at the tips of the leaf and spread down one or both sides of the leaf and maybe through the middle leaf. The disease occurs in both tropical and temperate environments, particularly in irrigated and rainfed lowland areas. The high wind and rain were believed to aid in rapid dispersal of the pathogen. In severe epidemics, yield losses can range from 20 - 40 percent (Sonti, 1998).

Bacterial leaf streak, caused by *Xanthomonas oryzae* pv. *oryzicola* (Ishiyama) Swings *et al.* is characterized by fine translucent streak between veins. Then the streaks become yellowish-gray, the lesions coalesce, then eventually turn to brown to greyish white causing the leaves to die. This disease is common in South and South-East Asia. Severe infection results in poor grain development, broken rice and deterioration in chemical and nutritional composition Ou (1985).

Diseases on the tiller include sheath blight, stem rot and bakanae

Sheath blight, caused by *Thanatephorus cucumeris* (A.B. Frank) Donk (anamorph: *Rhizoctonia solani* Kühn) is characterized by irregular lesions usually found on the leaf sheaths (initially water-soaked to greenish gray and later becoming grayish white with a brown margin). Sclerotia (small brown-to-black, rocklike reproductive structures) may be present on the stems. Symptoms are usually observed from tillering to milk stage in a rice crop. This fungus lives in the soil and floats to the top when fields are flooded, contacts rice plants and spreads to adjacent plants. Spread of sheath blight is thus favored by dense crop canopies Ou (1985). It occurs throughout the rice growing areas. Sheath blight causes a yield loss of 6% across lowland rice fields in tropical Asia Savary *et al.*

(2006).

Stem rot, which is caused by *Magnaporthe salvinii*, anamorph: *Sclerotium oryzae* Catt, has been reported from most rice-growing countries. The infected plants showed initially small, irregular black lesions on the outer leaf sheath near water level. The lesions enlarge and reach the inner leaf sheath. The stems became rot causing plants lodge. The small black sclerotia, fungal reproductive can be found in the rotting stem, and carried in stubbles after harvest. The presence of this disease was reported in most of rice growing countries Ou (1985). Reviewed by Ou (1985), this disease was the most common in Vietnam, caused yield loss around 50%. In the Phillipines, yield loss was estimated range 30 to 50 % caused by this disease.

Bakanae caused by *Gibberella fujikuroi* Sawada (anamorph: *Fusarium moniliforme* J. Sheld) is commonly found throughout Asia. Infected seedlings elongate abnormally, becomes slender and the leaves turn pale yellow green. Infected plants develop roots at the upper nodes and the whole plant turns yellow. At booting stage, some infected plants die. This disease was reported that it can cause yield loss up to 20 percent in outbreak areas Ou (1985).

Panicle diseases commonly include sheath rot, dirty panicle complex or grain discoloration, and false smut.

Sheath rot, caused by *Sarocladium oryzae* (Sawada) W. Gams & D. Hawksworth (Synonym: *Acrocyldrium oryzae* Sawada), reduces grain yield by retarding or aborting panicle emergence, and producing unfilled seeds and sterile panicles. Sheath rot also reduces grain quality by causing panicles to rot and grains to become discolored. The typical sheath rot lesion starts at the uppermost leaf sheath enclosing the young panicles. It appears oblong or as irregular spot with dark reddish, brown margins, and gray center

or brownish gray throughout Ou (1985).

Dirty panicle or grain discoloration is characterized by darkening of glumes of spikelets, brown to black, including rotten glumes. This disease caused by multiple fungal species which include, (*Alternaria alternata*, *Altrernaris padwickii*, *Cochilobolus miyabeanus* (synonym: *Bipolaris oryzae*) *Cuvularia* spp., *Fusarium* spp., *Magnaporthe salvinii*, *M. grisea*, *Nigrospora oryzae*, *Sarocladium oryzae*. These pathogens contributed to deleterious effect on the quality of seed lots. Rice seedlings are poor at field emergence, and survival. Finally, they are vulnerable to pests and diseases (Ou, 1985) and (Mew and Gonzales, 2002).

False smut caused by *Villosiclava virens* gen. nov., comb. nov. (anamorph: *Ustilaginoidea virens* (Cooke) Takahashi) is characterized by the grain transformed into a mass of spore ball, which is initially orange, then become olive-green, and then greenish black at maturity (Tanaka et al., 2008). False smut of rice is reported in most rice-growing areas of the worlds. In favorable conditions, which are high humidity, and soils with high nitrogen content, yield loss caused by this disease can reach to 7 - 75 percent (Ou, 1985). Additionally, the pathogen could produce two kinds of mycotoxins; ustiloxins and ustilaginoidins. They exhibit a variety of biological activities such as antimitotic activity, cytotoxic activity Meng et al. (2015).

Viral diseases include rice grassy stunt disease, rice ragged stunt disease, and rice tungro disease, etc.

Rice grassy stunt disease, caused by *Rice grassy stunt virus* is transmitted by brown planthopper. Infected rice plants show stunting and proliferation of short, erect, and narrow leaves that are pale green or pale yellow. The virus reduces yields by inhibiting panicle production. Plants can be infected at all growth stages. They are

most vulnerable to infection at the tillering stage. Infected stubble and volunteer rice are sources of rice grassy stunt virus. The virus cannot be transmitted via brown planthopper eggs (Ou, 1985; Ling et al., 1972).

Rice ragged stunt disease, caused by *Rice ragged stunt virus* is transmitted by brown planthopper. Leaves of infected plants have a ragged appearance. Rice ragged stunt virus infection is particularly high in tropical conditions where rice is planted all year around and provides a continuous host for the brown plant hopper vector. Infected stubble and volunteer rice are sources of rice grassy stunt virus. The disease reduces yield by causing partially exerted panicles, unfilled grains and plant density loss. Infected crops will have significant yield losses of up to 80 percent (Ling et al., 1972).

Rice tungro disease, caused by *Rice tungro spherical virus* and *Rice tungro bacilliform virus*, is transmitted by green leafhoppers. Tungro disease can occur during all growth stages of the rice plant. It is most frequently seen during the vegetative phase. Plants are most vulnerable at tillering stage. It causes leaf discoloration, stunted growth, reduced tiller numbers and sterile or partly filled grains. Tungro is one of the most damaging and destructive diseases of rice in South and Southeast Asia. In severe cases, Tungro susceptible varieties infected at an early growth stage can have as high as 100 percent yield loss (Ou, 1985).

Farmers encountered an estimated average of 37 percent of their yield loss because of pests and disease. This proportion will be reduced when the farmers have efficient pest management. An important concept applied in pest management is “disease triangle”^{To do (??)}, which explains that an occurrence of plant disease epidemic need three components, virulent pest, susceptible plant and conducive environment. At the farm level, we emphasize the interactions at population level. These interactions depend on

not only the physical, biological environment but also man-made activities (Fig.??). According to Savary et al. (2006), pest management tetrahedral first discussed by Zadoks and Schein (1979) consists of four elements, a pest, crop, the environment and human. Human is recognized the important role in agroecosystem and should be considered to sustainable pest management (Zadoks, 1985). Humans are not limited to farmers, but also included to farmers' communities, social networks, agro-technology suppliers, food-chain stakeholders, research and extension, and policy-makers.

Crop, C, incorporates elements pertaining to the host plant genetic make-up, including host plant resistance (HPR), the crop physiology, the crop phenology, and their interactions. C also incorporates microclimate factors that may influence the behaviour of crop-pest systems. This is because, while the microclimate in a crop is driven by physical meso-environment (under E), micro- climate also depends on the crop structure (i.e., its density and architecture). Thus, C not only accounts for the direct effects of HPR, but also for the conditions under which HPR may operate. Furthermore, the expression of resistance depends on the physiology of the crop (predisposition; Schoeneweiss, 1975).

The interactions between human activities and pathogens are considered indirect, but often very strong. For example, crop establishment (e.g., a crop rotation, the choice of a varieties, a crop establishment method), water and nutrient management, and pesticide use are strongly linked to the pest and disease outbreak Zadoks and Schein (1979). This is of importance to design effective pest control management, which is involved the sustainable plant protection (IPM). Moreover, IPM strategies are field-dependent due to the fact that the combination of pests and diseases is vary from different location and crop management practices Mew et al. (2004); Savary et al. (2012).

According to Savary et al. (2000a,b), The amount of yield reduction varies from the different the injury profiles.

Crop establishment and pests

Crop establishment methods of rice are various, but mainly can be categorized into two types, direct seeding and transplanting.

Weed composition in the rice field is constantly changing. Some weed species are favorably affected by production practice while some are adversely affected. Direct seeded rice more possibly encounters with weed competition than transplanted rice because they emerge simultaneously with rice seedlings and because of the absence of flooding in the early stages. Generally, weeds such as grasses, sedges, and broadleaf weeds are found in direct seeded rice fields, which dominant weeds are *Echinochloa crus-galli*, and *Leptochloa chinensis* among grasses, *Cyperus difformis*, and *Fimbristylis miliacea* among sedges, and *Ammania baccifera*, *Eclipta prostrata*, and *Sphenoclea zeylanica* in the broadleaf category Juraimi et al. (2013).

Compared with transplanted rice, the occurrence of insect pests and diseases is more intense in direct seeded rice because of high plant density and the consequent cooler, more humid, and shadier microclimate inside the canopy Pandey (2002), are conducive to different epidemiological conditions Willocquet et al. (2000). The major insect pests of direct seeded rice are brown planthoppers, stem borers, leafhoppers, and gall midges. Important diseases that affect direct seeded rice are blast, ragged stunt disease, yellow orange leaf disease, sheath blight, and dirty panicle (Pongprasert, 1995). In addition to insect pests and diseases, other pests that attack emerging rice seedlings are the golden apple snail (*Pomacea canaliculata*) and rats, which are more serious

problems in direct seeded rice than transplanted rice Pandey (2002).

Nutrient management and pest occurrence

Nutrition management is one of the most important practices of a high production system, but nutrition management will affect the response of rice to pests, as well as development pattern of pest populations due to the change of conditions Zadoks and Schein (1979); Willocquet et al. (2000). Soil fertility practices can impact the physiological susceptibility of crop plants to insect pests by either affecting the resistance of individual plant to attack or by weakening plant vulnerability to certain pests. Some studies have also documented how the shift from organic soil management to chemical fertilizers has increased the potential of certain insects and diseases to cause economic losses Castilla et al. (2003).

Nitrogen, phosphorus and potassium are most often managed by the addition of fertilizers to soils. The others are most often found in sufficient quantities in most soils and no soil amendments are needed to ensure adequate supply unless soil pH limits them. However, I will briefly summarize the effects of nitrogen, phosphorus, and potassium with their relationship to some of the most important rice diseases and insects.

Nitrogen can prolong the vegetative period and increases the proportion of young to mature tissues in rice plants. It can also reduce the amount of cellulose in plant cell walls, predisposing plants to lodging. Aside from these, increased nitrogen was found to reduce phytoalexins in plant cells. Phytoalexins are anti-microbial compounds that build up in plants as a result of infection or stress, and are associated with resistance to fungal and bacterial diseases. On the other hand, if applied correctly, nitrogen allows better plant compensation and tolerance to injury. Bacterial leaf blight can be aggravated by

too much nitrogen, and it is even more problematic during the wet season. This is one reason why the recommendation for nitrogen fertilizer during the wet season is lower compared to the recommendation during the dry season. Other than bacterial leaf blight, excessive nitrogen can also be conducive to the development of many other diseases, such as bakanae, bacterial leaf streak, false smut, leaf blast, sheath blight, and sheath rot Castilla et al. (2003).

As to the effect of nitrogen on the disease, excessive nitrogen application enhanced occurrence of insect pest, especially stem borers, whorl maggots, brown planthoppers and leafhoppers (Chau et al., 2003; Litsinger et al., 2011; Rashid et al., 2014). Except whorl maggot and rice thrips, their population did not increase following as the use of nitrogen increase (Chau et al., 2003).

The effects of phosphorous on pest occurrence

Just as important as nitrogen, phosphorous is another essential nutrient element that promotes vigorous root development and is responsible for hardening plant tissue. Phosphorus also shortens vegetative period of the plant, as opposed to the effect of nitrogen. Proper timing in the application of phosphorus may lower the incidence of brown spot and leaf blast Castilla et al. (2003).

In relation to disease incidence, what we know is that phosphorus can reduce the occurrence of bacterial leaf blight and brown spot, but may promote leaf blast and sheath blight of rice. The effect of phosphorus on the population of rice insect pests was not strong (Chau et al., 2003; Rashid et al., 2014), but the results of Rashid et al. (2014) showed that increased phosphorus application enhanced the population of brown planthoppers.

The effects of potassium on pest occurrence

Among the three essential nutrients mentioned, potassium appeared to be the most consistent and effective in minimizing disease incidence. It was found to lower the infection rate of bacterial leaf blight, leaf blast, and brown spot, sheath blight, sheath rot, stem rot and narrow brown spot Ou (1985). This is because potassium is found to increase the concentration of inhibitory amino acids in the plants as well as phytoalexins Castilla et al. (2003). Potassium also hardens the plants tissues which can minimize lodging incidence and support faster recovery of injured or stressed plants Harrewijn (1979). There were not many studies about the effect of potassium on the rice insect pests. Rashid et al. (2014) showed the result in Bangladesh that high potassium application decreased population build up and dry weight of brown plant hoppers and Salim et al. (2002) reported that deficiency of potassium in rice plants increased population build up of white backed planthopper and application of high dose of potassium to rice plants decreased population build up of the insect.

Crop establishment and pests

Crop establishment methods of rice are various, but mainly can be categorized into two types, direct seeding and transplanting.

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Common techniques for analysis of survey data

Methodologies used to interpret the survey data are developed for studies of the relationships for injuries - yield losses. The development of these studies started focusing on single-disease (pest) pathosystem to more complex. The classic analytical approaches aim to assess group-wise differences, either in a univariate *i.e.* parameter-by-parameter fashion or using multivariate techniques (*e.g.* multivariate analysis of variance (MANOVA), correspondence analysis (CA), multiple correspondence analysis (MCA), or principle component analysis (PCA)). Univariate methodologies are frequently used to reduce a possibly large number of measured analyses to only those that show the strongest response under the investigated conditions. Examples for such univariate approaches are significance testing methods and simple linear regression model. However, univariate methods fail to apply to different production situation or the scope that model can be applied.

Therefore, multivariate analysis methods seek to capture not only changes of a single pest. Probably the most prominent multivariate analysis techniques applied in the field of survey data are principal component analysis, cluster analysis and correspondence analysis. These analyses do not always; and to yield loss estimate^{To do (??)}. They can inform us the close relationship between changing production situations and shift crop health syndromes (changes in the dynamics of group disease or crop-limiting factors in an area)^{To do (??)}.

Farmers' fields survey datasets are of immense value to plant pathologists; they, however, are generally heterogeneous in format (being qualitative or quantitative) and/or precision. A number of methods, some now old and seldom used, and other more recent and comparatively easier to use, are available to process this type of information. The approach used to analyse these data included three main steps: cluster analysis, multiple correspondence analysis and logistic regression.

Cluster analysis represents another unsupervised multivariate technique suitable for the analysis of survey data with hierarchical cluster analysis and k -means clustering being the most prominent representatives. In general, clustering methods group and visualize samples according to intrinsic similarities in their measurements, irrespective of sample groupings.

Survey in the field of epidemiological research

To do (??)

The rice crop system is complex that the number of components is large and their interaction are complex To do (??). The dynamic of the system is encompassed by a growing crop, its physical environment, its diseases and pests, and farmers' actions, which influence the whole system. The number of rice pests (diseases, insects, and weeds) to be considered and the levels at which they vary from field to field. A rationale is often needed to delineate the limits of the system to be addressed To do (??).

A survey is a useful tool to provide an adequate account of this diversity and deal with the several pests or variables affecting crop production, which are essential components of systems research in plant protection Zadoks and Schein (1979). Field surveys generate information at the level of the population of crop stands rather than of

the individual plot, field, or orchard. Such surveys raise questions regarding variation in injury and its relationships to crop management, crop environment, crop performance, and the occurrence of multiple injuries.

Application of surveys can be found in a huge diversity of research fields, including crop characteristic, cropping practices and single disease or multiple disease and insect. The pest are measurable biological variable that can be used for characterize the injuries profiled (disease and insect injuries syndrome) to stratify farmer's fields account into the characteristic of their production situations.

Savary et al. (1995) emphasized field surveys as the basic means for acquiring information and expanding knowledge to higher scales. Conversely, field surveys and the associated analytical approaches provide a framework in which to characterize disease intensities and their relationships with attributes (including crop management) of the environments where injuries occur; but they usually do not generate crop loss data *per se*.

The concept of production situation described the set of factors - physical, biological, and socioeconomic - that determine agricultural production^{To do (??)}. This concept is used here within the restricted scope of an individual rice field and from the limited view point of plant protection. A quick overview of the variables listed in the following survey procedure indicates how this concept is operationalized in a survey: this list includes a few key components of rice crop management (which reflect the physical and socioeconomic environments of a crop and their interactions) and a series of rice pests (that is, insects, pathogens, and weeds).

This approach has the considerable advantage of providing quantitative estimate of the contribution of each constraint to the total yield loss. They study may be

considered among the first series, where a set of pest constraints of a crop, simultaneously with environmental variables, and variables representing the current cropping practices were considered. Pests only considered some of the “yield determining variables”. the approaches may also be considerate an attempt to incorporated the concept of production situation *i.e.* the set of environment that lead to a given yield output of the system^{To do (??)}.

The applications of network analysis have increased exponentially over the past two decades in various disciplines. Even though documented applications of network analysis in plant pathology are still relatively sparse, network applications in the social sciences, systems biology and ecology have been increasingly found. Here I review the empirical works that exist and argue that network analysis is a promising approach for exploring questions in the context of plant pathology.

Network Analysis

Introduction to network analysis

Network analysis is used for determining relationships between elements of interest. It offers toolkits for visualizing data in a network model and measuring its properties, and network thinking (Proulx et al., 2005). It has been widely used by various branches of science, such as social sciences, ecology, biology, computer science, and many others to study the interactions between elements, *e.g.*, the relationships of students in school (Moody, 2001), species in food webs (Krause et al., 2003), interactions of genes or proteins in cells (Guimera and Amaral, 2005), or the connections of computer in the network (Pastor-Satorras and Vespignani, 2001; Newman, 2006).

Newman (2003) loosely categorized four types of networks based on different complex data. The first category is social network, representing sets or groups of people forming some patterns of contacts or interactions between them such as the patterns of friendship or business relationships. Analyzing the structure of whole social entities gives us the perspectives from a social network, which enables us to explain the patterns observed. Moody (2001) analyzed the social behaviors in high school students using social network approaches. (Kasari et al., 2011) applied network analysis to compare the social relationships and friendships between children with and without autism spectrum disorder (ADS). The second type of network is an information network or knowledge network. The classic example of this network is the network of citations between academic papers (Newman, 2003). The articles cited other papers, which have related topics. They formed a citation network that has vertices as articles and direct links as citations. The citation network visualizes the structure and the movement of the information. The third category, technological network, is object connected network, or man-made network which represents a physical connection between objects. This network is mostly applied for illustrating physical structures and systems such as the electrical power grid, the connections of rivers, transport systems, etc. The fourth category of network is a biological network. It represents the biological systems such as genes to genes, genes to protein, protein to protein interactions, which enable biologists understand the connections and interactions between individual constituents including genes, proteins, and metabolites at the level of the cell, tissue and organ to ultimately describe the entire organism system. Biologists use biological networks in various branches of biology at different levels (from a single molecule to an entire organism). For example, Yang et al. (2014); Barabási and Oltvai (2004) studied in the patterns of gene expression

in different conditions and different types of cells (normal cells and cancer cells) in order to characterize the genes that change and do not change following the particular conditions; Freilich et al. (2010) applied a molecular ecological network analysis to study the communities of soil microorganisms. Networks revealed the complex relationships between microbial species in soils and their communities. Moreover, network analysis enables ecologists to understand ecological properties and predict the ecological roles of species in a soil ecosystem. Although the application of each type of network approach varies, all four categories of networks share a common empirical focus on relational structure and a similar set of mathematical analysis.

Network analysis can be a powerful tool to study plant disease. Moslonka-Lefebvre et al. (2011); Jeger et al. (2007) applied network models and concepts to study disease spreads in regional networks of plant nurseries and garden centers of *Phytophthora ramorum*, the oomycete causing Sudden Oak Death in the West Coast of the USA and leaf blight and dieback in many ornamental shrubs both in America and Europe. The result could be applied to design measures to control plant disease epidemic from movement of infected material among plant nurseries. Shaw and Pautasso (2014) recently reviewed critically about network analysis to plant disease management (*i.e.*, to design ways to reduce the flow of disease in traded plants, to find the best sites to monitor as warning sites for annually reinvading diseases, to understand the fundamentals of how plant pathogen spreads in different structures of simulated trade network models) To do (??). Windram et al. (2014) reviewed the applications of network analysis in molecular plant pathology. Network models were applied to reveal plant defense mechanism during plant-pathogen interactions.

Concepts, principles, and methods of network analysis

A network represents relationships between elements of interest, which are defined by links (edges) among nodes (vertices). Nodes can be units of interests or studies, and links represent interactions between nodes. Network analysis aims to identify the patterns of associations among nodes, not only features or attributes of particular nodes.

Network analysis follows three principles. Nodes and their behaviors are mutually dependent, not autonomous; links between nodes can be channels for transmission of both material (*e.g.*, money, disease) and non-material (*e.g.*, information, knowledge, relationship, interaction) and; persistent pattern of association among nodes create structure that can define, enable, or restrict the behavior of a node.

Network models have two different structures depending on goals of the representation and analysis (Borgatti et al., 2013). Flow models, commonly named directed graphs, represent the network as a system of pathways along which something move such as transportation networks (*e.g.*, highways, railways, and airlines) or communication networks. Since flow networks show the directions of movement between nodes, thus these networks are interesting to study their behaviors. Analysis of flow networks can identify which nodes are more active or which ones are more important connectors. Jeger et al. (2007); Shaw and Pautasso (2014) applied such network models to study plant disease spread. Architectural models, or undirected graphs, are mainly used to determine the structure of the network, seeking to discern whether specific structures lead to similar outcomes or whether nodes in similar network positions behave in similar ways. Ecological applications related to the ecology and spatial structure of “community” tend to be organized and analyzed as architectural models. For example,

Faust and Raes (2012) studied networks of soil microbial interactions. Network models can describe how microbial populations change over time, which will require the use of dynamic models of microbial communities. Beyond these basic principles, network analysis enables the calculation of structural properties of nodes, groups, or the entire network.

Measuring network properties

A network is made up of nodes and links from relational data. It is constructed from an adjacency matrix, which is obtained from analysis using metric algebra techniques. The row and column headings for an adjacency matrix are identical, listing the names of the components involved in the network. In the simplest case, the cells of the matrix are coded with “1” if a link exists between the node or “0” if no edge exists. However, a link can be valued. Value indicates a characteristic of the relationship that the research has quantified. The values may be binary, such as whether two friends recognize each other, or variable strength (*e.g.*, the number of mutual friends between two friends). A network link need not imply positive or cooperative interaction; they can also be a negative or competitive interaction between two individuals.

The distribution of links in a network suggests two important structural characteristics: centrality (importance) of nodes in the network and division of the network into subgroups. Variants of centrality in a network include degree, closeness, and betweenness. Degree centrality of a node is the sum of the value of the links between that node and every other node in the network. This measure tells us how well-connected a particular node is to the other nodes. Closeness centrality is calculated using the length of the path between a node and every other node. This measure could estimate the time required for information or resources to propagate to a given node in a network. Be-

tweenness centrality corresponds to the number of paths in the network that pass through a particular node, and therefore measures the dependence of a network on a particular node for maintaining connectedness (Toubiana et al., 2013). Deng et al. (2012); Newman (2003); Toubiana et al. (2013) are recommended references for descriptions of the theory and uses, as well as the formal calculation of these measures.

The unique values of network analysis

There are four key points that will help to understand network analysis 1) how it differs from traditional approaches of scientific research; 2) how it relates to those traditional approaches; 3) how networks are constructed, manipulated and measured; and 4) what value network analysis offers beyond traditional approaches.

The first point of network analysis is that there are two types of data represented in the network graphs; technical and rational data. The first is data characterizing the actors or variables being studied referring to attributes. Attributes describe characteristics of individual actors or variables, for example their race, income or physical location, and are the primary variables considered in traditional approaches. The second type of data is relational data, that is, data about the relationships between individual nodes. For example, Lazega and Pattison (1999) represented the network model of cooperation among lawyers in three law firms, through the exchange of various type of resources among them. This model consisted of over 70 lawyers in three different offices in three different cities. Rational data reflected to resources exchange and additional attribute information were included type of practice, genders, and seniority of each lawyers.

Relationships are also referred to as edges (links) in network analysis. Edges cannot be attributed to any single actor. Rather, edges only exist between nodes. This

leads to the second point about network analysis that it requires a different conceptual approach. Because edges only exist between nodes, it is useful to think of edges existing in a separate dimension from nodes, who are connected in physical space. This dimension is sometimes referred to as relational space. To visualize the difference, think of someone far away with whom you correspond regularly, say using a phone, email, or Facebook. Even though the two of you are not physically close, you have a strong relationship. The two of you are distant in physical space but close in relational space. This notion of relational space is in part what means when he refers to the space of flows as something distinct from the space of places (Castells, 2001).

The third point that distinguishes network analysis from other approaches is it involves different methods of analysis. Because traditional research methods consider variable attributes in a wide variety of statistical analyses such as measures of center (*e.g.*, mean, median, etc.) and dispersion (*e.g.*, standard deviation, range, etc.), these methods are sometimes referred to as variable analysis, whereas, network analysis models relational data and to measure various characteristics of network structure. For example, for lawyers data (Lazega and Pattison, 1999), it is natural to ask to what extent two lawyers that both work with third lawyer are likely to with each other as well. This notion corresponds to the social network concept of transitivity and can be captured numerically through an enumeration of proportion of vertex triples that form triangles, so-called cluster coefficient.

The idea that network structure may be correlated with variable attributes and behaviors is the fourth point to consider in comparing network analysis to other approaches. In network analysis, the arrangement of the network in relational space is basically correlated with the behavior and attributes of those variables. For example, in the

network created by Lazega and Pattison (1999) lawyers of the same firm may share similar attributes such as office location or department, and lawyers in similar roles within that network may share similar behaviors. Basically, conventional approaches measure various attributes of variable (nodes in a network) and attempts to discern something about the relationships between actors (edges in a network) based on those attributes. When the network structure is simple and the differences in node attributes are clear, the conventional analytic approach is sufficient. However when relationships are complex or node attributes are more nuanced, clear answers using conventional analysis may prove elusive. As a result, network analysis offers a tool to help researchers visualize the large network and disentangle some of the relational complexities within the network, just as cluster analysis and multivariate analysis for help research disentangle the complex data.

Networks and Plant Pathology

Recently, a broad expansion of applications of network analysis has occurred across many disciplines. It has been evaluated as a promising tool to study a complex system. Plant pathologists have applied network analysis for their research. Moslonka-Lefebvre et al. (2011); Jeger et al. (2007); Windram et al. (2014) supported that network analysis can be fruitful models in many applications relevant to plant pathology because of its generality and flexibility. For example, the network of main fresh cut flowers movements among European countries was determined the likelihood of introduction of new pathogens and other organisms associated with plants (Eagling, 2007), and plant-pathogen interaction network models were applied to present plant defense mechanisms (Dietz et al., 2010).

The development of network analysis challenges conventional approaches to uncover rational complexities of plant pathology studies. Two fields of research relevant to plant pathology presented particularly strong growth and proved that network analysis has significant potential to augment traditional analysis methods. The first is plant disease epidemiology, which investigates questions related to plant disease spread. The second is plant molecular biology, which investigates questions related to biological networks.

Using Network analysis to understand plant disease spread

Network analysis challenges conventional approaches of studies in plant disease epidemiology, especially underling the spatiotemporal flow of plants and their pathogens (Pautasso et al., 2010). When plant pathological studies were restricted to a single geographical location, there was a limit to thinking about the connections or relationships between plants or fields in different locations. However, network analysis can enlarge the view of studies and can consider whether or not plant pathogens are moving from one field to others in the regions of interest. For example, networks of plant disease spread in trade networks presented the flows of plant disease from infected units (infected plants or epidemic areas) to susceptible units (susceptible plants or areas).

Network models of epidemic development

The idea of plant epidemics is that the probability of infection embedded in the connection or the contact patterns between susceptible/infected plants, and it forms as the networks. Pautasso et al. (2010) showed a network model of epidemic development (susceptible-infected-susceptible model) in a directed network. In the network model, vertices were represented plant, and their attributes were presented the infectious sta-

tus (healthy or infected). The epidemic is started at a single node, then nodes with a connection from the starting infected node will be infected at the next time step with a certain probability of transmission. In turn, already infected nodes will be infected at the next time step depending on their infection status and on a certain probability of persistence. The probability of infection transmission is the same for all connections between infected nodes and susceptible nodes over times. Similarly, the probability of infection persistence is the same for infected nodes in a certain network replicate. For each network structure, the two probabilities of persistence and transmission define an epidemic threshold, which is independent of the starting node of the epidemic. This epidemiological model does not result in either susceptible or infected nodes, as nodes will have an infection status along a continuum. Key quantities for epidemiological dynamics in networks were reviewed in Moslonka-Lefebvre et al. (2011).

Analysis of plant trade network

Phytophthora ramorum epidemic networks in the horticultural trade are an example of the application of network models in the study of plant disease spread (Harwood et al., 2009; ?). Simulations of spread of *P. ramorum* in different network structures (random, small-world and scale-free network) were found that epidemic threshold, the boundary between a no epidemic and an epidemic outcome, is significantly lower for scale-free network, a network is dominated by a small number of nodes with many connections, compared to local, random and small-world network structure. Modeling suggested that it was possible to control an epidemic by changing the structure of network, without having to decrease the probability of infection persistence at a nursery site and/or of transmission between sites. Simulation showed that correlation coefficient between link in and out nodes increased with connectivity level for all the structures investigated

and underlined the importance of targeted control towards node with more connections than others.

Regardless of the network structure and connectivity level, epidemic threshold is negatively correlated to the correlation coefficient between link in and out nodes, (Moslonka-Lefebvre et al., 2009). In presence of high-connected nodes, the most effective way to control disease spread is to move from two-way to a one-way network. That is move from network where overall there is positive correlation among links-in and -out to one where the correlations are negative. In practice this would mean that a nursery network would be dominated by major node, which receives plant materials from many production sites but supply relatively few retail sites, or by major nodes which received plant materials from a few production sites but supply many retail sites. However, the scenario where there are major nodes which both receive plant material from many production sites and supply many retail sites is the most problematic control. The better way to control should target towards the cluster (group of nodes), not only to high-connected nodes.

The last point, the modeling of disease spread in small-size directed networks showed that increasing the proportion of wholesalers (*i.e.*, traders without a preponderance of incoming or outgoing links) tends to decrease the epidemic threshold in local, random, and small-world network. The opposite result is obtained for the proportions of produces and retails. Scale free networks appear instead to be tolerant to changes in these hierarchical categories as the epidemic threshold in this case is governed by the presence of hub rather than by the features of the majority of nodes in the network (Pautasso et al., 2010).

Network models to design strategies of plant disease management

Due to globalization, plant trades among countries are increasing and convenient. They potentially caused risks to plant health when they were not under good control. To control the risks of plant disease epidemic through plant trade, network models were applied to present the flows of trade network of plants and plant products across the world and within countries and to develop strategies of plant disease management (Pautasso, 2015). Networks could be found hubs or highly connected nodes, which represented locations or countries where imported and exported plants or plant parts. Hubs or highly connected nodes were targets for control disease flows in the scenarios that network presented because they were considered to the likelihood of pathogens actually infecting along particular links. Strategies for disease management should be designed by focusing on links to and from hubs, nodes which have high degree of connectivity, so that it increases efficiency to control plant pathogen spreads. The strategies may aim to remove them from the network (Jeger et al., 2007; Moslonka-Lefebvre et al., 2011). Alternatively, strategies may pay attention on them in order to prevent disease spreads. ? suggested placing quarantine efforts on hubs or on connections between major hubs.

Additionally, Pautasso et al. (2008) showed the good examples, which are co-occurrence networks of the *Phytophthora ramorum* infected plant genera different environments. The networks may be helpful in identifying host taxa playing an important role in spreading a certain disease in the seminatural environment, in crop plants, and plants in the trade. Combining genetic network analysis and data on trace forward and trace back on movement of plants nursery trade supported to identified confidentially *P. ramorum* migration. From this approach, it was clear that the pathogen was introduced originally from nurseries, which *P. ramorum* populations in nurseries are genetically ancestral to all Californian forest populations.

Using Network analysis to understand molecular plant pathology

For understanding mechanisms of plant-pathogen interactions, network analysis offers tools to visualize interactions of biological components including genes, proteins, and metabolites, which are related to plant-pathogen interactions. Network concepts enable us to characterize interplay of those components following the properties of network structure. Analyzing structural properties of networks may provide useful clues about the biological functions of individual genes, genes complexes, proteins, protein complexes, pathways they participate in.

Presenting biological data with network model

Networks are used in different contexts as ways to represent relationships between entities, such as interactions between genes, proteins or metabolites. Wu et al. (2007) gave the example of a network model built from gene-for-gene relationships between rice and various avirulence genes of the pathogen *Xanthomonas oryzae* pv. *oryzae* causing bacterial leaf blight of rice. Nodes represented isogenic lines of rice and weighted edges reflected the number of shared genes with high resistance (with respect to avirulence genes) in the two isogenic lines of rice. For a plant breeder, this graph can help in identifying particularly promising genes for developing a disease resistant variety.

Network analysis to study biological systems

Network analyses have been applied to visualize the myriad information and analyze the complex relationships. To better understand the collective impact of genes on complex traits and determine what governs their organization, biologists are most likely to apply gene co-expression networks (Usadel et al., 2009). Co-expression networks most commonly use the Pearson's correlation coefficient to establish linear pairwise cor-

relations between gene pairs in an adjacency matrix. Another associative metric that can be used is the Spearman correlation coefficient, which captures nonlinear correlations between genes to be uncovered (??). Once a co-expression network has been generated, identifying modules by clustering can help extract biological meanings weership, to study the relationships between modules, and to compare the network topology of different network.

Differential Network Analysis

Networks can respond differently under various environments or with external signals. They can be simplified by focusing on key components and capture only the essential components differently responding between environments in which they play a key role in the modeled response (Pe'er and Hacohen, 2011). Networks are examined by adding or deleting some variables. This allows predicting interactions or components that change following the changed structure of networks.

Differential network analysis is applied to identify and describe differences between two networks under different conditions. Differential networks from different data sets with different conditions might display different interactions from the single network based on the whole data set, which are disregarded the conditions. The strongest interactions in differential networks are not necessarily those that are strong in the single network. Conversely, they may be weak or absent in single network. Compared between differential networks under different environment conditions, the differential interactions can be implied that they are a result of response to environmental conditions. Moreover, it can be implied that environments influence on the interaction between pairs of nodes contributing to the differential interactions.

The objective of differential network analysis is to identify the different in-

teractions, that may reveal when condition has changed. For example, the differential network analysis of gene co-expression networks to understand the differences between human and chimpanzee brain was focused on how expression patterns of genes were different in human and chimpanzee brains. These networks are contrasted to find a) non-preserved modules (group of genes with sharing similar expression profiles and presumably similar functions), b) differentially occurred genes, and c) differentially connected genes. The result showed that

Dynamic Network

Biological systems are highly dynamic. They must continuously respond to external signals or the internal state of the system. The responses can be altered slowly or quickly over time (Pe'er and Hachohen, 2011). Thus, realistically the corresponding biological network models must evolve as well. It seems clear that dynamic networks enable us to see and understand the systems and how their dynamic effects change over time. Some understandings previously have been obtained from studies of dynamics of large networks, for example, gene expression or metabolic fluxes network (Ideker and Krogan, 2012).

The main goal of dynamic network analysis moves away from single network analysis, which characterizes absolute properties of the system. It aims to concentrate on a specific dynamic response. Rather than answer what the key factors in the system, it answers what parts of the system are most affected by perturbation. Most commonly, dynamic networks are applied when the edges among a set of vertices or the sets of vertices itself are changing as a function of time. For example,

Summary

This literature review presented a brief introduction of network analysis and concise concepts and methods^{To do (??)}. Briefly, a network is usually represented by sets of nodes (or vertices) connected by edges (or links) in various ways. Networks can be categorized to four types, social network, information, technology network, and biological network. Even though four types of networks are described and applied in different context, they share a common empirical focus on relational structure and a similar set of mathematical analyses. Network models are cable of presenting unique values, which traditional approaches cannot present. Network analysis was discussed as applied to two broad areas of study of plant diseases. It firstly was applied to study plant disease epidemics. For example, networks of *P. ramorum* spread through plant nurseries trade. The results enabled us to understand the directions and processes of disease spread. Additionally, they could predict the movement of disease flows, and improve the implementation of plant disease policy. Secondly, molecular plant pathology showed two applications of network applications. The first use is to apply networks to model large and complex biological datasets. Another use is the consideration network structure to understand biological system. Emergent properties of network structure influences may be identified, measured and analyzed to yield better explanations of the experiments being observed. While numbers of documented plant pathological studies using network analysis are sparse, the literature presented in this review showed a clear and compelling case for plant pathologists to expand the understanding of and use network concepts and methods. Biological networks are context-specific and dynamic in nature. Under different conditions, the topology of the network changes. Network approaches (differential network, dynamic network analysis) are successful to yield insight on biological sys-

tem under different conditions or dynamic processes. Network analysis concepts and methods augment existing approaches and provide tools for exploring complex relationships, which have been widely acknowledged as influential but difficult to measure using traditional methods.

CHAPTER III

Evaluation of correlation methods for constructing a network of rice injuries of rice crop health survey data

III.1 Evaluation of correlation methods for co-occurrence network construction of rice crop health survey data

Introduction

Network analysis is a promising tool frequently used to describe the pairwise relationships of a large number of variables. Network analysis of pest and disease incidence co-occurrence patterns offers new insight into pest management. Since different correlation measures lead to structurally different co-occurrence network and provide different information, selecting the optimal correlation method is critical. Pests are the blocks in the big wall of yield losses that plant protection specialists must scale due to the need to increase crop productivity. Surveys of farmers' fields are useful sources of data to help us determine the importance of pests and understand complex relationships within agroecosystems. Our objective is to find an appropriate method to.

Materials and methods

Survey data

Survey data collected in 450 farmers' fields in irrigated lowland rice growing areas across South and Southeast Asia Tamil Nadu, India (TMN); Odisha, India (ODS), West Java; Indonesia (WJV); Suphanburi, Thailand (SPB) and Mekong river delta, Vietnam (MKD) were collected from 2013 - 2016. The number of survey were summerize in Table.

??

The survey procedure and data were based on a standardized protocol described in “A survey portfolio to characterize yield-reducing factors in rice” developed by Savary and Castilla (2009). Rice injuries were collected in this study including the injuries caused by animal pests, and pathogens, which are harmful to rice plants, and importantly considered to reduce yield productivity. They were evaluated at booting and ripening stage according to survey procedure. Injuries on leaves such as whorl maggot injury (WM), leaffolder injury (LF), bacterial leaf blight (BLB), bacterial leaf streak (BLS), leaf blast (LB), brown spot (BS) were determined as a proportion of injured leaves. Injuries on tillers or hills such as stem rot (SR), sheath rot (SHR), sheath blight (SHB), whitehead (WH), deadheart (DH), silver shoot (SS) caused by gall midges were determined as a proportion of injured tillers or panicles.

Before analysis, data were compacted over time during crop growth. Two types of data were computed, depending on the nature of injury Savary and Castilla (2009). One is an area under injury progress curve (AUIPC) used for injury variables, which present on the leaves, and for weed infestation. Another is the maximum level at any of the two observations used for injury variables that can be observed on tillers, panicles, and hills, and insect pest count. The area under injury progress curve (AUIPC) (Campbell et al., 1990) were calculated by the mid-point method using the following

equation:

$$AUIPC = \sum \frac{1}{2(X_i + X_{i-1})(T_i - T_{i-1})} \quad (III.1)$$

where X_i is percentage (%) of leaves, tillers or panicles injured due to rice pests (e.g., leaf blast, leaf folder), or number of insects (e.g., plant hoppers, leaf hoppers) per quadrat, or percentage (%) of weed infestation (ground coverage) at the i th observation, T_i is time in rice development stage units (dsu) on a 0 to 100 scale (10: seedling, 20: tillering, 30: stem elongation, 40: booting, 50: heading, 60: flowering, 70: milk, 80: dough, 90: ripening, 100: fully mature) at the i th observation and n is total number of observations.

Evaluation

In this study, correlation measures including, Pearson, Spearman, Kendal and Biweight mid correlation were evaluated to discover the true functionally related variables in crop health survey data. The data will have to follow the assumption of correlation measures. The correlation measures will also be able to effectively capture biological relationships that are well published. I proposed three steps for correlation methods selection:

- Checking whether or not the data are normally distributed by visual assessments and statistical tests. I performed Shapiro-Wilk test to check the normality of data distribution.
- Comparing correlation measures by testing similarity of correlation coefficients.
- Identifying the most suitable correlation measure that can capture biological relationships between variables confirm with the published relationships.

III.1.1 Result

Evaluation of four correlation measures, including Pearson, Spearman, Kendall correlation, and Biweight mid-correlation shows two groups clustered according to hierarchical clustering using Euclidean distance. The Spearman and Kendall correlation were grouped in rank-based methods, and another group is non-rank-based correlations including Pearson correlation and Biweight mid-correlation. When choosing a measure, Spearman correlation was selected because its robustness to noise and to outliers. Briefly, network analysis revealed that brown spot is the highest connection. Bacterial leaf streak, brown spot, white-backed planthopper, and brown planthopper have high node of connectivity

For example, a clustering method focuses on the analysis of (dis)similarities, whereas principal component analysis (PCA) attempts to explain as much variation as possible in as few components as possible. Changing data properties using data pre-treatment may therefore enhance the results of a clustering method, while obscuring the results of a PCA analysis.

Heteroscedasticity To determine the presence or absence of heteroscedasticity in the data set, the standard deviations of the metabolites of the analytical and the biological repeats were analyzed (Figure 4). Analysis of the analytical and the uninduced biological standard deviations showed that heteroscedasticity was present both in the analytical error and in the biological uninduced variation (Figure 4A and 4B). In contrast, the relative biological standard deviation (Figure 4C), and also the relative analytical standard deviation (unpublished results), showed the opposite effect. Thus, metabolites present in high concentrations were relatively influenced less by the disturbances resulting from the different sources of uninduced variation, and were therefore

more reliable.

III.1.2 Discussion

The appropriate correlation measures for studied data should closely associate to the prior knowledge of biological correlation. For this study, nonlinear relationship between the abundances of brown plant hopper and white backed plant hopper was detected by a Spearman correlation but not by a Pearson correlation.

However, Pearson correlation coefficient is sensitive to outliers. Biweight mid-correlation is considered to be a good alternative to Pearson correlation since it is more robust to outliers [23Wilcox R: Introduction to Robust Estimation and Hypothesis Testing. Academic Press, San Diego; 1997.].

CHAPTER IV

Using network analysis to examine co-occurrence patterns of animal pests and disease in farmers' fields in irrigated lowland rice growing areas in South and South East Asia

IV.0.1 Introduction

Agricultural crop plants are frequently injured, or infected by more than one species of pests and pathogens at the same time. Many of these injuries may affect yields. Because of this co-occurrence in injuries, the idea of “crop health” has been highlighted and implemented to manage the combination of injuries or so-called injury profiles (Savary et al., 2006). Co-occurrence patterns of injuries are beginning to provide important insight into these injury profiles, which possibly present co-occurring or anti-co-occurring (mutually exclusive) relationships between injury-injury. Uncovering these patterns is important to implications in plant disease epidemiology and management. However, there are only a few reports of injury–injury relationships in rice crop systems are currently unknown. This could be a difficult task since complex patterns of injury profiles are related to environmental conditions, cultural practices, and geography (Wilocquet et al., 2008).

To address this issue, we used in-field surveys as a tool to develop ground-truth databases that allowed us to identify the major yield reducing pests in irrigated lowland rice ecosystems. These sorts of databases provide an overview of the complex relationships between crop, field management, pest injuries, and yields. Several previous studies Savary et al. (2000b,a); Dong et al. (2010) and Reddy et al. (2011) involved surveys that were used to characterize injury profiles in an individual production situation (a set of factors including cultural practices, weather condition, socioeconomics, *etc.*) that determine agricultural production, and the injury profiles using nonparametric multivariate analysis such as cluster analysis, correspondence analysis, or multiple correspondence analysis. Their results led to the conclusions that injury profiles (the combination of disease and pest injury that may occur in a given farmer's field) were found co-occurrence patterns across sites, which are associated at regional scale. For example, stem rot, sheath blight, planthopper, and rice whorl maggot injuries, are high incidence, with low incidence of brown spot, and absence of bacterial leaf blight, leaf blast, and neck blast are a common pattern in tropical Asia from the study of Savary et al. (2000b).

Co-occurrence analysis and network theory have recently been used to reveal the patterns of co-occurrence between microorganisms in the complex environments ranging from human gut to ocean and soils (Faust et al., 2012; Ma et al., 2016). Co-occurrence patterns are ubiquitous and particularly important in understanding community structure, offering new insights into potential interaction network. Recent reviews of network based approaches reveal that these tools have demonstrated previously unseen co-occurrence patterns, such as strong non-random association, topology based analysis of large networks has been proven powerful for studying the characteristics

of co-occurrence pattern of the communities in ecological community (Williams et al., 2014; Barberán et al., 2012), or key actors in social networks (Crowston and Howison, 2006). Here, we significantly advance this study by providing a comprehensive understanding of the topological shifts of animal pest injury and disease co-occurrence networks at regional scale.

South and Southeast Asia represent big bowl of rice for the world population. Comparing the topological properties of the node associated with occurrence in the different countries and examining network level topological features can provide us with insight into variation in the co-occurrence patterns of rice injuries in different countries. This approach helps contextualize the animal pests – disease association by taking to account the complex network of potential association among animal pest and disease occurring in farmers's fields in theses countries. Specifically, we addressed the following questions: (i) How can the co-occurrence relationships of rice injuries be examined from the perspective of network analysis (ii) Which animal injuries and diseases are found commonly close co-occurrence patterns among other variables in order to target to control or monitor. To answer these questions, we performed crop health survey at the farmers's fields in two different seasons and five countries in South and Southeast Asia and implemented co-occurrence network analysis to examine the topological feature differences across countries. Our main objective was to characterize and better understand co-occurrence networks in the association of rice animal pests and diseases.

IV.0.2 Materials and Methods

Study sites, data collection

We surveyed farmers' fields in 5 different locations; Tamil Nadu, India (TMN); West Java; Indonesia (WJV), Laguna, Philippines (LAG); Suphanburi, Thailand (SPB) and Mekong river delta, Vietnam (MKD) for consecutive years (2009 - 2013). These survey sites are in the major irrigated lowland rice growing areas, where rice is intensively cropped at least two seasons per year. The 412 individual fields were surveyed in both dry and wet seasons of 2010 to 2012 in India, Indonesia, Thailand, and Vietnam, except Philippines, which the surveys were only conducted in 2011. The number of survey were summerize in Table ??.

The survey procedure and data were based on a standardized protocol described in "A survey portfolio to characterize yield-reducing factors in rice" developed by Savary and Castilla (2009). Rice injuries were collected in this study including the injuries caused by animal pests, and pathogens, which are harmful to rice plants, and importantly considered to reduce yield productivity. They were evaluated at booting and ripening stage according to survey procedure. Injuries on leaves such as whorl maggot injury (WM), leaffolder injury (LF), bacterial leaf blight (BLB), bacterial leaf streak (BLS), leaf blast (LB), brown spot (BS) were determined as a proportion of injured leaves. Injuries on tillers or hills such as stem rot (SR), sheath rot (SHR), sheath blight (SHB), whitehead (WH), deadheart (DH), gall midge injuries (GM) were determined as a proportion of injured tillers or panicles. Insect pests including brown planthopper (BPH), white backed planthopper (WPH), green leafhopper (GLH), rice bug (RB) or stink bug were determined as number of insect found on the rice hill.

Before analysis, data were compacted over time during crop growth. Two types of data were computed, depending on the nature of injury Savary and Castilla (2009). One is an area under injury progress curve (AUIPC) used for injury variables, which present on the leaves, and for weed infestation. Another is the maximum level at any of the two observations used for injury variables that can be observed on tillers, panicles, and hills, and insect pest count. The area under injury progress curve (AUIPC) (Campbell et al., 1990) were calculated by the mid-point method using the following equation:

$$AUIPC = \sum \frac{1}{2(X_i + X_{i-1})(T_i - T_{i-1})} \quad (IV.1)$$

where X_i is percentage (%) of leaves, tillers or panicles injured due to rice pests (e.g., leaf blast, leaf folder), or number of insects (e.g., plant hoppers, leaf hoppers) per quadrat, or percentage (%) of weed infestation (ground coverage) at the i th observation, T_i is time in rice development stage units (dsu) on a 0 to 100 scale (10: seedling, 20: tillering, 30: stem elongation, 40: booting, 50: heading, 60: flowering, 70: milk, 80: dough, 90: ripening, 100: fully mature) at the i th observation and n is total number of observations.

Network construction

The co-occurrence network was inferred based on the Spearman correlation matrix constructed with R function `cor.test` with parameter method ‘Spearman’ (package stats) was used for calculate Spearman’s correlation coefficient (ρ), which is defined as the Pearson correlation coefficient between the ranked variables R Core Team (2015). The nodes in this network represent injuries and the edges that connect these nodes rep-

resent correlations between injuries. Based on correlation coefficients and P -values for correlation, we constructed co-occurrence networks. The cutoff of P -values was 0.05. Network properties were calculated with the **igraph** package (Csardi and Nepusz, 2006). All farmers' fields were divided into groups by country and season. The impact of each sample group on the Spearman correlation value of each edge in the network was assessed by Spearman correlation value of these fields. The network of each of group was detected the community structures by maximizing the modularity measure over all possible partitions by using `cluster_optimal` function of **igraph** package Brandes et al. (2008).

Topological feature analysis

We calculated the topological features for each network with the *igraph* package. We measured two levels of network topologies. The node features that we focused on are node degree, betweenness, and clustering coefficient. Node degree is measured by the number of the edges (connections) of a node has. Betweenness of a node is defined by the number of shortest paths going through a node, and the local clustering coefficients of a node is the ratio of existing edges connecting a node's neighbors to each other to the maximum possible number of such edges.

Global features including network clustering coefficient, average path length, and diameter were measured for each network. The network clustering coefficient measures the degree to which nodes of the network tend to cluster together and is a measure of the connectedness of the network and is indicative of the degree of relationships in the network. Average path length is the average number of steps along the shortest paths for all possible pairs of network nodes, and diameter is the greatest distance between any pair of nodes. In our analysis, both diameter and average path length are considered

measures of the size of the network. Larger networks are less connected, meaning that the likelihood of a strong connection between any two randomly selected species is low. The network clustering coefficient are considered measures of the complexity of the network. The networks are more complex, the network has higher clustering coefficient, and shorter average path length.

Node were further classified by ranking all nodes according to three node features, partitioning this ranked list into three equally value of each node property. Nodes with high rank value in top third proportion of node degree, and high rank value in top third proportion of betweenness are recognized as indicator in co-occurrence network of rice injuries.

IV.0.3 Results

Crop health survey data

Prior to using the crop health survey data to construct networks, some exploratory plots of the data are presented and examined. The figure shows the distribution of the data. A total of 415 farmers' fields was surveyed in Tamil Nadu, India (TMN); West Java; Indonesia (WJV), Laguna, Philippines (LAG); Suphanburi, Thailand (SPB) and Mekong river delta, Vietnam (MKD) from 2009 to 2013. The data collected pertain to rice injuries caused by animal pests, and pathogens. The protocol gives more emphasis on the nature of injuries and not on the causal organism. The levels of injuries during a cropping season were computed or summarized according to the nature of injuries and thus varied in scale. For example, foliar injuries, diseases on leaves, and insect pests were summarized as area under the disease incidence progress curve, and tiller injuries and diseases as a maximum incidence.

The injuries caused by animal pests observed during the survey period were rat injury (RT), deadheart (DH) and whitehead (WH) caused by stem borers, whorl maggot injury (WM), leaf folder injury (LF), gall midge injury or silver shoot (GM). Rat injuries were observed at all 5 survey locations with low incidence (less than 20% incidence). We could observe 75% incidence of the rat injury in MKD in dry season. They were also observed at WJV in dry season, and both season in TMN, LAG, SPB. Gall midge injuries during survey period were not observed in TMN and LAG, but it was found in SPB, MKD, but WJV at 25 % incidence. Deadheart were observed all survey sites, and it was severe in dry season at SPB and MKD, but in WJV and LAG, it was severe in dry season. The trend of whitehead incidences observed was opposite the deadheart in-

cidence, which whitehead incidences were more severe in wet season at SPB and MKD, but less severe at WJV and LAG. Leaf folder injury was observed at all survey locations. The leaf folder incidences were more severe in wet season than the dry season at WJV, TMN, and LAG. As apposite to SPB and MKD, they were more severe in dry season than wet season. Whorl maggot injury was observed at all locations. Mostly, they were more severe in wet season than dry season at all surveyed locations, except LAG.

The diseases we recorded were bacterial leaf blight (BLB), bacterial leaf streak (BLS), brown spot (BS), leaf blast (LB), narrow brown spot (NBS), red stripe (RS), sheath blight (SHB), sheath rot (SR), false smut (FS), stem rot (SR). Rice diseases we observed in this study were commonly found at all locations, but there were some diseases that could not find especially in TMN such as BLS, BS, NBS, DP, RS, SHB, and SR. BLB was observed at all location. BLB incidence was higher in dry season than wet season in WJV and MKD except in TMN, LAG, SPB. Wet season was more favorable for BLS than dry season because the incidence was higher in wet season than dry season. As same as BLS, BS incidence was higher in wet season than dry season, and it was severe in SPB. Even though, LB is common disease in these survey locations, but there were some farmer's fields observed LB. In WJV and MKD, the LB incidence was higher in dry than wet season. There were many fields in SPB found high level of NBS incidence. Like BLS and BS, NBS incidence was more severe in wet season than dry season. There were many fields found high RS incidences. The highest incidences of DP were found in MKD. FS were commonly found at all location. The high incidents were observed in SPB, and MKD. NB were observed at all location. They were found many observations in TMN. SHB commonly was found at all location, and high incidence was in TMN, and LAG.

Brown planthoppers (BPH) were found all location of survey sites. During the survey period, they were found higher population in wet season than dry season in TMN and MKD, but there was some observation of BPH at other locations. White backed planthoppers (WPH) were commonly found at MKD in both of dry and wet season, and there are a few observations in WJV and LAG, but at TMN and SPB, they were not observed. Rice bug (RB) could be observed all survey location. They were highly found in dry season than wet season at WJV, LAG, and MKD. but in SPB, they were found only in wet season during the survey period. Green leafhoppers were observed all location. They were found in LAG higher than other locations.

A global network of microbial co-occurrence and mutual exclusion within and among body site niches of the human microbiome

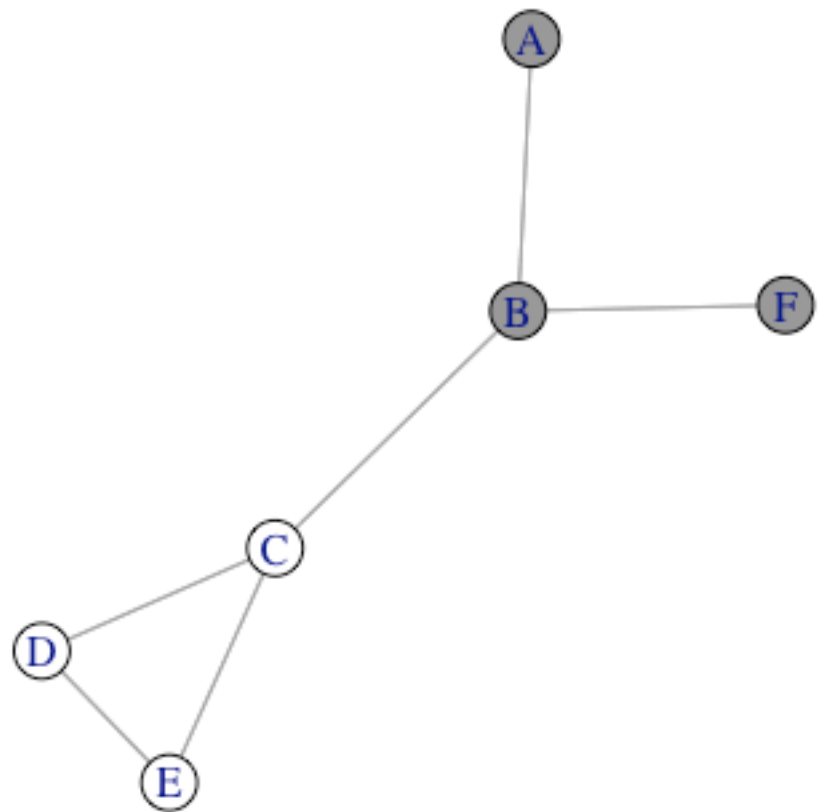
Co - (and anti) occurrence of injury profiles based on network analysis

Co-occurrence correlations of injuries were explored using network inference based on significant correlations using non-parametric Spearman's rank coefficient at $P < 0.05$. The setup of Spearman's coefficient cutoff at significant correlation coefficient at at $P < 0.05$ could efficiency reduce the sparse correlation and highlight the significant correlation between variables. The networks were determined for the co-occurrence analysis of rice injuries based on the survey data. Ten networks constructed were based on the survey data grouping different location (west java; Indonesia (WJA), Tamil Nadu; India (TMN), Laguna; Philippines (LAG), Suphan Buri; Thailand (SPB), and Mekong Rive Delta; Vietnam (MKD)) and seasons (dry, and wet season). Nodes represent rice injuries, and edges represent Spearman's correlation coefficients at P -value < 0.05 .

Once a network has been constructed, analytic tools and measures can be used to quantify by determining the structural properties.

Table IV.1: Node topology

Node	Node degree	Clustering coefficient	Betweenness
A	2	0	0
B	6	0	14
C	6	0.06	12
D	4	0.17	0
E	4	0.17	0
F	2	0	0



Measures of node properties of networks seek to identify the most important

nodes in a network. Different measures indicate the different contexts for the word “importance”. In this study, we consider 3 features of node properties.

- **Node degree** defined as the number of connections of a node. Nodes with high degree can be implied that the node has many connections or relationships with other nodes. In co-occurrence network, the high degree node can be inferred that it was found together with occurs with connected nodes. For example, in fig(), node B, and node C are the highest-degree nodes in this network. Node B has the relationships with 3 nodes. We can consider node B as the important node of this network because if we remove this node, there will be two nodes (node A and F) would disappear from this network.
- **Local clustering coefficient** is a measure of the degree to which nodes tend to cluster together. It is defined as how often a node forms a triangle with its direct neighbors, proportional to the number of potential triangles the relevant node can form with its direct neighbors. In the context of this network, local clustering coefficient is the probability that two neighbors of connected with each other in the network. These measures are indicative of the complex forming of co-occurrence patterns through the network. As presented nodes can be observed other nodes, a more closely connected network facilitates nodes co-occurrence. The higher clustering coefficient injury has high possibility to form complex. Node D and E have high clustering coefficient. They formed the complex relationships, which Node D connected with Node E connected to Node C, and Node C also connected with Node D. It indicated that they have close relationships. In the context of co-occurrence network, these three nodes usually occur together.
- **Betweenness** is measure the number of paths, which a node is present on the

shortest path between all other nodes. Nodes with high betweenness have been shown that there are many pair-wise relationships across a network pass through the node, which is implied that the nodes has high possibility to occur. We determine high-betweenness injuries as “indicator”.

Moreover, I inspected the community structure of the networks based on the optimal clustering algorithm. Such communities correspond to groups of injuries that are closely associated among themselves than other nodes in one others groups.

Communities, Structures and compositions of co-occurrence network of rice pest injuries

Tamil Nadu, India (TMN) Dry season network (Fig) was composed of 6 associated injuries (DH, NB, SR, FS, RB, and LF) and captured 7 associations. The network showed two groups of injury syndromes (the combination of injuries). The groups of the injury profiles corresponded to each community based on the optimal clustering algorithm. The first group is composed of DH, LF, RB and SR. Another group consists of FS and NB. Analysis of network properties revealed that LF and FS are high-betweenness nodes. They presented co-occurrence relationships within group 1 and group 2. SR and RB have high clustering coefficient. It indicated that these two injuries usually formed complex of co-occurrence relationships. As opposed to other injuries, NB and DH have low scores on all three centrality measure. Apparently, these injuries less possibly co-occur with other injuries (low betweenness), and do not have complex co-occurrences with other injuries (low degree and clustering coefficient).

Wet season network (Fig) was composed of 12 nodes (injury variables, DP SR, FS, BLB, NB, LF, GLH, RT, RB, DH, WM, BPH) with edges. Fig reveals three groups

of injury profiles. DH, GLH, SR and BPH are in the group 1 (green). FS, NB, DP, BLB is in group 2 (orange). RT, LF, RB and WM are in group 3 (purple). Top four injuries with high betweenness, DP, SR, LF, and BLB, are the members of each of group. They possibly are found co-occurrence within the group and inter-groups of injury profiles. Considered in each group, DH and GLH have high clustering coefficient in the group 1. NB has high clustering coefficient in the group 2. RT has highest clustering coefficient comparing other injuries in the group 3. Group 2 has high clustering coefficients indicating that this group is closed clustered, and the injuries in this group formed co-occurrence. DP and SR are important as the linkage to occur with other groups of injuries profiles because of high betweenness and degree. WM, BPH have low value of the 3 local properties. It indicated that BPH and WM were less possible to occur, and present co-occurrence patterns, and when they were observed, they were also not able to relate to many injuries.

West Java, Indonesia (WJV) Dry season network (Fig.) composed of 20 nodes and 51 associations. The network reveals four groups of injury profiles. Group 1 (green color) include DH and RT. Group 2 (orange color) is NBS RB, FS, BLS, LB, DP, BLB, NB, BS. Group 3 (purple color) included GM, LF, BPH, SR, SHB, GLH, RS. Group 4 (pink color) include WM and WH. The second and third group are formed closely clusters. RT in group 1, BLB and BS in the group 2, LF of group 3, and WM in group 4 are high-betweenness nodes with intermediate clustering coefficient. NBS, NB, RB, BLS, FS, DP of group 2 have high clustering coefficients. Compared to group 2, the injuries in group 3 have relatively smaller than. It indicated that the injuries in the group 2 are tightly formed complex co-occurrence. DH, WH have low value of the three features, and located far from the center of the network. BPH and WM were less

possible to occur, and present co-occurrence patterns, and when they were observed, they were also not able to relate to many injuries. connectivity.

Wet season network (Fig.) composed of 19 injuries (WM, LF, GLH, DH, BLS, WH, NBS, FS, DP, RS, LB, WPH, SR, SHR, SHB, NB, BS and BLB) with 25 associations. The network was loosely connected (low clustering coefficients). It reveals 3 connected groups and one isolated group of injury profiles. Group 1 (green) DH, WM, SHB, WH, GLH, LF, LB, and RS. Group 2 (orange) is composed of WPH, SR, GM, BLS and DP. Group 3 consisted of FS, NBS, SHB, BLB. Group 4, which is isolated, is composed of BS and NB. In group 1, WM is the injuries with high betweenness, and WH is the injury with high clustering coefficient. In group 2, BLS has high betweenness, and DP has high clustering coefficient. FS in group 3 node with high betweenness and high clustering coefficient. Group 1 appeared to form complex co-occurrence patterns because the average of clustering coefficient of injuries in this group are higher than other groups of injuries.

Central Luzon, Philippines (LAG) The dry season network reveal three clustered groups of injury profiles. Group 1 composed of WM, RB, NB, SHB, and FS. WM and RB in this group have high rank of betweenness. Group 2 consist of LB and DP. DP in this group connected with BPH (high betweenness) from group 3, which have GLH, BPH and BLB. Interestingly, SHB in group1, and GLH in group 3 have high clustering coefficients. NB, LB, FS, and BLB featured low in three of centrality.

Figure revealed co-occurrence network of injury profiles in dry season at Laguna, the Philippines. Network resulted four groups of injury profiles, which there are three connected and one isolated. Group 1 (green) has SHR, RS, RT, which is isolated. Group 2 (orange) has NB, RB, GLH, BLS, and DP. This group has more more complex

combination than others because the clustering coefficients of injuries in this group are higher. Group 3 (purple) has LB, SHB, LB. This group is between group 1 and group 4 (pink), which has NBS, LF, BS, and BLB. The position of SHB, LB and WM in the group with high betweenness, but they do not form the the complex combination of the injuries. They tend to link the co-occurrence of the first and the fourth group of injury profiles, which connected to NB in the first group and LF in the fourth group. So they are potentially good target to be monitored too. For example, when LF presented without the present of LB, SHB, WM, it is less likely NB would present, and other injuries in the first group would less likely to present neither.

Suphan Buri, Thailand (SUP) Dry season network shows 8 injuries (DH, BS, SR, SHB, NBS, SHR, GLH, and WH) showing 12 significant relationships. The network revealed three closed cluster of co-occurrence patterns of injuries. Group 1 (green) is composed of NBS, DH, BS, and SHB. Group 2 (orange) is SHB and GLH, and group 3 (purple) is SR and WH. Group 1 is more complex than other two groups because three out of four nodes in the groups presenting high clustering coefficient. NBS, DH, SHB, and SR seem to be clustered together. BS present high betweenness in group 1, which is associated with the other two groups. SHB has high moderate degree and high betweenness, whereas it has low betweenness. It can form complex association with the injuries in group 1 and group 2. WH and GLH have low betweenness and node degree. Apparently, it is less easily found with other injuries, do not tend to complex combination (low clustering coefficient), or less possible for expressing co-occurrence through the network (low betweenness).

Network of co-occurrence patterns of injury profiles wet season at Suphan Buri, Thailand revealed 18 associated injuries (DH, BS, SR, SHB, NBS, SHR, GLH, and

WH), and 79 associations. Network analysis resulted three closely clustered groups. Group 1 (green) composed of NBS, FS, RT, SHB, LB, WM, RS and RB. Group 2 (orange) consist of GLH, BS, GM, SR, BLB, BLS, LF and DP. Group 3 consisted of SHR and NB. The group 1 are bigger and 3 were relatively high clustering coefficients than other two groups of injuries. RB, NBS, LF, and BS were highly associated in wet season. Even though they were not in the same group, but group 1 and 2 were close, which is indicated by the positions in the network. Interestingly, SHR in group 3 has high betweenness, which is in-between the association of injuries of group 1 and group 3. So SHB is more likely to present in wet season, and form complex association with other injuries because it connected to the high clustering coefficient groups.

Makong Rive Delta, Vietnam (MKD) Co-occurrence network of injury profiles of dry season in Mekong Rive Delta, Vietnam presented 20 injuries (DH, BS, SR, SHB, NBS, SHR, GLH, and WH) with 61 associations. The network reveals the three groups of injury profiles. The group 1 (green) consisted of LB, DP, RB, DH, BS, and FS. The group 2 (orange) was composed of GLH, WPH, SR, WH, BPH, RS, WM. And the group 3 (purple) consisted of RT, NB, SHB, BLB, LF, and NBS. RB in the group 1 has high rank of betweenness and clustering coefficients. The betweenness of injuries in group 2 ranged low to intermediate, but their clustering coefficients are relatively high. This indicated that they are more likely to form complex association within group than between groups. BLB in group 3 and BS in group 1 have high rank of betweenness and they were associated. The average clustering coefficient of group 1 and 3 are smaller than the group 3. So group 1 and 3 are more likely to have chance to form association between the groups.

Co-occurrence network of injury profiles in wet season at Mekong River delta, Vietnam presented 21 injuries (DH, BS, SR, SHB, NBS, SHR, GLH, and WH), and 54 associations. From the structure of this network (Fig), it seems to have three clustered groups base on optimal clustering algorithm. The group 1 (green) is composed of NB, SR, BS, WPH, RS, WM, DH, GLH, FS, RT. The second group (orange) has BLB, BPH, LB, NBS, SHB, SHR, DP. The third group is the smallest groups, which has RB, LF and WH. The members within the first groups are relatively close following the layout, and have similar level of clustering coefficients. SHB and RB, RT, LB have high node degree and betweenness, which are inferred that they have possibility to occur in wet season because they shared many co-occurrence patterns with others. Even though, SHB have high level of betweenness and node degree, but intermediate clustering coefficient. It

connected to the high-betweenness injuries such as RB and RT, where are in different groups. SHB also acted like a “bridge”, which link the injuries of the group 1 and group 2.

IV.0.4 Discussion

Rice injuries were found commonly in South and South east Asia, but at different levels of incidence. Some injuries of this study is relatively low prevalence of areas that have been reported such as leaf blast, brown spot. It could be implied that these injuries strongly depended on locations or climatic conduction to develop, so they were not observed at all locations or seasons during survey were conducted. Another reason is that there are some factors such as the utility of resistance varieties in the farmer's fields surveyed, so we could observe those injuries at low level of incidence. The similar reasoning could explain to many injuries such as BLB, RB, NB, which widely occur in rice growing areas.

From the survey data of rice injuries observed in farmers' fields, I analyzed the interaction and build the network based on that data. The methods applied for building a co-occurrence network of rice injuries were adapted from ecological studies. Usually, relationships were assessed using Pearson correlation. However, the use of the Pearson correlation coefficient is problematic because it requires the variables are applied with similar measure, and the variable values are normally distributed. Additionally, Pearson correlation can only capture linear relationships. Due to the fact that the assumptions of Pearson correlation are not fit with the survey data. The alternative is provided by using Spearman's rank correlation coefficient, which is also widely used in ecological studies.

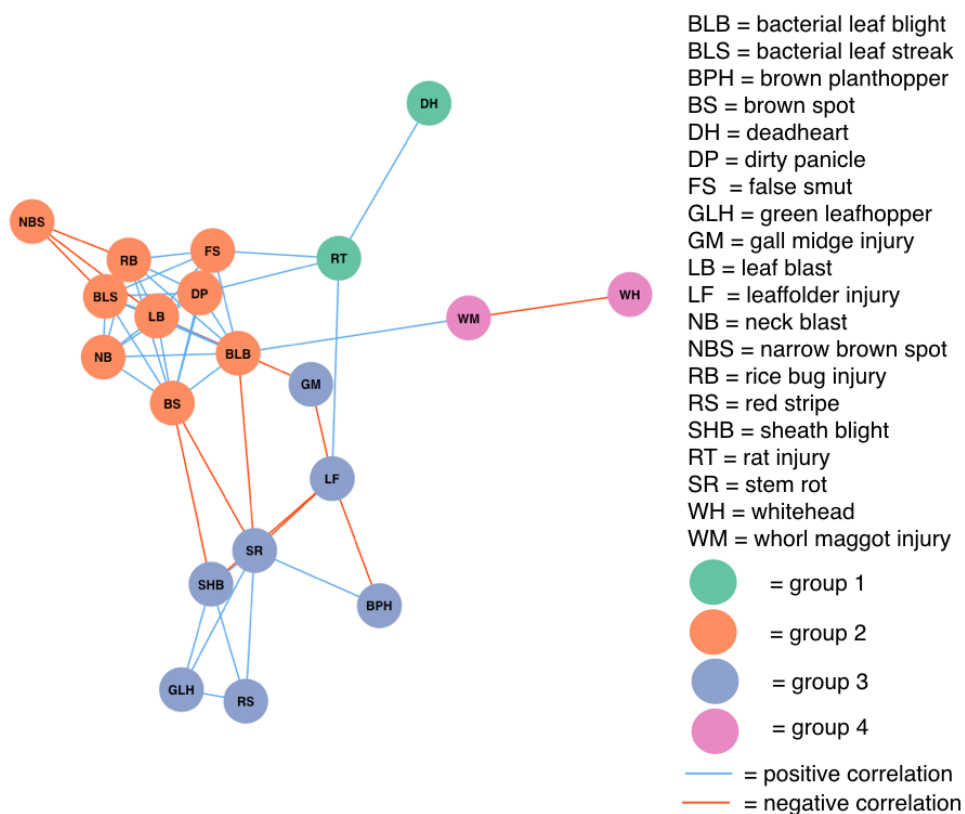
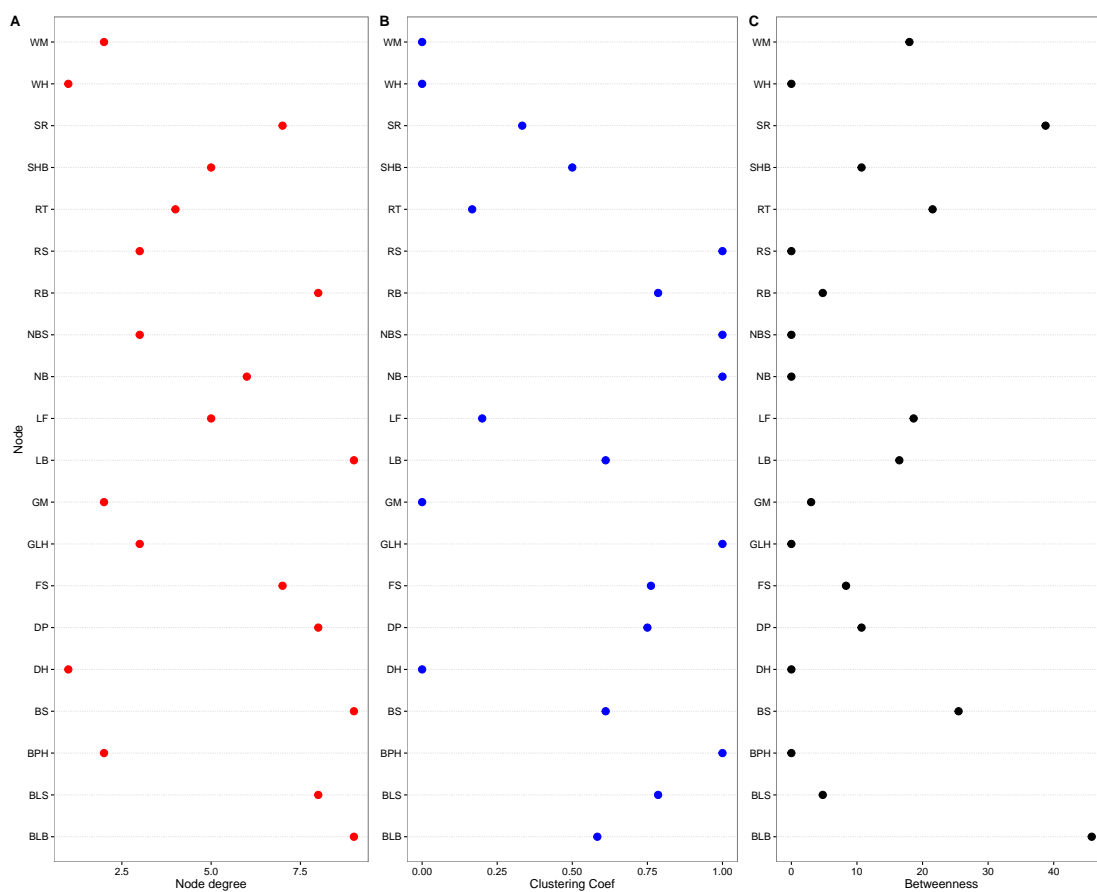


Figure 4-1: A picture of the same gull looking the other way!



The exploration of co-occurrence networks is a useful method for determining interactions of co- occurring injuries. Network analysis has also suggested important injuries in networks. The important injuries were selected from the node features such as node degree, clustering coefficient, and betweenness. The betweenness represents the importance of the control potential that an injury exerts over the associations of other injuries in the network. The clustering coefficients are indicative of the potential spreading of the incidence of injuries through the network. As activated injury can activate other injuries, a more densely connected network facilitates injury activation Williams et al. (2014). In the network of dry season in West Java, Indonesia, BLB and SR can be the targets to be monitored because they have high betweenness, which indicated that they are more likely to present than others injuries.

It is good attempt to detect communities in a network because it can reveal information about the networks that is maybe not easy to detect by simple observation. The communities are groups of nodes that are densely connected among their node members, and slightly connected with the rest of the network. In this study, we detected node community based on the optimization of the modularity of a sub-network, which is an approach that widely applied in many fields ?. Even though, the groups of injury profiles from this study are different from seasons and countries, but they are reasonable because some rice injuries present in some seasons (seasonal occurrence) such as gall midge injury ?. The result also showed similar groups of injuries to the patterns of injuries profile from the study of Savary et al. (2000b).

CHAPTER V

Differential networks reveal the dynamics of animal pests and disease co-occurrences conditioned by yield levels

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V.0.1 Abstract

The season network »»»»> origin/master

V.0.2 Introduction

Rice (*Oryza sativa*) is a major crop in South and Southeast Asian. Generally, rice farmers cultivate 2 rice crops per year, with the typical seasonal crop cycles or rotations being rice-rice-fallow or rice-rice-secondary crops (corn, soybean, peanut). The Food and Agriculture Organization of the United Nations (FAO) estimates that approximately 70 percent of total lowland rice area produces 2 rice crops each year. The first crop is cultivated in the wet season, while another is in the dry season. The important role of seasonal cropping in the temporal dynamics of animal pests and diseases has been studied under farmers field survey in South and Southeast Asia by the use of multivariate techniques Savary et al. (2000b); Willocquet et al. (2008). The previous studies

showed that Injuries profiles (the combination of injuries) differ from season to season in term of weather pattern. In the dry season, crop losses were lower than in the wet season. A previous study based on surveys done in farmers' rice fields in the region of lowland rice were shown to be strongly associated with injury profiles.

In the previous chapter, the co-occurrence networks to yield co-occurrence networks, a methodological approach which has already proved fruitful in a variety of different applications. Basically, plant injuries caused by pests maybe affect on yield production. Therefore in this chapter, I attempted to characterize the yield-reducing factors by studying the changes in the co-occurrence patterns of rice injuries (e.g disease incidence, animal pest injury incidence) at different yield levels.

Differential network analysis aims to compare the connectivity of two nodes at 2 different conditions. As demonstrated by several studies, differential networks can identify important nodes implicated in my fields, and also provide critical novel insights not obtainable using other approaches. In this work, I explore the the properties of network of a complex association of rice injuries at different yield levels. Elucidating the rice injuries association represents a key challenge, not only for achieving a deeper understanding of injury association (injury profiles) but also for identifying the unique association. Given that the injury association is governed by a complex network of injuries association, it seems natural to explore network properties which may help elucidate some of different association presenting in the different seasons.

I explored the relation between local differential association and discuss the meaning of the results in the context of the crop health management, and discuss the potential implications of our findings for development of pest management with a view to future studies.

V.0.3 Materials and Methods

I used survey data were collected from farmers' fields ,analyzed co-occurrence relationships of rice injuries, and performed network analysis as described previously. Actual yield estimates were collected from each farmers' fields surveyed. Before analyzing. Survey data were grouped into 6 groups by different yield levels and seasons.

Network Comparison Measures For the pair of x_i injury and y_i variable, I denoted the correlation coefficient based on Spearman's correlation coefficient by c_{xy}^1 and c_{xy}^2 in networks 1 and 2, respectively. To test whether the 2 correlation coefficients were significantly different of each network, I used Spearman's correlation coefficients, the p-values of the correlation test, the difference of the 2 correlations, the corresponding p-values, and the result of Fisher's z-test. First, I applied the Fisher z -transformation in order to stabilize variances due to sample size.

$$z_{xy} = \frac{1}{2} \log \left[\frac{1 + c_{xy}}{1 - c_{xy}} \right] \quad (V.1)$$

if we let z_{xy}^D and z_{xy}^W denote the z - transformation for dry and wet season variable pairs, respectively. Next, differences between the two correlations can be test using following.

$$\Delta z_{xy} = \frac{z_{xy}^D - z_{xy}^W}{\sqrt{\frac{1}{N_D-3} + \frac{1}{N_W-3}}} \quad (V.2)$$

N_D and N_W represent the sample size for each of season network for each country. The Z has an approximately Gaussian distribution under null hypotheses that the population correlations are equal. The pair-wise correlation significants are considered at $p\text{vlaue} < 0.05$.

More and more researchers realized that gene module is high related with disease, but not individual gene. In gene expression network, gene is only related with other genes. Based on the characteristic of no self-loop, the graph of gene coexpression network is a simple undirected graph, and the diagonal elements of gene coexpression matrix are all 0. The gene coexpression matrix is a square and symmetric matrix whose rows and columns correspond to the genes and whose element A_{ij} denotes the coexpression relationship between genes. The graph of maximum clique network is a complete graph that every pair of nodes is joined by edge, and the adjacency matrix elements of the complete graph are all 1 except the diagonal elements. For a simple undirected graph G containing N nodes, its adjacency matrix $A = (a_{ij})_{N \times N}$ contains only 1 and 0. It is a square and symmetric matrix obviously. $a_{ij} = 1$ represents that gene i and j is coexpressed, $a_{ij} = 0$ means that gene i and j is not connected. We set two thresholds $T1$ for adjacency matrix $A1$ in normal condition and $T2$ for adjacency matrix $A2$ in disease condition. $A1(i,j)$ set to 1 if the value of $A1(i,j)$ greater than or equal to $T1$, otherwise, $A1(i,j)$ set to 0. $A2(i,j)$ set to 1 if value of $A2(i,j)$ less than or equal to $T2$, otherwise, $A2(i,j)$ set to 0. We integrated $A1$ and $A2$ into a matrix A after we had intersection the corresponding elements of $A1$ and $A2$. $A(i,j) = 1$ means coexpression value of gene i and gene j in $A1$ greater than or equal to $T1$, and coexpression value of gene i and j in $A2$ less than or equal to $T2$. Equation 6 summarized the process. We excavated cliques which have biological significance from A adjacency matrix to further investigate gene regulatory networks.

V.0.4 Results

It is possible to construct other networks with the same data using a different criteria. It is interesting to see if there is a difference in the network structure dependent on different level of yield

Shared and unique co-occurrence and anti-co-occurrence in injuries at different yield level

V.0.5 Discussion

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