ENVIRONMENTAL CONDITIONS ASSOCIATED WITH STRIPE RUST AND LEAF RUST EPIDEMICS IN KANSAS WINTER WHEAT

by

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B.S., Arizona State University, 2006
M.S., Kansas State University, 2010

AN ABSTRACT OF A DISSERTATION

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Abstract

Stripe rust (caused by *Puccinia striiformis* f. sp. *tritici*) and leaf rust (caused by *Puccinia triticina*) are the top two diseases of winter wheat (*Triticum aestivum*) with a 20-year average yield loss of 4.9% in Kansas. Due to the significant yield losses caused by these diseases, the overall objective of this research was to identify environmental variables that favor stripe and leaf rust epidemics. The first objective was to verify the environmental conditions that favor *P. triticina* infections in an outdoor field environment. Wheat was inoculated with *P. triticina* and exposed to ambient weather conditions for 16 hours. Number of hours with temperature between 5 to 25°C and relative humidity >87% were highly correlated and predicted leaf rust infections with 89% accuracy. The results of this outdoor assay were used to develop variables to evaluate the association of environment with regional leaf rust epidemics.

Before regional disease models can be developed for a forecast system, suitable predictors need to be identified. Objectives two and three of this research were to identify environmental variables associated with leaf rust and stripe rust epidemics and to evaluate these predictors in models. Mean yield loss on susceptible varieties was estimated for nine Kansas crop reporting districts (CRD's). Monthly environmental variables were evaluated for association with stripe rust epidemics (>1% yield loss), leaf rust epidemics (>1% yield loss), severe stripe rust epidemics (>14% yield loss) and severe leaf rust epidemics (>7% yield loss) at the CRD scale. Stripe rust and leaf rust epidemics were both strongly associated with soil moisture conditions; however, the timing differed between these diseases. Stripe rust epidemics were associated with soil moisture in fall and winter, and leaf rust epidemics during winter and spring. Severe stripe rust and leaf rust epidemics were associated with favorable temperature (7 to 12°C) and temperature (15 to 20°C) with relative humidity (>87%) or precipitation in May

using tree-based methods of classification, respectively. The preliminary models developed in this research could be coupled with disease observations and varietal resistance information to advise growers about the need for foliar fungicides against these rusts in Kansas winter wheat.

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Dedication

To Gabe, Arlo and Maebe for their unconditional love and support.

Chapter 1 - Literature Review

Introduction

Wheat (*Triticum aestivum* L.) is the most widely grown cereal in the world and is a staple food for 35 percent of the world's population (IDRC 2016). Wheat is historically the most economically important crop in Kansas. In 2015, Kansas was the leading grower of winter wheat in the United States (U.S.) producing 321.9 million bushels (NASS 2015). The 2015 Kansas winter wheat crop was worth approximately 1.8 billion dollars (Price $5.80 * 321.9 million bushels).

Plant pathogens can significantly reduce yields in winter wheat. The twenty-year average for Kansas yield losses associated with pathogens of wheat is 8.49 percent (Appel et al. 2015). The most yield-limiting diseases of wheat are the foliar fungal pathogens leaf rust (*Puccinia triticina* Erikss.) and stripe rust (*Puccinia striiformis* f. sp. *tritici* Erikss.). The twenty-year average yield loss for these two diseases is 4.9 percent (Appel et al. 2015). The highest statewide yield losses were 15.4 percent and 13.9 percent for stripe rust and leaf rust, respectively (Appel et al. 2015). Yield losses from rust pathogens are attributed to the reduction of biomass, kernel weight, kernels per spike, and test weight (Chester 1946; Herrera-Foessel et al. 2006; Singh and Huerta-Espino 1997). Reductions in yield are more severe when the diseases are present before and after anthesis and into the grain filling growth stages (Burleigh et al. 1972; Chester 1946; Herrera-Foessel et al. 2006; Singh and Huerta-Espino 1997). While stripe rust and leaf rust represent fungi from the same genus, they require different environmental conditions for infection processes which will be discussed in the subsequent sections.

Leaf Rust

Wheat leaf rust occurs wherever wheat is grown and the center of origin is the Fertile Crescent region of the Middle East (Chester 1946; Samborski 1985). Leaf rust is an obligate parasite that requires a living host in order to produce an infection. Leaf rust is heteroecious and macrocyclic, producing five spore stages. The urediniospore, teliospore and basidiospore stages are produced on the primary host, wheat. The pycniospore and aeciospore stages are produced on the alternate hosts *Thalictrum speciosissimum*, *Isopyrum fumarioides*, *Anchusa* spp. and *Clematis* spp. (Samborski 1985). *Thalictrum speciosissimum* is considered to be the main alternate host with the other hosts only occurring in specific regions (Samborski 1985). The *Thalictrum* species native to North America are resistant to leaf rust (Kolmer et al. 2007; Saari et al. 1968); therefore, sexual recombination does not result in new races in North America (Kolmer et al. 2007). In the absence of sexual recombination, new races are the result of mutation and selection for virulence against leaf rust resistance genes (Kolmer 2005). When susceptible winter wheat varieties are widely grown, large populations of rust survive which increases the reservoir for mutation and selection for new rust races (Kolmer 2005). Approximately 50 to 60 races of leaf rust are found each year by the virulence survey conducted by the USDA Cereal Disease Laboratory (Kolmer et al. 2007).

The polycyclic cycle of urediniospores is responsible for the widespread epidemics experienced in the Great Plains (Eversmeyer and Kramer 1994). New infections can occur every 7 to 10 days depending on environment, host age and genotype (Bolton et al. 2008; Tomerlin et al. 1983). Urediniospores are wind dispersed from northern Mexico and southern Texas following the developing wheat crop in the spring (Roelfs et al. 1989). The movement of spores occurs from southern Texas north into Canada annually (Roelfs et al. 1989). Leaf rust can survive the hot, summer period between harvest and planting on volunteer wheat as pustules (uredinia) or mycelium (Eversmeyer and Kramer 1994). Leaf rust is not suspected to overwinter in Kansas in most years due to the wheat crop dormancy and cold temperatures that kill the leaves (Eversmeyer et al. 1988). Eversmeyer et al. (1988) investigated the survival of leaf rust inoculum from winter to early spring in Kansas (Eversmeyer et al. 1988). When leaf rust overwinters, statewide yield losses can exceed 2% (Eversmeyer et al. 1988). Final leaf rust severity can be between 80 to 100% when infection occurs at the flag leaf to early dough stages (Burleigh et al. 1972; Eversmeyer and Kramer 1998).

Leaf rust produces round to ovoid pustules up to 1.5 mm in diameter on the adaxial and abaxial leaf surfaces (Bolton et al. 2008). The urediniospores are brown to orange, round to subgloboid and on average 20µm wide (Bolton et al. 2008). Approximately 20,000 spores can be produced per pustule when leaf rust infections occur at the heading growth stage through senescence (Tomerlin et al. 1983).

Leaf rust can infect over a wide range of temperatures depending on the infection process being investigated. The leaf rust urediniospore infection process involves six major steps: spore germination, germ tube formation, appressorium development, penetration into the substomatal space, growth and development of haustorial mother cell (HMC), and sporulation. Urediniospores imbibe water in order to begin germ tube development. Optimal spore germination occurs with temperatures between 15 to 20°C with continuous dew for 4 to 8 hours (Chester 1946; de Vallavieille-Pope et al. 1995; Hogg et al. 1969; Roelfs et al. 1992). However, urediniospore germination has been documented to occur between 2 to 30°C (Roelfs et al. 1992). Germination is severely limited at 30°C and no germination was observed at temperatures greater than 35°C (de Vallavieille-Pope et al. 1995). More time is required for the germination process when temperatures are less than 10°C (de Vallavieille-Pope et al. 1995). Low temperature thresholds are more difficult to evaluate as the fungus can survive as mycelium within the leaf (Chester 1946). Mycelium from a fall infection can overwinter in living leaf tissue and can resume sporulation when the temperatures increase and the plant breaks dormancy (Eversmeyer and Kramer 2000). After germination, the germ tube will move across the leaf surface to a stoma (Bolton et al. 2008). The optimum temperature for germ tube growth is 15 to 20°C (Chester 1946) with growth restricted at temperatures less than 5°C and greater than 31°C (Hogg et al. 1969). When a stoma is encountered, the germ tube protoplasm will concentrate at the hyphal tip and form an appressorium (Bolton et al. 2008). An appressorium is a swollen flattened portion of a fungal hypha that will adhere to the surface of a plant (Bolton et al. 2008; Hawksworth et al. 1995). A septum will form between the germ tube and the newly formed appressorium (Bolton et al. 2008). The optimal temperature for development of the appressorium is between 15 to 20°C (Chester 1946; Hogg et al. 1969). The stoma will close in response to the appressorium and a penetration peg will push through the closed stoma to enter the host substomatal space (Bolton et al. 2008). The penetration peg hypha forms a substomatal vesicle. Infection hyphae and HMC are developed from the substomatal vesicle when in contact with mesophyll cell (Bolton et al. 2008). The HMC penetrates the host cell wall forming a haustorium (Bolton et al. 2008). The HMC will produce more infection hyphae that will form new HMC and haustoria when in contact with other host cells (Bolton et al. 2008). The optimum temperature for penetration and growth within the host ranges from 15 to 25°C (Chester 1946; de Vallavieille-Pope et al. 1995; Hogg et al. 1969). At optimal temperatures, the latent period is approximately 7 to 10 days after inoculation (Bolton et al. 2008; Tomerlin et al. 1983). Rime et al. (2005) found longer latent periods on resistant cultivars than susceptible cultivars with environmental conditions constant. Resistant varieties with longer latent periods are considered slow rusting wheat. Slow rusting wheat generally has longer latent periods and fewer, smaller pustules compared to susceptible varieties (Kolmer 1996). The combination of the genetic resistance and environmental conditions could reduce the number of reproductive generations for leaf rust in a wheat growing season (Kolmer 1996).

Many experiments have been conducted evaluating the temperature and moisture conditions that are conducive for leaf rust infection and infection processes in controlled growth environments. The findings of these experiments varied depending on the isolate of leaf rust, temperature ranges used in the controlled environment and host genotype. Because of the variation in experimental conditions, many of the temperature ranges have been summarized to describe optimal temperatures for infection. Commonly summarized leaf rust infection ranges include temperatures between 5 to 25°C (de Vallavieille-Pope et al. 1995), 15 to 20°C (Chester 1946; Hogg et al. 1969), 10 to 25°C (Bolton et al. 2008), and 10 to 30°C (Roelfs et al. 1992).

Due to the severe yield losses that can occur from leaf rust on wheat, many researchers have attempted to model and predict leaf rust infections using environmental conditions that may limit yield in the field. The earliest forecast model in the U.S. was based on the concept of the "critical month" by K. Starr Chester in Oklahoma (Chester 1943). The critical month correlated leaf rust severity and environmental conditions starting in midwinter. The author found leaf rust severity and development was correlated with early spring (March) mean temperature greater than 10°C and precipitation. These conditions were associated with the future development of the fungus and the potential for increased number of generations for a severe epidemic. Building on the previous research, Eversmeyer and Kramer (1996) found winter and early spring variables representing temperature or the deviation from optimal temperatures explained most of the variation in overwintering of leaf rust in Kansas. Moisture conditions in July and September were also associated with leaf rust overwintering. These time periods correspond to the potential establishment of volunteer wheat in July and the start of planting and establishment of winter wheat in Kansas. The resulting “green bridge” is an important inoculum source for fall leaf rust infections. For the March prediction date, snow and temperature variables were highly associated with overwintering of leaf rust. The authors continued investigating the environmental conditions favorable for severe leaf rust epidemics using three time periods representing environmental conditions prior to planting, beginning of winter dormancy, and final tiller development growth stage for winter wheat in Kansas (Eversmeyer and Kramer 1994, 1998). Overwintering was associated with temperature and precipitation variables prior to planting. Warmer and wetter than average conditions and the presence of snow cover; which insulates infected wheat leaves from temperature fluctuations that would generally kill wheat tissue, favors overwintering of leaf rust at the beginning of winter dormancy. A model combining the environmental conditions favorable prior to planting and winter dormancy was highly associated with severe leaf rust epidemics at final tiller development.

In addition to the work in the U.S., environmental and cultural practices favorable for leaf rust disease development have been investigated in other wheat growing regions. Moschini and Perez (1999) evaluated the environmental variables associated with leaf rust severity by planting date in Argentina. Leaf rust severity at early planting (June to July) was associated with temperatures (accumulation of heat units), relative humidity (days with relative humidity greater than 70 percent), and cultivar resistance. When wheat was planted late (August), meteorological variables were not as predictive of leaf rust severity as the early-planted model. For the late planting date, cultivar resistance and temperature (total accumulated degree-day base mean temperature of 11°C) were the most predictive variables with final leaf rust severity. In the United Kingdom (U.K.), temperature (accumulated degree-days from planting), frost (days with minimum temperatures less than 0°C), and amount of inoculum could be used to predict disease severity prior to fungicide decisions (Audsley et al. 2005). In Luxembourg, El Jarroudi et al. (2014) found nightly temperatures (temperatures between 8 to 16°C), relative humidity (relative humidity greater than 60 percent), and precipitation (rainfall less than 1mm) were associated with leaf rust infections on the flag minus 3 leaves through early dough growth stages in the spring (El Jarroudi et al. 2014).

Stripe Rust

The center of origin for wheat stripe rust is most likely western China, the Caucasus, central Asia and eastern Africa (Ali et al. 2014; Jin et al. 2010). Like leaf rust, stripe rust is an obligate parasite that requires a living host in order to produce an infection. Stripe rust is heteroecious and macrocyclic, producing five spore stages. The urediniospore, teliospore, and basidiospore stages are produced on the primary host, wheat. The pycniospore and aeciospore stages are produced on the alternate hosts *Berberis chinensis, Berberis holstii, Berberis koreana*, and *Berberis vulgaris* (Jin et al. 2010). While the alternate hosts listed previously are not native to North America, there may be potential for ornamental *Berberis* species to act as an alternate host (Jin et al. 2010). Despite the presence of potential alternate hosts, stripe rust virulence surveys indicate that the races observed in the Great Plains are not the result of sexual recombination (Chen 2005). The virulence diversity observed in the Great Plains is from mutation within the stripe rust population (Chen et al. 1993; Hovmoller et al. 2011; Steele et al. 2001; Stubbs 1985). In 2013, 34 races of stripe rust were detected from a total of 417 stripe rust isolates in the U.S. and Ontario, Canada (www.striperust.wsu.edu/races/raceData/stripe-rust-race-summary_2013.doc).

Prior to 2000, stripe rust was commonly found west of the Rocky Mountains and rarely in the Great Plains (Chen et al. 2002). In 2000, a severe epidemic occurred in Arkansas and Louisiana (Chen 2005). The first major Kansas statewide yield loss of 7.3 percent was observed in 2001 (Chen et al. 2002; Cereal Disease Laboratory 2001). The change in virulence was attributed to new races that were more aggressive and better adapted to the warm temperatures experienced in the Great Plains (Milus et al. 2006). Milus et al. (2009) compared the post-2000 (new) isolates to the pre-2000 (old) isolates found in the Great Plains. The new isolates were more aggressive than the pre-2000 isolates with respect to shorter latent period, larger pustule size, increased sporulation per inoculation, and more spores produced per mm^2 at low (10 to 18°C) and high (12 to 28°C) temperature ranges. The urediniospore stage is polycyclic with a 9 to 14 day latent period depending on environment and host genotype (Chen 2005; Line 2002; Milus et al. 2006). Markell and Milus (2008) evaluated the phenotypic and genotypic variability of the old and new isolates. The new isolates were genetically different and likely the result of a new introduction and not from mutation (Markell and Milus 2008). Since 2000, the combination of the introduction and increased aggressiveness of the race at higher temperatures and susceptibility of the widely planted varieties resulted in the severe stripe rust epidemics experienced in the Great Plains (Markell and Milus 2008).

Stripe rust inoculum overwinters in the southern Great Plains and northern Mexico (Chen 2005; Sharma-Poudyal et al. 2013; Stubbs 1985). The environment in Kansas is considered generally unfavorable for stripe rust overwintering and the inoculum for infection is usually the result of long distance dispersal (Rapilly 1979; Sharma-Poudyal et al. 2013). The spread of stripe rust through the Great Plains is facilitated by the prevailing winds, temperature and storms that push northward in the spring following the developing wheat crop in the Great Plains and Canada (Chen 2005; Hovmoller et al. 2011; Rapilly 1979). The transport of viable rust spores from the southern region could be hindered by ultraviolet radiation exposure of the spores suspended in the atmosphere and the presence of high relative humidity causing the spores to clump together preventing long distance aerial transport (Rapilly 1979).

Research in other regions of the world suggests temperature and moisture are likely important variables influencing the development of stripe rust epidemics. Stripe rust follows the same infection process previously discussed in the leaf rust section except some isolates do not form an appressorium and stripe rust requires different environmental conditions for infection (Allen 1928). Stripe rust will germinate after 3 hours of leaf wetness with temperatures between 5 to 10°C (Rapilly 1979). However, stripe rust can germinate from -2.8 to 21.7°C (Rapilly 1979). With dew present, spore germination, germ tube growth, and appressorium development occurs between 2 to 15°C with an optimum temperature of 7°C (Rapilly 1979). At 6°C, de Vallavieille-Pope et al. (1995) found spore germination to begin after 2 hours of dew. After 6 hours of dew, 93 percent of the spores germinated (de Vallavieille-Pope et al. 1995). The optimal temperature range for maximum spore germination was 5 to 16°C and the maximum germination occurred with temperatures between 8 to 12°C and no germination occurred above 20°C (de Vallavieille-Pope et al. 1995). The optimum temperature for infection was between 5 to 12°C and no infections occurred above 15°C (de Vallavieille-Pope et al. 1995). Eddy (2009) evaluated the environmental conditions required for infection in an outdoor exposure assay. Eddy (2009) found infections to occur between 2 to 23°C when moisture was not limiting. Many experiments have evaluated the optimum ranges for infection and the results vary depending on the isolate and growing conditions. Therefore, many authors have summarized the temperature ranges evaluated for infection processes and developed general infection ranges for stripe rust including 5 to 12°C (de Vallavieille-Pope et al. 1995), 7 to 12°C (de Vallavieille-Pope et al. 1995; Rapilly 1979), 10 to 18°C (Milus et al. 2006; Milus et al. 2009), 12 to 28°C (Milus et al. 2006; Milus et al. 2009) and 2 to 23°C (Eddy 2009).

Many models have been developed to predict stripe rust severity. A review of this research suggests that temperature and moisture conditions in the fall and winter months influence stripe rust severity in many areas of the world. For example, in the Pacific Northwest (PNW) region of the U.S., temperature degree-days in the fall (October and November) and winter (December and January) and precipitation in the spring (April) are correlated with severe stripe rust epidemics (Coakley 1983; Coakley and Line 1984; Coakley et al. 1982, 1984, 1988). Recently, Sharma-Poudyal and Chen (2011) evaluated yield loss caused by stripe rust in the PNW. Stripe rust was predicted based on a series of models using sum of maximum daily temperatures and a temperature degree-day based variable in late winter (February) and number of days with precipitation in the winter (December through January). In eastern Australia, temperature and precipitation in the fall (April and May in Australia) are highly correlated with stripe rust epidemics (Park 1990). The impact of environmental conditions during winter months was also documented in Europe (Gladders et al. 2007; Papastamati and van den Bosch 2007; Te Beest et al. 2008; van den Berg and van den Bosch 2007). Te Beest et al. (2008) identified temperature (maximum temperature) from February to June as the most important variable for predicting stripe rust epidemics and disease severity in the U.K. The models developed by Papastamati and van den Bosch (2007) found temperature, dew point and light quality as important weather variables associated with stripe rust disease progress and epidemics. Van den

Berg and van den Bosch (2007) and Gladders et al. (2007) indicated that overwinter survival was associated with mild temperatures. Spore reproduction was reduced with high temperatures in the summer (van den Berg and van den Bosch 2007).

Research Objectives

Stripe rust and leaf rust of wheat are the most yield limiting diseases in Kansas and lead to significant yield losses worldwide. Despite the detailed research on these diseases, there are only a few analyses evaluating the weather factors influencing leaf rust and no research on the weather factors influencing stripe rust epidemics in Kansas. The objectives of the following research are to identify environmental conditions associated with regional stripe rust and leaf rust epidemics of wheat in Kansas and to develop parsimonious predictive models for the risk of yield loss caused by these diseases. The objectives of the first experiment are to identify environmental conditions that are favorable for leaf rust infection events in an outdoor environment and to develop simple predictive models (Chapter 2). The objective of the second experiment was to identify the local weather and soil moisture conditions associated with stripe rust epidemics in Kansas (Chapter 3). The third objective was to identify the local weather, soil moisture conditions, remote sensing and climate variables associated with leaf rust epidemics in Kansas (Chapter 4). Ultimately, the objective of this research is to provide the basis of developing decision tools for Kansas wheat producers to increase their productivity and profitability.

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Chapter 2 - Effect of temperature and moisture on infection of wheat by *Puccinia triticina* under field conditions in Kansas.

Abstract

Leaf rust (caused by *Puccinia triticina*) is an economically important wheat disease in Kansas. On average, Kansas experiences 2% annual yield loss; however, yield losses >13% in 2007 resulted in monetary losses of 272 million US dollars from this disease alone. A bioassay was used to identify weather conditions that were favorable for leaf rust infection in an outdoor field environment. The bioassay included inoculating potted seedlings with leaf rust and exposing the plants to an outdoor environment overnight (16 hours). During the nightly exposure period, two pots were misted with distilled water and then covered with a plastic bag to retain moisture, and two pots were exposed to ambient conditions. Temperature, relative humidity, leaf wetness, and precipitation were collected on-site. Following exposure, the plants were then placed in a growth chamber at 20°C and evaluated for disease severity after 13 days. The bioassay was repeated on 125 nights over 3 years representing both fall and spring growing seasons for winter wheat in Kansas. The misted treatment was used to determine the temperature range for infection when moisture was not limiting. Infections occurred at a wide range of temperatures (3 to 25°C), and were most frequent when more than 6 hours of leaf wetness occurred during exposure. Temperature and relative humidity combination, leaf wetness, and relative humidity variables were the most highly correlated variables considered in this analysis. Only 5.3 hours were required for a leaf rust infection event when temperature and humidity were favorable. The results from this analysis will be used to determine weather conditions that are associated with leaf rust epidemics at a regional scale and to develop preliminary regional prediction models for Kansas.

Introduction

Leaf rust of wheat (caused by *Puccinia triticina* Erikss.) is an economically important disease in Kansas. In Kansas, leaf rust causes on average 2.1% annual yield loss but can cause severe yield loss when conditions are favorable (Appel et al. 2015). In 2007, Kansas experienced 13.9% yield loss that cost Kansas wheat growers approximately 272 million U.S. dollars (Appel et al. 2015).

Leaf rust forms characteristic round, orange to brown uredinia that are commonly referred to as pustules on the upper and lower leaf surface (Bolton et al. 2008). Each pustule can produce over 20,000 urediniospores on a susceptible host (Tomerlin et al. 1983). Urediniospores are wind-dispersed from northern Mexico and southern Texas through the Great Plains region and into Canada annually (Eversmeyer and Kramer 2000). Leaf rust reduces the amount of green leaf area thereby decreasing the photosynthetic rate of the plant and reducing yield (Robert et al. 2005).

Given sufficient inoculum and a susceptible host, temperature and moisture are the limiting environmental factors for leaf rust disease development. Many researchers have focused on temperature range requirements for infection processes in controlled environments including 15 to 20°C (Browder 1971), 20 to 25°C (Chester 1946), 18 to 25°C (Asuyama 1935) and 5 to 25°C (de Vallavieille-Pope et al. 1995). Higher temperatures have been shown to decrease the latent period, increasing the amount of spore production over time (Eversmeyer and Kramer 2000). Tomerlin et al. (1983) found latent period was shortest at higher temperatures for all growth stages. In the seedling assay, temperatures between 21.1 to 26.7°C resulted in the shortest latent period (8.3 days). Latent period was shortest at 23.9°C (10.1 days) and 29.4°C (8.3 days) for heading and anthesis wheat growth stages, respectively. Urediniospore germination was negatively impacted at temperatures greater than 32.2°C.

Moisture is the other limiting factor for disease development (Huber and Gillespie 1992). Previous researchers exposed leaf rust spores to varying moisture conditions to determine optimum spore germination (Chester 1946). Eight hours of high relative humidity and free moisture were required for germ tube and appressorial development (Chester 1946). Fewer hours of free moisture may be required for spore germination when optimum temperatures are present. Four to 8 hours of leaf wetness at 20°C are the optimum conditions for spore germination (Bolton et al. 2008). de Vallavieille-Pope et al. (1995) found 92% of urediniospores germinated after 4 hours at 15°C with continuous dew. Germ tube development ceased if dew formation was interrupted during the first 4 hours of exposure (de Vallavieille-Pope et al. 1995). These interruptions to infection processes are more likely to occur at suboptimal temperatures when longer dew events (wetness events) are required (de Vallavieille-Pope et al. 1995).

Outdoor exposure assays have been conducted to determine the environmental conditions required for infection by the wheat tan spot pathogen (*Pyrenophora tritici-repentis* (Died.) Drechsler) and the leaf and glume blotch pathogen (*Parastagonospora nodorum* (Berk.) Quaedvlieg. Verkley & Crous) (Francl 1995). In these experiments, potted plants were exposed to an ambient field environment for 24 hours to determine the conditions favoring natural field infections (Francl 1995). Rainfall was an important variable and facilitated the splash dispersal of spores into the crop canopy (Francl 1995). A forecast system was developed based on the outdoor exposure bioassay (De Wolf and Francl 1997). De Wolf and Francl (1997) developed an artificial neural network (ANN) to predict tan spot of wheat. Tan spot infections were predicted with high accuracy using variables describing moisture conditions including precipitation, proportional relative humidity and leaf wetness. The ANN resulted in 99% accuracy on a developmental dataset and 87% accuracy on the validation dataset; however, when leaf wetness was excluded from the model, accuracy decreased to 81% on the validation dataset. Eddy (2009) found stripe rust (*Puccinia striiformis* f. sp. *tritici*) infections occurred with temperatures between 2 to 23°C when moisture was not limiting in an outdoor field environment. Exposure assays have also been implemented to determine dispersal distances of the Septoria leaf spot pathogen (*Septoria lycopersici* Speg.) on tomato (*Lycopersicon esculentum*) at a variety of distances from the trap plants (Ferrandino and Elmer 1996). Dispersal was positively correlated with rainfall amount and duration prior to the exposure of trap plants. The authors hypothesized that rainfall prior to exposure facilitated the production of cirrhus containing conidia from the pycnidia.

Temperature and moisture ranges for *P. triticina* pathogenicity have been developed in controlled environments removing much of the environmental variability that is often seen under field conditions. While controlled growth environments are important to develop infection ranges, it is also important to confirm the environmental conditions that are favorable for plant pathogen infection events in natural field conditions. Therefore, the objective of this research was to identify local weather conditions that are favorable for leaf rust infection events in an outdoor field environment and to develop predictive models for infection events in Kansas.

Methods

Wheat leaf rust spore increase

The wheat variety 'Jagger' was evaluated for susceptibility to 10 Kansas leaf rust isolates based on Stakman infection type classes (McIntosh et al. 1995). Race TPBQJ (virulences 1, 2a, 2c, 3, 9, 24, 26, B, 10, 28, 39/41), collected in 2010 from Hutchinson, Kansas, was selected based on the moderate susceptibility rating of 33+ indicating medium sized uredia with or without chlorosis. Two week old seedlings were inoculated with the TPBQJ isolate using a single pustule inoculation procedure. Leaf rust urediniospores from one pustule were suspended in Soltrol 170 light paraffin oil (Chevron Phillips Chemical Company, The Woodlands, TX) and applied using an atomizer (G-R Manufacturing Co., Manhattan, KS) with compressed air (137.9 kPa). Pots were rotated during the inoculation procedure to ensure coverage of the solution. The oil was allowed to evaporate at room temperature for approximately 20 minutes or until the oil was no longer visible. Plants were then placed in a mist chamber at 19 to 21°C without light for approximately 16 hours. Following the 16 hours of moisture, plants were placed into a controlled growth chamber for approximately 12 days at 20°C with 16 hours of light (95µmol m^{-2} s^{-1}) per day for disease development. Urediniospores from the resulting infections were collected in glass vials with a cyclone spore collector (G-R Manufacturing Co., Manhattan, KS) and a DeWalt vacuum (Model DC500, Baltimore, MD). Spores were dried at room temperature (20°C to 25°C) for 24 hours in an airtight container with desiccant packs. Dried inoculum was placed into 2 ml polypropylene Cryogenic Storage Vials (Fisherbrand, Thermo Fisher Scientific Inc., Pittsburgh, PA) and stored at -80°C.

Plant production

Thirty seeds of wheat variety Jagger were planted in a 10 cm × 10 cm × 8.9 cm pot with Metro-Mix 360 potting soil (Sun Gro Horticulture, Agawam, MA). Plants were grown in a controlled growth chamber at a constant temperature of 20°C with 16 hours of light (95µmol m^{-2} s^{-1}) per day. The plant growth regulant, Cycocel (active ingredient chlormequat (2-chloroethyl) trimethylammonium chloride, OHP, Inc, Mainland, PA), was applied at a rate of 0.2ml/100ml water per pot prior to emergence. Pots were fertilized with Miracle-Gro® All Purpose Plant Food

(Scotts, Marysville, OH) at 0.325g/100ml water per pot when the first leaf was fully emerged. Plants were watered in shallow plastic trays from below. This avoided leaf wetness and reduced the risk of other foliar diseases. Marathon 1% Granular Insecticide (active ingredient Imidacloprid, 1-[(6-Chloro-3-pyridinyl)methyl]-N-nitro-2-imidazolidinimine,OHP, Inc, Mainland, PA) was applied at 1.4 g per pot at planting to control aphids. Approximately 2 weeks after planting, plants were inoculated with leaf rust urediniospores.

Wheat leaf rust inoculation procedure

A cryogenic storage vial was removed from storage at -80°C and heat shocked to activate spores when inoculum was ready to use. To heat shock inoculum, spores were warmed in a 42°C water bath for 6 minutes in the vial. Each batch of spores was used within 3 days of removal from storage to avoid possible losses in spore viability.

Leaf rust urediniospores were suspended in Soltrol 170 light paraffin oil at a concentration of 10^6 spores per ml or 0.0111g/ml and applied with an atomizer at 137.9 kPa. Pots were rotated to ensure full coverage of inoculum and kept at room temperature until the leaves were dry.

Bioassay

Two-week old Jagger seedlings were grown and inoculated using the previously mentioned procedures and exposed to three treatments. For the first treatment, inoculated plants were misted with double distilled water, covered with a bag to ensure moisture was not limiting spore germination, and placed overnight in a controlled environment at 20°C for approximately 16 hours (6:00 p.m. to 10:00 a.m.). This control treatment ensured that the inoculum was viable and provided an estimate of the possible disease severity given conditions known to be highly favorable for infection. The second and third treatments were exposed to ambient field conditions at the Kansas State University Rocky Ford Experiment Station (Manhattan, KS). For the second treatment, hereafter designated “ambient”, inoculated plants were exposed to the naturally fluctuating ambient temperatures and moisture conditions. The third treatment, hereafter designated “mist”, consisted of inoculated plants that were misted with double distilled water and covered with a bag, thus ensuring that moisture was not a limiting factor. Ambient and mist experimental units were placed in the field with the pots submerged at ground level for approximately 16 hours (6:00 p.m. to 10:00 a.m.). Bags were removed from the control and misted treatments after the 16 hour exposure and all treatments were removed from the field and incubated in the growth chamber at 20°C with 16 hours of light (95μmol m^{-2} s^{-1}) per day. Thirteen days after inoculation, 20 leaves were rated for percent severity (0-100%) per pot with 2 pots per treatment (40 leaves per treatment).

Weather conditions

Weather conditions at the Rocky Ford Experiment Station (Manhattan, Kansas) were collected using a Campbell data logger (Model CR-10X, Logan, UT). The weather station recorded ambient temperature (Model CS215, Logan, UT), temperature inside of the bag used to maintain the mist treatment (Model CS215, Logan, UT), relative humidity (Model HMP45AC, Vaisala, Vantaa, Finland), leaf wetness (Decagon Devices Model LWS, Pullman, WA), and precipitation (Model TR-525I, Texas Electronics, INC, Dallas, TX). The weather station recorded observations every minute, and these data were used to create hourly summaries of temperature, moisture, and temperature and moisture combinations.

Data analysis

The bioassay was conducted in Fall 2011 (22 days), Spring 2012 (53 days), Fall 2012 (42 days), and Spring 2013 (8 days). Disease incidence and average severity per treatment were summarized daily and paired with the weather data collected during the exposure period. A leaf contributed to incidence when the ambient treatment had leaf rust severity greater than or equal to 2%. A case was classified as an infection event if incidence was greater than 10%. Sixteen exposure days where the control had less than 1% disease severity or the plants were killed from hot or cold temperatures were removed from the dataset. Variables were created from the information gained from the data of the bioassay as well as known temperature ranges from the literature. Weather variables included four temperatures ranges: 5 to 25°C (de Vallavieille-Pope et al. 1995), 18 to 25°C (Asuyama 1939), 15 to 20°C (Browder 1971), and 20 to 25°C (Chester 1946); relative humidity greater than or equal to 80%, 87%, and 90% (Chester 1946; Rowlandson et al. 2015), leaf wetness duration (Chester 1946; de Vallavieille-Pope et al. 1995), precipitation (mm) and hours with precipitation (Chester 1946). Combination variables were created to evaluate the accumulation of time with temperature within a particular range and relative humidity greater than or equal to 87%. For example, the variable T0525RH87 describes the accumulation of hours where temperatures were between 5 to 25°C and relative humidity was greater than or equal to 87% during the exposure period. Variables were also developed with information gained from the bioassay. A total of 17 variables were evaluated in this analysis (Table 2.1).

The relationship between the binary response variable and the predictor variables was evaluated with the non-parametric Kendall Tau rank correlation coefficient with the Multivariate Platform in JMP (JMP®, Version *11*. SAS Institute Inc., Cary, NC, 2014). Variables with a Kendall Tau rank correlation coefficient greater than 0.40 were used to identify variables potentially associated with infection cases. The variables selected from the correlation analysis were evaluated as potential predictors in a logistic model framework. The potential single variable models were developed using Proc Logistic in SAS (SAS Institute, Version 9.4, Cary, NC). Models were evaluated based on the Akaike Information Criterion (AIC) and Receiver Operator Curve (ROC). The AIC evaluates model performance and was used to compare different models. The model with the lowest value suggests that the model performs the best and will provide the highest accuracy compared to the other models. The predictive ability of the variables was evaluated using ROC. ROC is sensitivity versus 1-specificity; sensitivity is the percentage of infection cases correctly predicted and specificity is the percentage of non-infection cases correctly predicted. ROC results in a value between 1 and 0 with the value closest to 1 indicating high performance of the variable. A value of 0.5 has no relationship and the predictive performance would be the same as random chance. As the predictive performance increases the accuracy also increases. Predictive accuracy of the models was evaluated using k-fold cross validation. The model dataset was randomly divided into 5 validation datasets and the misclassification rate was calculated for the sum of the 5 datasets. Equation (2.1) was used to calculate the predicted probability of each case. The classification table in Proc Logistic was used to determine the predicted probability threshold (p*) for classifying an infection event (Equation 2.2 & 2.3). The predicted probability threshold was chosen by balancing sensitivity and specificity.

EQ 2.1 $\mathbf{Predicted\ probability} = \frac{(\mathbf{EXP}(\mathbf{b}_{\boldsymbol{o}}+(\mathbf{b}_{\mathbf{1}}*\mathbf{x}_{\mathbf{1}})))}{(\mathbf{1}+(\mathbf{EXP}(\mathbf{b}_{\boldsymbol{o}}+(\mathbf{b}_{\mathbf{1}}*\mathbf{x}_{\mathbf{1}}))}$

EQ 2.2 $\mathbf{P}^{*}(\mathbf{probability\ threshold}) = \mathbf{ln}\,\frac{\mathbf{p}}{\mathbf{1-p}}$

EQ 2.3 $\mathbf{Threshold\ value} = \frac{\mathbf{p}*-\mathbf{b}_{\boldsymbol{o}}}{\mathbf{b}_{\mathbf{1}}}$

For equations 2.1 through 2.3, b_o is the intercept, b_1 is the parameter estimate, x_1 is the value for the parameter for a specific case, p is the probability value that balances specificity and sensitivity, and P* is the predicted probability threshold.

Results

The bioassay resulted in a total of 125 cases with 31 infection events and 94 non-infection events. Two infection events occurred in Fall 2011, 11 in Fall 2012, and 18 in Spring 2012. No infection events occurred during the 8 sampling days in Spring 2013. Average air temperature for infection events in the ambient treatment was 16.0°C with mean temperature ranging from 10.5 to 26.0°C. The average air temperature for non-infection events in the ambient treatment was 13.0°C with mean temperature range of -3.5 to 30.0°C. In the mist treatment, air temperature ranged from -4.0 to 30.2°C for non-infection events. In the mist treatment infection events, temperature ranged on average from 9.8 to 27.1°C. In most cases mist and ambient treatments both had infection with the exception of 2 cases in June 2012. The cases had infection in the ambient treatment and no infection in the misted treatment. The difference in infection was likely due to the mist treatment having temperatures greater than 39.0°C while the ambient treatment temperatures were below 37.0°C. While these temperatures are well above the optimum conditions for both wheat and leaf rust development, this could explain the difference in infection. Only 8 of the 31 infection cases had measurable precipitation during the exposure period. Low amounts of precipitation over short time periods occurred for 36 non-infection cases. Every infection event had periods of leaf wetness with a range of 2.0 to 15.0 hours. For non-infection events the mean leaf wetness was 6.2 hours suggesting that while leaf wetness was present for infection, temperature or other factors during the exposure period likely limited infection.

Variable selection

Correlation analysis identified hours with temperatures between 5 to 25°C and relative humidity greater than or equal to 87% (T0525RH87), leaf wetness duration (LWD), temperatures between 15 to 20°C and relative humidity greater than or equal to 87% (T1520RH87) and relative humidity (RH90, RH87 and RH80) as the variables with the highest correlation to infection events (Table 2.2). When moisture was not a limiting factor (mist treatment), infection occurred between 5 to 25°C (T0525) (Fig. 2.1). The combination variable T0525RH87 resulted in a stronger correlation with the response variable than RH87 and T0525 independently. All three relative humidity predictors RH87, RH90 and RH80 had similar correlation with the binary response variable. This is expected given the similarity of these variables.

Model fit

Variable T0525RH87 had a low AIC indicating that this model likely had a better fit than the other variables considered in the analysis (Table 2.3). T0525RH87 resulted in a ROC of 0.96 indicating that this variable can highly predict infection cases from non-infection cases. RH87 and T0525 had AIC values of 106.6 and 117.2, respectively. This suggests that the model fit improved with the combined variable when compared to the variables separately. An improvement of 10 or more in the AIC is considered to be markedly improved and increases the strength of evidence that the lowest AIC model is better fit when comparing models (Burnham and Anderson 2004). The ROC of T0525RH87 improved by 0.11 and 0.21 compared to RH87 and T0525, respectively. T0525RH87 had 88.8% accuracy when testing the model using the k-fold cross validation method. T0525 had 68.0% accuracy while RH87 had a 77.6% accuracy indicating that RH87 was the better single predictor variable. The predicted probability threshold for T0525 resulted in 14.5 hours of temperatures between 5 to 25°C required for an infection event. The predicted probability threshold for RH87 was 7.4 hours of relative humidity greater than 87% for an infection event. T0525RH87 had a predicted probability threshold of 5.3 hours. T1520RH87 had the largest AIC and lowest ROC of the variables considered in the analysis. While T1520RH87 had the poorest fit, the variable resulted in 81.6% accuracy. The poor fit was likely from 51.6% of the infection cases misclassified (false negatives). LWD had the second lowest AIC value and second highest ROC. The predictive probability threshold was 6.3 hours of leaf wetness for an infection to occur. Many of the infection events had one long period of leaf wetness however some of the cases had interrupted periods of leaf wetness. The cases with interrupted leaf wetness generally had a moist period soon after the start of the exposure period.

Discussion

The weather conditions that favor leaf rust infections had not previously been explored in a field environment. This analysis explored the weather conditions that favor leaf rust infections over the course of 125 exposure periods during two falls and two springs. The most significant variable identified from the analysis was T0525RH87. This variable combined the accumulated hours of temperatures between 5 to 25°C and relative humidity greater than or equal to 87%. Only 5.3 hours of favorable temperature and moisture conditions are required for an infection event based on the predicted probability threshold. This information suggests that when temperature and moisture conditions are conducive, infections can occur with fewer hours at optimal conditions than with moisture or temperature independently.

Measurements of leaf wetness were highly correlated with infection events. Relative humidity and leaf wetness variables were more strongly correlated with infection events than the temperature ranges considered in this analysis (Table 2.2). RH87 required 1 additional hour than the LWD model based on the predicted probability thresholds chosen for the analysis (Table 2.3). In a true field environment, the duration of RH87 may be higher in an overnight period due to the planting population, narrow rows, and canopy closure that increases leaf wetness as well as relative humidity in the canopy. Precipitation was poorly correlated with ambient treatment leaf rust infection events (Table 2.2). In other pathosystems, relative humidity and leaf wetness have been identified to be more associated with infections than precipitation alone (De Wolf et al. 2003; Eddy 2010; Paul and Munkvold 2005; Payne and Smith 2012; Wilks and Shen 1991). While leaf wetness was an important variable for predicting leaf rust infections in experiments reported here, the sensors may not be useful in regional prediction systems due to the lack of sensors at airports and variability among sensors (Bourke 1970; Giesler et al. 1996; Huber and Gillespie 1992; Rao et al. 1998; Kim et al. 2004; Magarey et al. 2001; Rowlandson et al. 2015; Wilks and Shen 1991; Windels et al. 1998).

The disease development temperature ranges were more weakly correlated with infection events than other variables. For example, T0525 alone fit more poorly when compared to RH87 and the combination variable T0525RH87. When moisture was not a limiting factor, infections occurred between 3 to 25°C (Fig. 2.1). de Vallavieille-Pope et al. (1995) also found leaf rust infections occurred within this similar temperature range in a controlled growth environment but, until this current study, their results had not been corroborated under field conditions. In the mist treatment, temperatures greater than 25°C restricted infection events. Other researchers have also found restrictive temperatures to inhibit infection processes and disease development (Chester 1946; de Vallavieille-Pope et al. 1995; Hogg et al. 1969; Tomerlin et al. 1983).

The results of this study indicate that while infection occurs in a particular temperature range, the presence of leaf wetness may be a dominant environmental condition required for infection. Relative humidity greater than 87% (RH87) was a stronger predictor than T0525 and resulted in two less false negative events. The combined model (T0525RH87) resulted in the best-fit model with the fewest number of false negative cases when balancing sensitivity and specificity.

Local weather conditions identified from this field exposure study confirmed previously reported leaf rust infection requirements for temperature and moisture under controlled growth environments (de Vallavieille-Pope et al. 1995). Combining temperature and relative humidity into a single predictor creates a parsimonious model while still obtaining the necessary information on the weather conditions that favor infection events. The individual variables that correlated with infection events could be applied to a risk analysis study of epidemics in Kansas. These variables could be coupled with regional and local disease observations as well as genetic resistance to determine the risk of leaf rust infection in Kansas.

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Figures and Tables

Table 2.1 Environmental variables descriptions.

Variable[a]	Variable Description
T0525RH87	Number of hours with temperatures between 5 to 25°C and relative humidity greater than 87%
LWD	Number of hours with leaf wetness
T1520RH87	Number of hours with temperatures between 15 to 20°C and relative humidity greater than 87%
RH90	Number of hours with relative humidity greater than 90%
RH87	Number of hours with relative humidity greater than 87%
RH80	Number of hours with relative humidity greater than 80%
RHAVG	Average relative humidity (%)
T1520	Number of hours with temperatures between 15 to 20°C
T1825RH87	Number of hours with temperatures between 18 to 25°C and relative humidity greater than 87%
T0525	Number of hours with temperatures between 5 to 25°C
T2025RH87	Number of hours with temperatures between 20 to 25°C and relative humidity greater than 87%
TBAG	Average air temperature inside misted treatment (°C)
T1825	Number of hours with temperatures between 18 to 25°C
TAVG	Average air temperature (°C)
T2025	Number of hours with temperatures between 20 to 25°C
PPT	Total of precipitation (mm)
PPTD	Number of hours with precipitation (mm)

[a] Acronyms describing the variables include temperature (T), relative humidity (RH), leaf wetness duration (LWD), average air temperature (TAVG), average relative humidity (RHAVG), precipitation (PPT), and precipitation duration (PPTD). Variable combining environmental conditions will have T and RH (e.g. number of hours with temperatures between 5 and 25°C and relative humidity greater than 87%).

Table 2.2 Correlation with binary infection events using Kendall Tau rank correlation coefficient.

Variable[a]	Kendall[b]	p-value
T0525RH87	0.63911	<0.0001
LWD	0.51025	<0.0001
T1520RH87	0.48490	<0.0001
RH90	0.46158	<0.0001
RH87	0.45223	<0.0001
RH80	0.42627	<0.0001
RHAVG	0.37961	<0.0001
T1520	0.37796	<0.0001
T1825RH87	0.35460	<0.0001
T0525	0.32470	<0.0001
T2025RH87	0.21556	0.0143
TBAG	0.18139	0.0137
T1825	0.18129	0.0174
TAVG	0.16245	0.0273
T2025	0.11796	0.1274
PPT	0.11245	0.1902
PPTD	0.10997	0.2002

[a] See table 1.

[b] Kendall Tau rank correlation coefficient.

Table 2.3 Model fit and performance of the logistic regression to predict leaf rust infections.

Variable[a]	AIC [b]	ROC [c]	P* [d]	Threshold (Hours) [e]	True Positive [f]	True Negative	False Positive	False Negative	Misclassification Rate (%)[g]
T0525RH87	61.0	0.96	0.30	5.3	26	85	9	5	11.2
RH87	106.6	0.85	0.30	7.4	24	73	21	7	22.4
T0525	117.2	0.75	0.34	14.5	22	63	31	9	32.0
LWD	100.1	0.88	0.28	6.3	24	73	21	7	22.4
T1520RH87	120.0	0.75	0.22	0.93	15	87	7	16	18.4

[a] See Table 1.

[b] The AIC statistic is used to compare different logistic regression models. Models with lower values are preferred.

[c] The ROC is used to evaluate predictive performance. Models with values closest to 1.0 are preferred.

[d] The cut-point for converting model-generated probabilities to classification of an observation as an infection event or non-infection event was based on balancing the sensitivity and specificity of the model.

[e] The model threshold is determined from equations B and C.

[f] There were 125 observations total. TP = true positives (number of infection events correctly classified) (31 cases); TN = true negatives (number of non-infection events correctly classified) (94 cases); FP = false positives (number of non-infection events incorrectly classified as epidemics); FN = false negatives (number of infection events incorrectly classified as non-epidemics).

[g] The percentage of the total observations that were incorrectly classified.

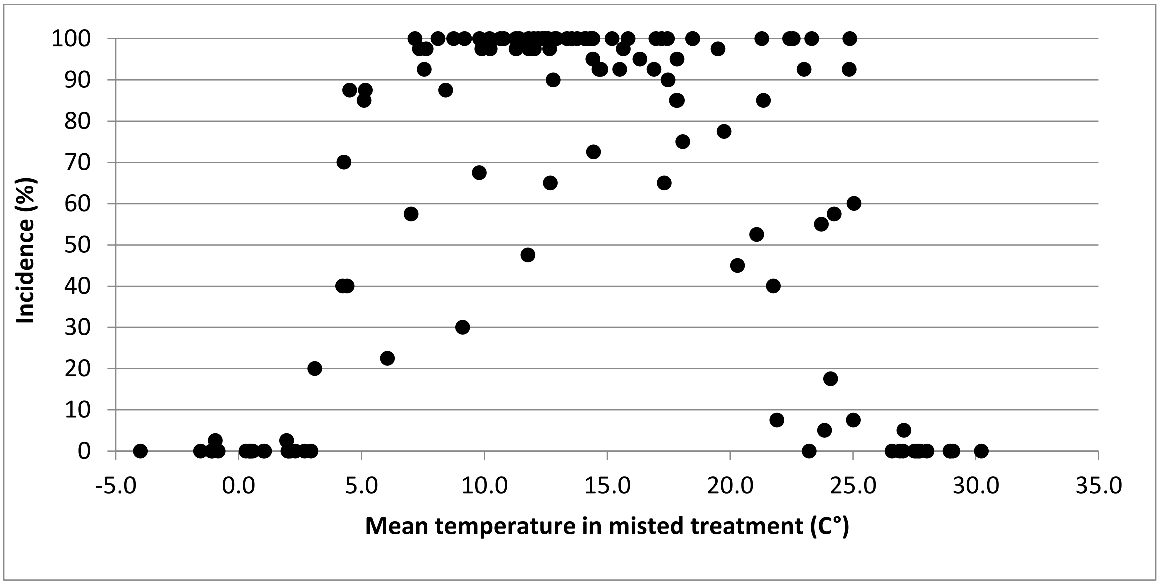

Figure 2.1 Incidence of leaf rust infection on bioassay plants following exposure in a field environment for 16 hours. Plants were misted with double distilled water and covered with a plastic bag to ensure moisture was not a limiting factor.

Chapter 3 - Environmental conditions associated with stripe rust in Kansas winter wheat

Abstract

Stripe rust has re-emerged as a problematic disease in Kansas wheat. However, there are no stripe rust forecasting models specific to Kansas wheat production. Our objective was to identify environmental variables associated with stripe rust epidemics in Kansas winter wheat, as an initial step in the longer-term goal of developing predictive models for stripe rust to be used within the state. Mean yield loss due to stripe rust on susceptible varieties was estimated from 1999 to 2012 for each of the nine Kansas crop reporting districts (CRDs). A CRD was classified as having experienced a stripe rust epidemic when yield loss due to the disease equaled or exceeded 1% and a non-epidemic otherwise. Epidemics were further classified as having been moderate or severe if yield loss was 1 to 14%, or greater than 14%, respectively. The binary epidemic categorizations were linked to a matrix of 847 variables representing monthly meteorological and soil moisture conditions. Classification trees were used to select variables associated with stripe rust epidemic occurrence and severity (conditional on an epidemic having occurred). Selected variables were evaluated as predictors of stripe rust epidemics within a general estimation equations framework. The occurrence of epidemics within CRDs was linked to soil moisture during the fall and winter months. In the spring, severe epidemics were linked to optimal (7 to 12°C) temperatures. Simple environmentally-based stripe rust models at the CRD level may be combined with field-level disease observations and an understanding of varietal reaction to stripe rust as part of an operational disease forecasting system in Kansas.

Introduction

Stripe rust, caused by *Puccinia striiformis* f. sp. *tritici* Erikss., re-emerged between 1999 and 2001 as a yield-limiting disease of wheat (*Triticum aestivum* L.) in the Great Plains region and Southern U.S. (Chen 2005; Chen et al. 2002; Chen et al. 2010; Line 2002). *P. striiformis* overwinters in the southern U.S.; urediniospores are wind-dispersed into the central Great Plains during spring (Sharma-Poudyal et al. 2013). Epidemics have become more frequent in Kansas since 2001; prior to that year, stripe rust had little impact in the state (Eversmeyer and Kramer 2000). From 2001 to 2012 average statewide yield loss due to stripe rust exceeded 1% in 5 out of 12 years. A 1% statewide yield loss translates to over 10 million U.S. dollars lost in grain production alone. Feedback from wheat producers in addition to results from foliar fungicide research plots indicate that yield losses can exceed 40% in individual fields (DeWolf, *unpublished*). These more recent stripe rust epidemics are associated with evolution in the pathogen population, which has overcome the genetic resistance of many popular wheat varieties (Chen et al. 2002; Markell and Milus 2008).

Wheat producers have turned to foliar fungicides for managing stripe rust given the loss of effective genetic resistance in preferred varieties. When stripe rust levels are moderate to severe, there is a high likelihood that foliar fungicides are profitable. However when the local environment favors low disease, the application of a fungicide could result in a net financial loss depending on varietal resistance category, yield potential of the crop, product and application costs, and market value of the grain (Edwards et al. 2012; Wegulo et al. 2011; Willyerd et al. 2015). Therefore the decision to apply a fungicide, given the importance of stripe rust and the narrow profit margins of wheat production in Kansas, is not straightforward. Plant pathologists, crop consultants and growers would benefit from having models to help them evaluate the risk of stripe rust epidemics when making foliar fungicide decisions. No such models currently exist for stripe rust in Kansas.

Environmentally based predictors have been used in stripe rust models for other wheat production regions. At the regional scale, temperature and moisture during the fall and winter months influence stripe rust severity in several wheat-growing regions worldwide. In the U.S. Pacific Northwest (PNW), autumn (October to November) and winter (December and January) temperatures, as well as precipitation during the spring (April) are correlated with stripe rust severity (Coakley 1983; Coakley et al. 1982, 1984, 1988). Sharma-Poudyal and Chen (2011) built on the earlier work of Coakley and colleagues by directly addressing stripe rust yield loss based on a series of simple single-variable models including the sum of maximum daily temperatures in February, accumulated negative degree days based on daily maximum temperatures in February, and the number of days with measureable rainfall between December and January. In Australia, autumn (April and May) temperature and rainfall are highly correlated with stripe rust epidemics (Park 1990). The importance of environment during the winter months was also noted in Europe (Gladders et al. 2007; Papstamati and van den Bosch 2007; Te Beest et al. 2008; van den Berg and van den Bosch 2007).

At the field scale, stripe rust is favored by temperatures between 2 to 23°C and non-limiting moisture (Eddy 2009). Temperature and moisture effects on stripe rust infection and latency studied under controlled environments (Coakley et al. 1982; de Vallavieille-Pope et al. 1995; Hoggs et al. 1969; Milus et al. 2009) gave optimal temperatures between 5 to 12°C, 7 to 12°C, 10 to 15°C or 10 to 18°C. Epidemiological research in North America and Europe also suggested that temperatures greater than 18°C suppress stripe rust in the field (Coakley et al. 1988; Hoggs et al. 1969; Newton and Johnson 1936; Shaner and Powelson 1971; Te Beest et al.

2008; van den Berg and van den Bosch 2007). The variability among reported temperature ranges for stripe rust infection are likely the result of differences among isolates considered; in research methods, and the specific stage of the disease cycle investigated. The effect of temperature on the pathogen is further complicated by the concomitant effect of temperature on expression of resistance by wheat to stripe rust. At higher temperatures the stripe rust fungus is less fit, but at the same time the wheat plant also expresses adult plant resistance (Qayoum and Line 1985). Adult plant resistance occurs from jointing onwards (Qayoum and Line 1985).

Before any type of model can be developed for stripe rust in Kansas wheat, suitable predictors must be identified. As environmentally-based predictors were used successfully in stripe rust modeling in other regions, it is likely that environmental variables are related to stripe rust epidemics in Kansas. Our objective was to identify environmental variables associated with stripe rust epidemics at a regional scale in Kansas. This objective is part of a longer-term goal of creating stripe rust predictive models which can be used in conjunction with field-level disease scouting and a consideration of variety genetic resistance when supporting fungicide use decisions.

Methods

Stripe rust yield loss data

Stripe rust disease occurrence and levels on susceptible varieties from 1999 to 2012 were derived from Kansas Cooperative Plant Disease Survey Reports (http://agriculture.ks.gov/divisions-programs/plant-protect-weed-control/reports-and-publications; Bockus et al. 2001; Bockus et al. 2011). Average stripe rust severity (0 to 100%) on the flag leaf was estimated for each CRD from non-fungicide-treated variety performance trials, county demonstration plots, and on-farm research locations. A wheat yield loss model (Mundy 1973) was then applied to the stripe rust disease estimates to obtain yield loss estimates on susceptible varieties in each CRD (DeWolf et al. 2012).

Variables from meteorological data

Hourly temperature (°C), relative humidity [RH] (%) and precipitation (mm) time series within the nine Kansas CRDs were downloaded from the Kansas Weather Data library (http://mesonet.k-state.edu/). These data were collected by automated weather stations at experiment fields maintained by Kansas State University. Weather stations were selected based on their location relative to wheat producing areas within each CRD, the availability of hourly weather data and completeness of weather records for the years considered in the analysis (Fig. 3.1). Erroneous or missing observations were identified by examining descriptive statistics and plots of the hourly time series. Common errors included out of range values (i.e. RH>100%), or atypical observations given other available data such as a temperature observation of -20°C when all other observations during that time series were between 15 and 22°C. Missing records or erroneous values in one or two contiguous hours were averaged using the adjacent data values. If data were missing or erroneous for three or more contiguous hours, data were filled from the nearest hourly reporting weather station.

Calendar-month summaries of weather were constructed from the hourly time series from August (of the planting year for winter wheat) to the following July (harvest year). These included descriptive variables such as average temperature, average RH and total precipitation. Other variables were constructed to reflect known relationships (as described in the Introduction) between weather and stripe rust etiology by summarizing the number of hours a specified condition was satisfied per month (e.g. hours with temperatures between 5 and 12°C). Multiple favorable or optimal temperature ranges (i.e. 5 to 12°C, 7 to 12°C, 10 to 15°C, 10 to 18°C and 2 to 23°C) have been reported for stripe rust and are conditional on the component of the disease cycle (spore germination, infection, etc.) and adaptations within the *P. striiformis* population (Coakley et al. 1982; de Vallavieille-Pope et al. 1995; Eddy 2009; Hoggs et al. 1969; Milus et al. 2009). Although these temperature ranges are highly correlated by definition, we decided it was important to consider multiple ranges to avoid any *a priori* bias in variable selection. Variables describing the accumulation of hours with temperatures that could restrict the development of stripe rust (i.e. temperatures greater than 12°C, 18°C or 23°C) were also considered (Coakley et al. 1988; de Vallavieille-Pope et al. 1995; Newton and Johnson 1936; Shaner and Powelson 1971; Te Beest et al. 2008). Relative humidity variables counted the number of hours that RH was greater than 87% or 90%. Precipitation variables included the number of days with any measurable precipitation (precipitation greater than 0.25 mm) or total precipitation per month. The final class of variables summarized temperature, RH or precipitation conditions being met simultaneously (e.g. the number of hours that temperature was between 5 and 12°C and RH>87%).

Regional moisture indices

The Palmer Drought Severity Index (PDSI; Palmer 1965), Palmer's Moisture Anomaly Index (ZNDX; Palmer 1965) and the Standard Precipitation Index (SPI; McKee et al. 1993) quantify moisture conditions at a regional scale. These three indices are highly correlated, but differ in what moisture variables and time periods are considered. The Palmer indices are dimensionless representations of the current moisture supply relative to a standard (Palmer 1965). The PDSI determines the severity of the wet or dry period per calendar-based month relative to a standard for a geographical region. The ZNDX expresses the departure of moisture conditions per month from the average moisture conditions for that month and is considered an agricultural drought index. It does not give insight to the duration or severity of the wet or dry period like the PDSI (Keyantash and Dracup 2002). The SPI expresses a precipitation deficit or surplus relative to historical records for the region. It can be used to compare moisture conditions between locations and years (Guttman 1998; Keyantash and Dracup 2002; McKee et al. 1993). We refer interested readers to Alley (1984), Guttman (1998), Keyantash and Dracup (2002) and Palmer (1965) for further information on these indices.

Monthly values for the PDSI, ZNDX and SPI for climate divisions in Kansas (9 divisions), Oklahoma (9 divisions) and Texas (10 divisions) were obtained from the National Climatic Data Center (NCDC). Index values were averaged over climate divisions to summarize moisture conditions within major wheat producing regions each month. Major wheat producing regions were defined as climate divisions with greater than 180,000 hectares of harvested winter wheat annually during 1999 to 2012. These included the western and central climate divisions of both Kansas and Oklahoma; and the northern climate divisions of Texas (National Agriculture Statistics Service Census of Agriculture, http://www.agcensus.usda.gov/Publications/2012/Full_Report/Census_by_State/; Fig. 3.1). Climate divisions in southern Texas were included to account for environmental conditions in potential stripe rust overwintering locations (Fig. 3.1). Climate divisions and CRDs represent the same geographical areas in Kansas.

Data analysis

There were 126 regional yield loss estimates from 1999 to 2012 (9 CRDs by 14 years). Each yield loss estimate was linked to 847 variables representing monthly (August to July) summaries of weather conditions within CRDs and monthly soil moisture indices. Variables were arranged into five groups representing the type of information conveyed: (i) soil moisture indices (571 variables), (ii) temperature (96 variables), (iii) RH (36 variables), (iv) precipitation (24 variables), and (v) combinations of temperature and RH (120 variables).

The analysis was done in two phases. In the first phase, a case was classified as an epidemic if the CRD-level yield loss due to stripe rust was greater than or equal to 1% and as a non-epidemic otherwise. The choice of 1% as the classification threshold stemmed from wheat yield losses above that threshold being financially relevant to the statewide agricultural economy. The emphasis was on identifying variables capable of separating stripe rust epidemics (43 cases) from non-epidemics. Phase one considered all 847 variables for the wheat growing season starting in August to July the following year.

Phase two focused on the 43 stripe rust epidemic cases by categorizing an epidemic as either moderate (yield loss of 1 to 14%) or severe (yield loss >14%). The goal of phase two was to identify variables which distinguished between moderate (21 cases) and severe epidemics (22 cases). The candidate set of variables summarized weather conditions from March to May. These months corresponded to the period when the penultimate and flag leaves were emerging in Kansas winter wheat, a phenologically important stage during which fungicide application decisions are made. Group (i) variables were excluded from the candidate set because of the timing of data availability. The NCDC data release cycle is one to two weeks after the month's end, making it impractical to include soil moisture indices in any phase two model that may eventually be used in a predictive capacity.

Classification trees (CT) were used to select variables from each group (Hastie et al. 2009). CT analysis was done with JMP Pro (Version 10.0, SAS Institute Inc., Cary, NC). For a given variable, the goal of recursive binary splitting was to identify a threshold value that minimized node impurity. Trees were restricted to two-way interactions and a minimum of 5 cases per node because of the small dataset. Variables were selected by ranking the likelihood ratio chi-square statistic (G^2) and the area under the receiver operating characteristic curve (ROC). Note that in the JMP software $G^2 = 2 \ln(D)$, where D is the entropy, which means that G^2 increases with node purity. The two variables with the highest G^2 and ROC scores in each of the five groups were selected for further evaluation.

Variables selected through CT were used as the independent variables in one- or two-variable generalized estimating equation (GEE) models of epidemics (Hardin and Hilbe 2013). A logit link function was used to model the binary responses (non-epidemic versus epidemic, moderate versus severe epidemic). The quasi-likelihood under the independence model information model criterion (QIC) statistic was used to compare models with different working correlation structures for within-year correlation. Model fitting was continued with the independent correlation structure after comparing the QIC estimates with different correlation structures. Models were compared using the QIC_u statistic (Pan 2001). Residuals were plotted to check for outliers and patterns indicative of model assumption violations. All cases were retained after examining the residual plots (i.e. there were no outliers). GEE modeling was done with the GENMOD and UNIVARIATE procedures of SAS® (Version 9.2, 64 bit, SAS Institute Inc., Cary, NC). GEE model output consisted of predicted probabilities which required conversion to a class membership (non-epidemic or epidemic for phase one models and moderate or severe epidemic for phase two models). We used the Youden Index (YI), which is the maximum difference between the true positive and false positive rates, as a guide to identify the cut-point for class assignment based on balancing sensitivity and specificity. Therefore, classification was based on maximizing the balance between the true positives and true negatives and without explicit consideration of the costs of misclassification errors.

To assess the utility of the environmental variables from a predictive standpoint, the prediction accuracy of the phase one GEE models was estimated by a modified form of *K*-fold cross-validation (Hastie et al. 2009) called cross-year validation (Landschoot et al. 2012), in which each fold *K* contained all observations for one year only. For the current study, this meant 14 folds (years), each with nine observations (CRDs). The cross-year validation approach is more useful for estimating predictive performance on future (unobserved) years (Landschoot et al. 2012). As there were only 43 cases for phase two models; leave-one-out cross validation was used instead (Hastie et al. 2009).

Results

Variable selection

In phase one, 10 variables (two variables with the highest G^2 and ROC per group) were selected through the CT analysis (Table 3.1). Group (i) (regional soil moisture) variables clearly stood apart when compared with the best variables from the other groups (Fig. 3.2). Seven of the ten environmental variables were associated with weather or soil moisture conditions from late winter to early spring (February to March). The other three variables represented soil moisture or temperature at planting or during crop establishment from fall to early winter (October to December). In phase two, variables in groups (ii) and (v) were most associated with high yield loss (Table 3.1; Fig. 3.3). Seven of the eight weather-based variables identified in phase two summarized conditions in April and May, which corresponds to jointing through early grain fill in Kansas wheat. Rainfall-based variables (group iv) were among those with the lowest G^2 and ROC scores (Figs 3.2 and 3.3) in both phases.

Phase one models

Single-variable GEE models with either ZNDX_Establish (Model 1) or ZNDX_02 (Model 2) had lower QIC_u and cross-validated misclassification rates than other variables identified by CT. Conditional on the cut-point for classification, Models 1 and 2 both had an accuracy rate over 85% (Table 3.2). However, Model 2 had a 41% less false negative rate (erroneously classifying a stripe rust epidemic as a non-epidemic) than Model 1.

Model 3 was a linear combination of ZNDX_Establish and ZNDX_02. Having both variables in the model resulted in a lower QIC_u statistic compared with both Models 1 and 2 (Table 3.2). However from a predictive standpoint, the cross-validated misclassification rate with Model 3 was the same as with Model 2. Nevertheless, Model 3 gave a relatively wide decision boundary separating stripe rust epidemics from non-epidemics (Fig. 3.4). An evaluation of model errors indicated that three false positives occurred in eastern Kansas.

Phase two models

All three phase two models were based on temperature conditions in May (Table 3.3). Model 5 had the lowest QICu and the highest classification accuracy rate of 79%. Model 5, combining the accumulation of temperatures between 7 to 12°C and RH>87%, had a 50% decrease in the false negative rate compared with Model 4 using temperature alone (Table 3.3). The estimated predictive performances of Models 4 and 6 were similar, with a classification accuracy rate of 70% for the two models. No pattern was discernable among the misclassified observations.

Discussion

There are no stripe rust forecast models for Kansas winter wheat. As a first step towards filling that need, the current study analyzed environmental variables for association with regional stripe rust epidemics over a 14-year period. Soil moisture indices appeared promising as predictors of stripe rust epidemics in crop reporting districts (CRDs) as a whole. Conditional on an epidemic occurring, temperature-based variables seemed to have worth in discerning between moderate and severe epidemics. Simple, single-variable models could be used in combination with field-level disease observations and varietal resistance to inform the need for foliar fungicides against stripe rust in Kansas wheat.

Variables summarizing soil moisture conditions were positively associated with stripe rust epidemics, with the ZNDX showing a stronger association than either PDSI or SPI. ZNDX values summarizing soil moisture conditions in autumn (October, November and December) and winter (February) were most strongly associated with stripe rust epidemics. The importance of moisture during similar time periods relative to crop growth was also noted in Australia (Park 1990) and Europe (Papstamati and van den Bosch 2007; Te Beest et al. 2008; van den Berg and van den Bosch 2007). In the Great Plains, October, November and December correspond with winter wheat planting and establishment. Soil moisture during this time may influence crop growth and the development of canopies creating microenvironments that favor (or suppress) the development of stripe rust. *P. striiformis* requires a wet leaf surface in order to infect. A possible source of this moisture is dew that originates from soil moisture itself, via a distillation process (Jacobs et al. 1990). The water vapor generated from soil moisture remains an important source of dew until the canopy grows, at which time atmospheric water vapor becomes the primary source of dew (Jacobs et al. 1990). Later on, soil moisture in February may influence winter survival and early stages of an epidemic when the pathogen is establishing foci or "hot-spots". These initial foci are an important source of inoculum for a developing epidemic (Cowger et al. 2005; Zadoks and van den Bosch 1994). The importance of moisture in the winter months is consistent with findings from the PNW region of the U.S. (Coakley et al. 1988; Sharma-Poudyal and Chen 2011).

In contrast to models proposed for the PNW (Coakley et al. 1983; Coakley et al. 1982, 1984, 1988; Sharma-Poudyal and Chen 2011) and Europe (Christensen et al. 1993; Gladders et al. 2007; Papstamati and van den Bosch 2007; Te Beest et al. 2008; van den Berg and van den Bosch 2007), temperatures during winter (January to February) were not strongly associated (either positively or negatively) with stripe rust epidemics in Kansas. It is possible that winter conditions in the Central and Southern Great Plains are not restrictive to *P. striiformis* in enough years to be useful as predictors of stripe rust epidemics. It is also conceivable that urediniospore dispersal from overwintering sites in southern Texas, where temperatures would rarely restrict winter survival (Sharma-Poudyal et al. 2013), masked the true effect of temperature on winter survival of stripe rust in Kansas. Winter survival of *P. striiformis* in Kansas is still an open research question.

Phase two of the study focused on whether environmental variables were useful in classifying moderate and severe epidemics, given an epidemic occurrence. Environmental conditions during April and May were associated with severe epidemics across a CRD. These two months are when Kansas winter wheat is progressing through jointing to grain fill, a period when stripe rust infections can damage leaf tissue, thus increasing the risk of severe epidemics and yield loss (Mundy 1973). The duration of favorable temperature, alone or in combination with RH, was a useful predictor of severe stripe rust epidemics. The regional association of temperature with stripe rust epidemics is consistent with previous research in Europe (Gladders et al. 2007; Papstamati and van den Bosch 2007; Te Beest et al. 2008; van den Berg and van den Bosch 2007) on the role of temperature in stripe rust disease development. In the current study, temperatures between 7 and 12°C were more strongly associated with severe epidemics at the CRD level than other temperature ranges reported in the literature. Consistent with field observations from Kansas (Eddy 2009), we found that temperatures greater than 23°C had a negative association with severe stripe rust epidemics in a CRD (i.e. the probability of a severe stripe rust epidemic decreased with 171 hours of temperatures above 23°C). The suppressive effect of warm temperatures on stripe rust is well-documented (Coakley 1988; de Vallavieille-Pope et al. 1995; Shaner and Powelson 1971). Spring precipitation was less associated with severe stripe rust which is consistent with results from the PNW (Sharma-Pouydal and Chen 2011).

Single- or two-variable models may be useful as components of a disease forecasting system for stripe rust in Kansas. There is a chronological progression from phase one to phase two, which suggests the sequential deployment of any future stripe rust models during the Kansas growing season. A preliminary forecast of the risk of a stripe rust epidemic across a CRD can be based on soil moisture conditions in autumn. Updated forecasts can be provided in March after the NCDC releases the February ZNDX values. As Kansas wheat approaches heading, phase two modeling could then inform disease scouting and varietal resistance to make a field-level forecast. Because phase two models would be used just before the critical time for fungicide decisions, it may not be practical to wait until the end of May to release an updated forecast. In this situation, it may be possible to monitor the accumulation of favorable or unfavorable weather throughout the critical time period.

The two-phase modeling approach to regional stripe rust forecasting was informally evaluated during the 2013, 2014, and 2015 wheat growing seasons. In 2013 and 2014 phase one models predicted non-epidemics due to dry soil moisture conditions in the Great Plains. In 2013 stripe rust was identified by the Kansas wheat survey. However the disease failed to progress because of unfavorable May temperatures which emphasized the importance of the phase two models. In 2014, the phase two models were unnecessary due to the dry environmental conditions for the entirety of the wheat growing season and the lack of regional observations of disease. There were trace losses to stripe rust in Kansas in 2013, and negligible losses to stripe rust in 2014.

For the 2015 growing season, phase one models predicted that the risk of a stripe rust epidemic was low, based on fall and early winter soil moisture; the Great Plains ranged from abnormally dry to exceptional drought during this time (U.S. Drought Monitor). Stripe rust was identified by 28 January 2015 in Texas. Then, Kansas and Oklahoma experienced numerous rainfall events (and hence increased soil moisture) in March 2015, which is outside the data space of phase one models. Stripe rust was detected in Kansas by the middle of April 2015. At this point, with the establishment of stripe rust in Kansas, the deployment of phase two models became important. Temperatures remained favorable for continued stripe rust development through April and May 2015, resulting in an estimated 15% overall yield loss due to stripe rust in Kansas wheat. The scenario described for 2015 highlighted the utility of having two modeling phases, as well as some shortcomings. Therefore, these types of stripe rust models need to be coupled with field-level disease observations and growers’ knowledge of variety resistance reactions when deciding to apply a fungicide against stripe rust.

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Figures and Tables

Table 3.1 Environmental variables used as input for phase one and phase two models

Phase[a]	Group[b]	Variable Code[c]	Description
One	i	ZNDX_02[d]	Average ZNDX in Kansas, Oklahoma, and Texas in February
	i	ZNDX_Establish	Average ZNDX in Kansas in October, Oklahoma in November, and Texas in December
	ii	TAVG_10	Average temperature (°C) in October
	ii	T1015_08	Number of hours in August with temperature between 10 and 15°C
	iii	RH87_02	Number of hours in February with relative humidity greater than 87%
	iii	RHAVG_02	Average relative humidity (%) in February
	iv	R_02	Sum of precipitation (mm) in February
	iv	DAYSR_02	Days with precipitation in February
	v	T0223RH90_02	Number of hours in February with temperatures between 2 and 23°C and relative humidity greater than 90%
	v	T0223RH90_03	Number of hours in March with temperatures between 2 and 23°C and relative humidity greater than 90%
Two	ii	T0712_05	Number of hours in May with temperatures between 7 and 12°C
	ii	T23_05	Number of hours in May with temperatures greater than 23°C
	iii	RH90_04	Number of hours in April with relative humidity greater than 90%
	iii	RHAVG_04	Average relative humidity (%) in April
	iv	R_04	Total rainfall (mm) in April
	iv	R_03	Total rainfall (mm) in March
	v	T0712RH87_05	Number of hours in May with temperatures between 7 and 12°C and relative humidity greater than 87%
	v	T12RH87_04	Number of hours in April with temperatures greater than 12°C and relative humidity greater than 87%

[a] Phase one modeled the probability of stripe rust epidemic occurrence whereas phase two modeled the probability of severe (>14% loss) stripe rust epidemics when epidemics occurred.

[b] Variables were grouped by type of information represented: (i) soil moisture indices, (ii) temperature, (iii) relative humidity (RH), (iv) precipitation, and (v) combinations of temperature and RH.

[c] Acronyms describe the variables summarizing regional soil moisture indices (ZNDX, Palmer's Moisture Anomaly Index), temperature (T), average temperature (TAVG), relative humidity (RH), average relative humidity (RHAVG), rainfall (R), and days with rainfall (DAYSR). The number after the variable acronym specifies the value or range used, and _number indicates the month (ranging from January (01) to December (12)). For example, T0712RH87_05 signifies the number of hours in May (_05) with temperatures (T) between 7 and 12°C and relative humidity (RH) greater than 87%.

[d] ZNDX_Establish describes the soil moisture conditions when winter wheat is planted and established in Kansas (October), Oklahoma (November), and Texas (December). Regions used for the average ZNDX_02 and average ZNDX_Establish are shaded gray in Fig.1.

Table 3.2 Phase one generalized estimating equation models for classifying stripe rust epidemics and non-epidemics in Kansas

Model number	Predictor[a]	QIC_u[b]	Youden Index[c]	TN[d]	TP	FN	FP	Misclassification rate[e]
Model 1	ZNDX_Establish	58.7	0.67	82	26	17	1	0.14
Model 2	ZNDX_02	82.8	0.50	80	33	10	3	0.10
Model 3	ZNDX_Establish + ZNDX_02	51.6	0.62	80	33	10	3	0.10

[a] See Table 3.1.

[b] The QIC_u statistic (Pan 2001) is used to compare different generalized estimating equation (GEE) models. GEE models with smaller values are preferred.

[c] The Youden Index was used as the cut-point for converting model-generated probabilities to the classification of an observation as a stripe rust epidemic or non-epidemic.

[d] There were 126 observations total. TN = true negatives (number of non-epidemics correctly classified) (83 cases); TP = true positives (number of epidemics correctly classified) (43cases); FN = false negatives (number of epidemics incorrectly classified as non-epidemics); FP = false positives (number of non-epidemics incorrectly classified as epidemics).

[e] The proportion of the total observations that were incorrectly classified.

Table 3.3 Phase two generalized estimating equation models for classifying moderate and severe stripe rust epidemics in Kansas

Model number	Predictor[a]	QIC_u[b]	Youden Index[c]	TN[d]	TP	FN	FP	Misclassification rate[e]
Model 4	T0712_05	54.4	0.70	20	10	12	1	0.30
Model 5	T0712RH87_05	50.6	0.54	18	16	6	3	0.21
Model 6	T23_05	57.3	0.62	17	13	9	4	0.30

[a] See Table 3.1.

[b] See Table 3.2, footnote b.

[c] The Youden Index was used as the cut-point for converting model-generated probabilities to the classification of an observation as a moderate or severe stripe rust epidemic.

[d] There were 43 observations total. TN = true negatives (number of moderate epidemics correctly classified) (21 cases); TP = true positives (number of severe epidemics correctly classified) (22 cases); FN = false negatives (number of severe epidemics incorrectly classified as moderate epidemics); FP = false positives (number of moderate epidemics incorrectly classified as severe epidemics).

[e] See Table 3.2 footnote e.

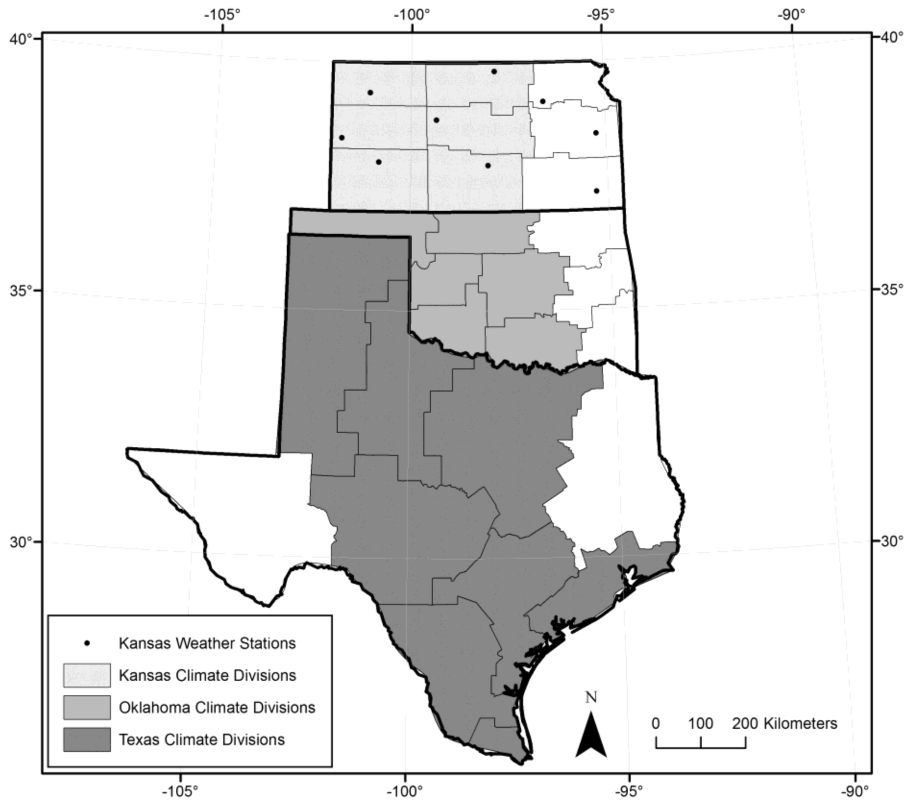

Figure 3.1 Climate divisions in Kansas, Oklahoma and Texas. Within Kansas, crop reporting districts and climate divisions are the same spatially. Points in Kansas indicate the locations of weather stations used to supply hourly weather data. Gray areas indicate climate divisions over which regional soil moisture conditions were averaged.

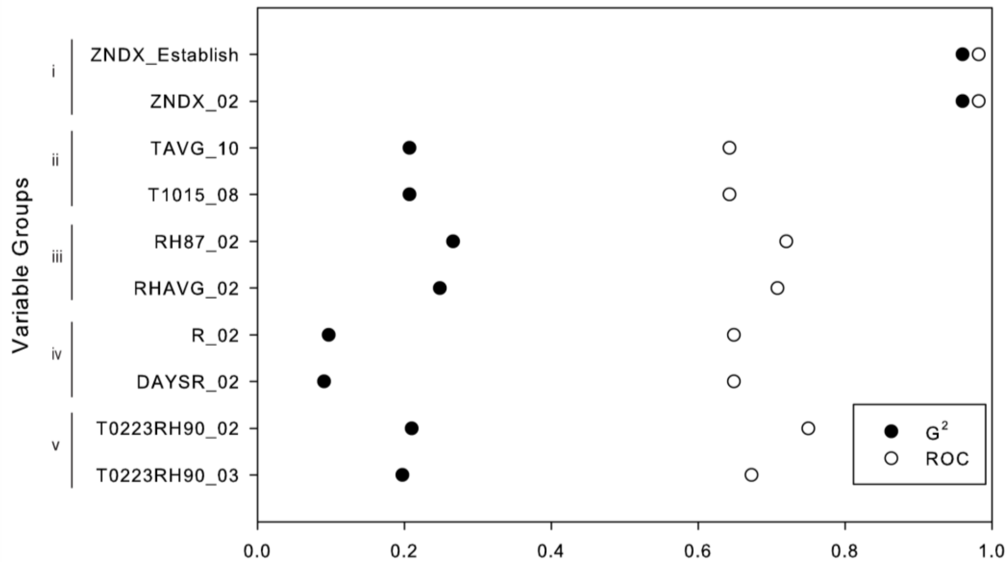

Figure 3.2 The likelihood ratio chi-square statistic (G^2) and receiver operating characteristic (ROC) curve statistics for the first-split variables in classification trees fit to stripe rust epidemics and non-epidemics in Kansas. Groups represent soil moisture indices (group i), temperature (group ii), relative humidity (group iii), precipitation (group iv), and combined temperature and relative humidity conditions (group v). See Table 3.1 for full descriptions of predictor acronyms.

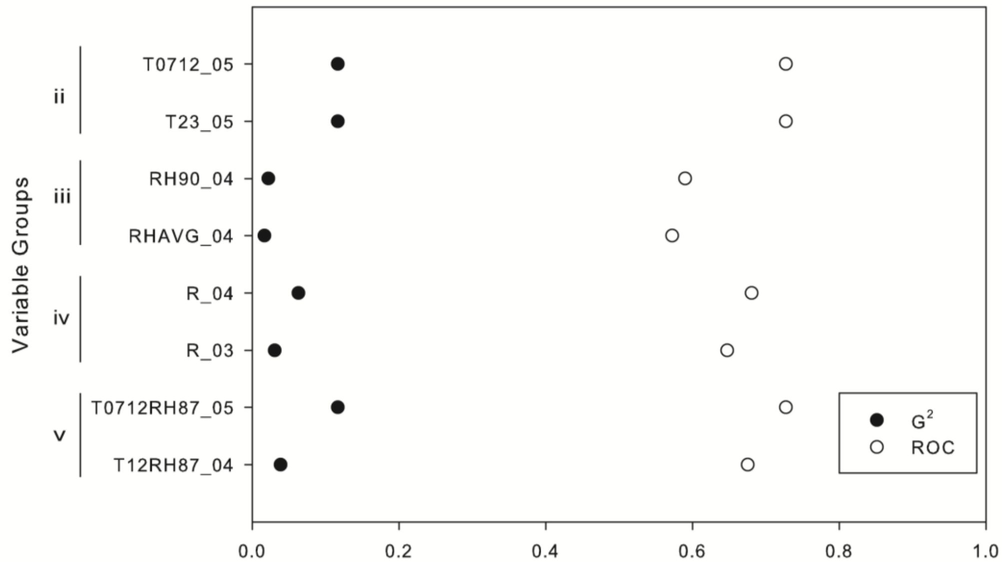

Figure 3.3 The likelihood ratio chi-square statistic (G^2) and receiver operating characteristic (ROC) curve statistics for the first-split variables in classification trees fit to moderate and severe stripe rust epidemics. Groups represent temperature (group ii), relative humidity (group iii), precipitation (group iv), and combined temperature and relative humidity conditions (group v). See Table 3.1 for full descriptions of predictor acronyms.

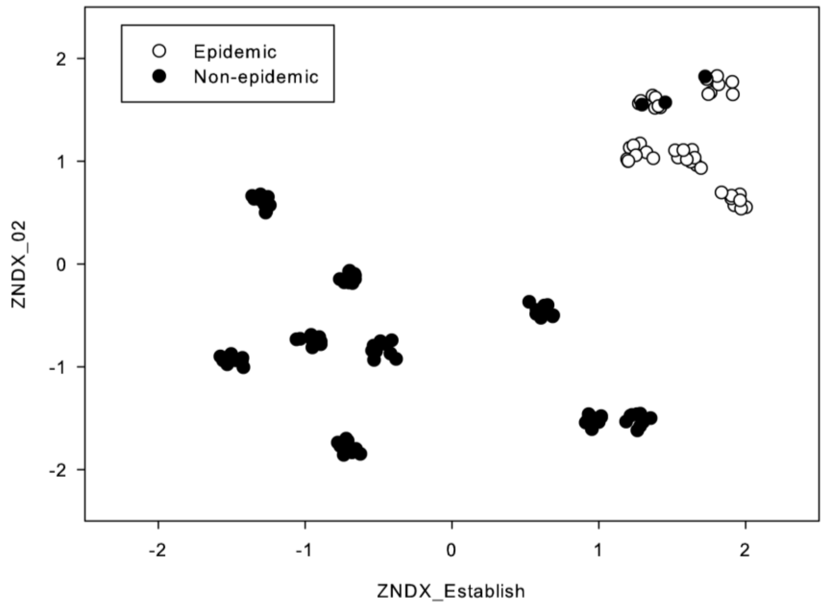

Figure 3.4 Separation of stripe rust epidemics and non-epidemics by phase one Model 3. Jitter was added to visualize overlapping points.

Chapter 4 - Environmental variables associated with leaf rust in Kansas winter wheat

Abstract

Leaf rust (caused by *Puccinia triticina*) of wheat is an economically important disease and on average causes losses of 18.4 million U.S. dollars per year in Kansas. The objectives of this research were to identify environmental conditions that are conducive for leaf rust epidemics in the central Great Plains region of the U.S., and to develop preliminary predictive models for Kansas. The analysis was divided into two phases. The first phase identified the monthly environmental variables associated with leaf rust epidemics (regional yield loss >1%). The second phase of the analysis considered the monthly variables that were associated with severe yield loss (yield loss ≥7%) within epidemic years. Variables used in this analysis included: temperature and moisture requirements for leaf rust infection based on previous literature, remotely sensed canopy characteristics, soil moisture conditions, and climate indices. Boosted regression trees were used to identify monthly variables associated with leaf rust epidemics in Kansas. Variables identified in the variable selection procedure were used to develop predictive models in logistic regression. The models indicate that leaf rust epidemics are influenced by soil moisture conditions in the southern Great Plains during winter and spring. The models also indicate that extended periods of favorable temperature and relative humidity combination (15 to 20°C and relative humidity >87%) and precipitation in May are most associated with severe yield losses in epidemics years. The variables and leaf rust models at the crop reporting district (CRD) level could be used in a disease forecasting system for Kansas wheat when coupled with regional disease observations and knowledge of the variety reaction to leaf rust infections.

Introduction

Leaf rust, caused by *Puccinia triticina* Erikss., is a common disease of wheat (*Triticum aestivum* L.) that causes significant yield losses worldwide. The 20-year average yield loss from leaf rust in Kansas is 2.1%; which can result in economic losses of 27.0 million U.S. dollars per year. Since 1976, the statewide yield loss has exceeded 2% in 19 out of 40 years. In 2007 the highest statewide yield loss from leaf rust was 13.9% resulting in 272 million U.S. dollars lost to Kansas's wheat growers.

Leaf rust regularly overwinters in the southern U.S. and the pathogen is wind dispersed into the central Great Plains but it has also been documented to overwinter in Kansas (Eversmeyer et al. 1988). Eversmeyer et al. (1988) found leaf rust to overwinter in 6 out of 7 years in Kansas and found yield losses greater than 2% when leaf rust overwintered in northeastern Kansas (Eversmeyer et al. 1988). The main method of leaf rust control is genetic resistance. However, the loss of avirulence genes within the pathogen population allow it to overcome wheat resistance genes in widely planted, popular varieties (Flor 1971; Hulbert et al. 2001). When genetic resistance is not effective, growers use foliar fungicides for managing leaf rust. The decision to apply a foliar fungicide depends on pathogen presence, variety susceptibility, favorable weather, application costs and grain market value (Carlson and Main 1976; Edwards et al. 2012; Guy et al. 1989; Wegulo et al. 2011; Willyerd et al. 2015).

Like most fungal pathogens, *P. triticina* requires specific temperature and moisture conditions for infection processes. Previous research has shown that leaf rust infections can occur over a wide range of temperatures including 5 to 25°C, 15 to 20°C, 10 to 25°C, and 10 to 30°C with moisture present (Bolton et al. 2008; Chester 1946; de Vallavieille-Pope et al. 1995; Roelfs et al. 1992). de Vallavieille-Pope et al. (1995) demonstrated that 6 hours of moisture were required for leaf rust infections at 15°C, the optimum temperature in the study. More hours of leaf wetness were required when temperatures deviated above or below the optimum temperature. Leaf rust infections can also be restricted by temperature (de Vallavieille-Pope et al. 1995; Hogg et al. 1966; Roelfs et al. 1992). Temperatures greater than 30°C will reduce leaf rust germination by 96% with no spores germinating at temperatures greater than 35°C regardless of the presence of moisture (de Vallavieille-Pope et al. 1995).

The relationship between weather conditions on wheat leaf rust disease severity and incidence has been investigated. The earliest model for leaf rust development in the Great Plains is based on the concept of the "critical month" (Chester 1946). Chester (1946) found March (stem elongation) to be the critical month for predicting leaf rust severities with final disease development with temperatures greater than 10°C associated with extensive leaf rust development in Oklahoma. Leaf rust severity has been found to be associated with free moisture and minimum temperatures in Kansas (Eversmeyer and Burleigh 1970; Burleigh et al. 1972). Minimum temperatures, deviation from optimum temperatures (December) and snow cover (December and February) during winter dormancy, and precipitation during oversummering and autumn crop establishment (July, September and October) were important weather variables for predicting overwinter and spring survival of leaf rust in Kansas (Eversmeyer and Kramer 1996; Eversmeyer and Kramer 1998). In Sweden, precipitation prior to planting (August) and temperature and precipitation during winter crop dormancy (January) were correlated with leaf rust severity and incidence (Wiik and Ewaldz 2009). Temperature during winter dormancy (February) and mid to late tillering (April) and precipitation at the beginning of tillering (March) were used to predict leaf rust disease incidence at the mid-anthesis growth stage with high accuracy (Wiik and Ewaldz 2009).

Environmentally based predictors have been previously utilized for predicting leaf rust severity and overwintering potential in Kansas. These models have largely focused on field-level disease and not on the overall regional risk. Therefore the objective of this research was to identify environmental variables associated with leaf rust epidemics at a regional scale in Kansas and to evaluate these predictors for utility as potential predictive models.

Methods

Leaf rust yield loss data

Records of estimated yield loss to leaf rust between 1994 and 2013 were obtained from the Kansas Cooperative Plant Disease Survey Reports (http://agriculture.ks.gov/divisions-programs/plant-protect-weed-control/reports-and-publications). The survey effort has been described previously in Bockus et al. 2001 and Bockus et al. 2011 and will not be detailed here. In general, these reports include observations of leaf rust incidence and flag leaf severity (0-100%) on susceptible varieties per crop-reporting district (CRD) from non-fungicide treated variety performance trials, county demonstration plots, and on-farm research locations. A wheat leaf rust yield loss model (Bowden et al. unpublished) is applied to the leaf rust disease estimates to obtain loss estimates on susceptible varieties. To determine the yield loss per CRD, the acreage planted to susceptible varieties from National Agriculture Statistic Service (NASS) was used to estimate the yield loss per CRD on susceptible varieties. The yield loss estimates are corroborated with observations from fungicide research plots and arbitrarily selected commercial wheat fields.

Variables from meteorological data

Hourly temperature (°C), relative humidity [RH] (%), and precipitation (mm) weather data were obtained from eight automated surface observing systems (ASOS) (ZedX Inc. Bellefonte, Pa) and one automated weather station from the Kansas Weather Data Library (http://mesonet.k-state.edu/). The National Weather Service and the Federal Aviation Administration maintain the ASOS weather stations and the Kansas Weather Data Library station is maintained by Kansas State University. Weather station locations were chosen based on their close proximity to wheat producing areas within each CRD and the completeness of weather records for the years considered in this analysis. Examining hourly time series plots of the weather data and descriptive statistics identified missing or erroneous observations. Common errors included out of range values (RH>100%) or abnormal values given the other available data in the time series. For example if a value was -20°C but the surrounding hourly time series were between 19 to 21°C. Short time periods with missing or erroneous values were addressed by averaging observations from the adjacent hours in the time series.

Calendar-based monthly summaries were constructed from hourly time series from August (planting) to July (harvest). These summaries included descriptive variables like average temperature, average relative humidity and total precipitation. Variables describing known relationships between leaf rust infection processes and environment were constructed by summarizing the number of hours a specific condition was fulfilled per month (e.g. accumulation of hours with temperatures between 5 to 25°C). Many optimal and favorable temperature ranges have been documented for leaf rust (i.e. 5 to 25°C, 15 to 20°C, 10 to 25°C, and 10 to 30°C). These differences are likely based on the component of the disease cycle studied, isolates tested, and research methodologies applied. Numerous temperature ranges were evaluated to avoid any *a priori* bias in variable selection. Variables describing temperatures that could restrict the development of leaf rust were also considered (i.e. temperatures greater than 30°C and 35°C). Two variables were created to recreate the concepts proposed by Chester in 1946; if the average temperature was greater than 10°C and the number of hours with temperatures greater than 10°C per month. A relative humidity variable summarized the number of hours with RH greater than 87% as a proxy for conditions that would favor leaf wetness. A leaf wetness variable was developed using dew point temperature to estimate if dew would be present based on the dew point temperature (Ham 2005). Precipitation variables included the number of hours with measureable precipitation (precipitation greater than 0.25 mm) or total precipitation per month. Variables summarizing temperature and relative humidity or precipitation conditions being met simultaneously were used to describe favorable temperature and moisture combinations (e.g. the number of hours that temperatures were between 5 and 25°C and RH>87%).

Regional moisture indices

The Palmer Drought Severity Index (PDSI; Palmer 1965), Palmer's Moisture Anomaly Index (ZNDX; Palmer 1965) and the Standard Precipitation Index (SPI; McKee et al. 1993) were included to quantify moisture conditions at a regional scale. These three indices are highly correlated, but differ in moisture variables and time periods considered.

The Palmer indices use a reference set of water balance variables compared to the observed meteorological conditions to develop a dimensionless index value representing the current moisture supply relative to a standard (Palmer 1965). The PDSI includes a duration factor to determine the longevity of a moisture anomaly (unusually wet or dry) by defining the severity of the wet or dry period per calendar-based month relative to the standard for a geographical region. The Palmer Moisture Anomaly Index (ZNDX) expresses the departure of the moisture conditions per month from the average conditions for that month. The ZNDX is considered an agricultural drought index but does not give insight into the duration or severity of the wet or dry period like the PDSI (Keyantash and Dracup 2002).

The SPI expresses a precipitation deficit or surplus relative to historical records for the region. The SPI normalizes the data distribution and facilitates comparison between locations and years (Guttman 1998; Keyantash and Dracup 2002; McKee et al. 1993). Numerous reviews are available on the development of the PDSI, ZNDX, and SPI. We refer interested readers to the following articles by Alley (1984), Guttman (1998), Keyantash and Dracup (2002) and Palmer (1965).

One-month PDSI, ZNDX, and SPI values for climate divisions in Kansas, Oklahoma, and Texas were obtained from the National Centers for Environmental Information (NCEI; formerly the NCDC). Climate divisions were averaged within major wheat producing and key epidemiological regions to summarize moisture conditions at the individual CRD, state, and multi-state level (Kansas, Oklahoma, and Texas combined) per month (Fig. 4.1). The CRD and climate divisions represent the same geographic area of Kansas; therefore the regional yield losses are associated with the climate divisions for the analysis.

Climate indices

Niño 3.4, Niño 4.0, and Oceanic Niño (ONI) climate indices based on sea surface temperatures were obtained from the Climate Prediction Center to determine if leaf rust epidemics are associated with the warming or cooling periods that influence Niño events in the central Great Plains (Rasmussen and Carpenter 1982). The Niño 3.4 and Niño 4.0 variables included the raw temperature values and anomalies with a 30-year base period (1971-2000) per month (Trenberth 1997). The Niño 3.4 region is used to classify the intensity of El Niño events when the sea surface temperatures anomalies greater than or equal to 0.5°C and La Niña events when less than or equal to -0.5°C for 3 consecutive months (Barnston et al. 1997). The region measures changes in sea surface temperatures from 5°N to 5°S and 170°W to 120°W and describes the average equatorial sea surface temperatures and anomalies. The Niño 4.0 focuses on the central equatorial pacific area from 5°N to 5°S and 160°E to 150°W and tends to have less variability than the Niño 3.4 region (Stenseth et al. 2003). One three-month averaged teleconnection pattern, ONI, was included in the analysis. The ONI is the primary index used to predict the El Niño-Southern Oscillation (ENSO) (Kousky and Higgins 2007). The ONI is based on the sea surface temperature departures (base period 1971-2000) in the El Niño 3.4 region. El Niño or La Niña events are characterized by greater than or equal to +0.5°C or -0.5°C ONI values, respectively. While these indices are highly correlated, they differ in the Pacific Ocean region and time periods considered.

The Southern Oscillation Index (SOI) is based on the observed sea level pressure anomalies between Tahiti (17.6509°S and 149.4260°W) and Darwin, Australia (12.4628°S and 130.8418°E) using the base period of 1951-1980 (Trenberth 1984). The negative SOI anomalies are associated with negative air pressure in Tahiti and positive air pressure in Darwin resulting in warm ocean waters classified as El Niño events (NCEI). Cool ocean temperatures occur when the opposite air pressure anomalies occur in the two locations resulting in positive values classifying La Niña events (NCEI).

The Niño 3.4, Niño 4.0, ONI and SOI were obtained from the Climate Prediction Center within the NCEI (http://www.cpc.ncep.noaa.gov/data/indices). Monthly values for the sea surface temperature, anomalies, and sea level pressure were assigned per calendar-based month and the ONI was assigned to the month based on the second month of the three-month period.

Regional biomass index

The normalized difference vegetation index (NDVI) was used to determine greenness and general health of winter wheat for each CRD. NDVI is the ratio of near infrared (NIR) and the red (RED) regions of the electromagnetic spectrum (Eq. 4.1) (Tucker 1979). NDVI data was collected from the Advanced Very High Resolution Radiometer (AVHRR) daily observations that are composited into a weekly image displaying maximum greenness (USGS). The AVHRR images have a 1 km spatial resolution. The NDVI Departure from Normal images was used to display areas of above average or below average greenness. The near infrared wavelength is measured from channel 2 (0.75-1.10 µm) and the red wavelength from channel 1 (0.58-0.68 µm). The analysis was completed in ArcGIS (Release 10.1, Environmental Systems Research Institute, Redlands, CA). The images were extracted using a climate division shapefile for Kansas, Oklahoma, and Texas and resampled to the same resolution. The USGS GAP analysis raster file was resampled and used to extract only the NDVI pixels from the Cultivated Cropland Ecological System Land Use Class (http://gis1.usgs.gov/csas/gap/viewer/land_cover/Map.aspx). This resulted in weekly images of NDVI on cultivated cropland per climate division. Zonal statistics were used to calculate the mean NDVI per climate division. The average Departure from Normal NDVI was calculated for Kansas, Oklahoma, and Texas individually per month and averaged over the three states using the same climate divisions as described in the regional soil moisture section.

EQ 4.1 $NDVI = (NIR - RED) \div (NIR + RED)$

Data analysis

There were 180 regional yield loss estimates from 1994 to 2013 (9 CRD's by 20 years). Each yield loss estimate was linked to 487 variables representing monthly (August of planting year to July of harvest year) summaries of (T) temperature (100 variables), (RH) relative humidity (24 variables), (LW) leaf wetness (12 variables), (P) precipitation (24 variables), (TRH) temperature and relative humidity combination (48 variables), (TP) temperature and precipitation combination (48 variables), (DRT) regional soil moisture (108 variables), (NDVI) vegetation greenness (48 variables), and (CLI) climate indices (72 variables).

The analysis was completed in two phases. In the first phase, any case with greater than 1% CRD-level yield loss due to leaf rust was classified as an epidemic or non-epidemic otherwise. The 1% classification threshold was based on the median yield loss for the dataset and the serious economic implications of wheat yield losses exceeding 1% statewide. This threshold resulted in 67 epidemic cases to identify variables capable of distinguishing leaf rust epidemics. Phase one considered all 487 variables for the entirety of the wheat-growing season.

Phase two emphasized identifying variables capable of distinguishing severe epidemics from moderate epidemics in epidemic years. A severe epidemic was classified as CRD-level yield loss greater than or equal to 7% (27 cases) and a moderate epidemic (40 cases) otherwise. The 7% threshold is the mean of the epidemic cases. Phase two considered only variables from March to May which corresponds to the period when the penultimate and flag leaves are emerging in Kansas winter wheat. These growth stages are important phenologically and temporally for fungicide application decisions in Kansas. DRT, CLI, and NDVI variables were not considered for this portion of the analysis due to the lag in data availability making it impractical to use if the variables will eventually be used in a predictive framework.

Boosted regression trees (BRT) were used to select variables based on the relative influence of the predictor variables (Friedman et al. 2000). The relative influence is based on the number of times a variable is chosen for splitting and weighted by the squared improvement of the model (Elith et al. 2008). For phase one, BRT models were fit to predictors within three time periods: Fall (August to November), Winter (December to February) and Spring (March to July). BRT models are optimized by adjusting the tree complexity (*tc*), learning rate (*lr*), and the bag fraction (*bf*) to result in a total number of trees (Hastie et al. 2009). In general, the goal is to achieve 1000 trees per BRT model (Elith et al. 2008). The *tc* indicates the number of nodes in a tree representing the maximum number of interactions in the model (*tc* = 1 [i.e. an additive model], *tc* =2 [i.e. pairwise comparisons] and higher order interactions are achieved with increasing *tc* values). For this analysis, the *tc* was set to 3 to minimize the model complexity. The *lr* is used to minimize the contribution of each tree as it is added to the models. The smaller the *lr*, the slower the model "learns" and increases the number of trees produced by the model. For this analysis, *lr* = 0.005. The *bf* is the proportion of the data to be used in each tree. For this analysis, the *bf* was 0.75 resulting in 25% of the dataset being withheld from each tree. The parameter values described above resulted in greater than 1000 trees produced for each time period. BRT per season was checked for variable interactions. The BRT analysis was completed with the dismo package (ver. 07-17) in R (64 –bit version 3.1.6; R Foundation for Statistical Computing, Vienna). Variables with a relative importance greater than 1.0 were advanced in the analysis and evaluated as predictor variables for each season. For more information on BRT models we refer interested readers to Elith et al. (2008) and Hastie et al. (2009).

Variables selected through BRT were used as independent variables in one- or two-variable logistic regression models for phase one (epidemics vs non-epidemics) and phase two (severe epidemics vs moderate epidemics). Single and multiple variable models were compared using the Akaike information criterion (AIC) and receiver operating characteristic curve (ROC). Residuals were plotted to check for outliers and patterns indicative of model assumption violations. After examining the residual plots and all the cases were retained in the dataset. Logistic regression modeling was completed with STATA (Version 14, StataCorp LP, College Station, TX). The logistic modeling process resulted in predicted probabilities that required class assignment (non-epidemic or epidemic for phase one or moderate epidemic or severe epidemic for phase two models). The Youden Index (YI) was used to identify the cut-point for class membership. YI is the maximum difference between true positive and true negative rates and is based on balancing sensitivity and specificity (Youden 1950).

Phase one models were evaluated for the potential use as predictive models. The prediction accuracy was assessed using k-fold cross validation (*k*=5), in which 20 percent of the dataset (36 cases) was randomly withheld and used to estimate the predictive performance on withheld data. For the phase two models, a leave-one-out cross validation was used instead because of the small dataset (*k*=67). The cross validation was performed in STATA using the CROSSFOLD module (Daniels 2012).

Results

Variable selection

In phase one, fall, winter and spring identified 19 (Table 4.1), 23 (Table 4.2) and 21 (Table 4.3) candidate variables, respectively. Fall precipitation (P) and temperature (T) variables had the highest mean relative influence of the groups considered in this analysis (Fig. 4.2). The least influential variables groups were leaf wetness, relative humidity, and NDVI variables. The most influential individual variables in the fall were the Southern Oscillation Index in August and September (SOI_08 and SOI_09), Kansas CRD ZNDX in November (ZDNXCRD_11), the accumulation of hours with temperatures between 5 and 25°C in October (SumT0525_10), and total precipitation in October (SumPrec_10). In the winter, soil moisture indices (DRT) and precipitation (P) variable groups had the highest mean relative influence (Fig. 4.3). The most influential variables were the Palmer Drought Severity Index in Kansas (PDSIKS_12), the PDSI multi-state region in December and January (PDSIKSOKTX_12 and PDSIKSOKTX_01), and the accumulation of hours with temperatures between 5 and 25°C and precipitation in February (SumT0525Precip_02) . In the spring, temperature (T) and soil moisture (DRT) variables were the most influential variable groups. The accumulation of temperatures between 5 and 25°C in May (SumT0525_05) was the variable with the highest relative influence in the spring (Fig. 4.4).

Ten variables were selected from the BRT for phase two (Table 4.4). Within the springtime period, variables representing conditions in May were selected most often followed by variables from March. Variables combining temperature and relative humidity (TRH) conditions were the most influential variable group considered in the phase two analysis (Fig. 4.5). The accumulation of hours with temperatures between 15 and 20°C and relative humidity greater than 87% in May had the highest relative importance (SumT1520RH87_05) (Fig. 4.5). Precipitation (P) and temperature (T) variables were the second and third most important variable groups with total precipitation in May (SumPrec_05) and temperatures between 5°C and 25°C in March (SumT0525_03) having the highest relative influence.

Phase one models

Spring variables had lower AIC and higher ROC values compared to the fall and winter time periods (Table 4.5). ZNDXKS_04 had the lowest AIC and highest ROC with 74% accuracy. SumT0525_05 had a slightly higher AIC than ZNDXKS_04; however, this variable resulted in 76% accuracy. ZNDXKS_04 had 15% less false negative rate than SumT0525_05.

In the fall, SumPrecip_10 had the highest ROC and lowest AIC. Although SOI variables were identified as having a high relative importance in the BRT analysis, it did not perform as well as the other variables with logistic regression with respect to AIC, ROC and accuracy. SumT1025Precip_10 combining temperature and precipitation had the highest AIC and misclassification rate of the fall variables.

Winter variables all had ROC values less than 0.70 and had some of the highest misclassification rates compared to the other time periods. The PDSIKSOKTX_12 had the lowest AIC and highest accuracy of the winter variables. PDSIKSOKTX_12 had the lowest false negative rate of any variable identified from the BRT analysis dependent on the YI.

The variables identified in the BRT analysis were also considered in two variable combinations (Table 4.6). For phase one, Model C was a linear combination of SOI_09 and SumPrecip_10 and improved the misclassification rate by 2% and decreased the false negative rate by 13% compared to the single variable SumPrecip_10 in the Fall. For models that considered only winter variables, Model F had the highest accuracy and improved the AIC by 17 and 36 compared to Models D and E, respectively. Model F increased the false negative rate by 9% but decreased the false positive rate by 14% compared to PDSIKSOKTX_12. Combining the ZNDXKS_04 with SumT1520_05 (Model H) increased the accuracy by 5% compared to the single variable spring models. Model H had a decrease of 5% and 7% false positive and false negative rates compared to ZNDXKS_04, respectively.

Phase two models

For phase two, SumT1025RH87_03, SumT1520RH87_05 and SumT1030_05 had similar AIC and ROC values with SumT1520RH87_05 having the highest accuracy of the single variable models considered in the analysis (Table 4.7). SumT1030_05, SumT1520RH87_05 and

SumT1025RH87_03 had 15% less false negative rate than SumPrec_05 based on the YI. While SumT0525RH87_03 had the highest misclassification rate, this variable had the lowest false negative rate based on the YI compared to the other variables. The single variable model accuracies ranged from 63% to 76%.

Phase two models were combined into multivariate models and improved the accuracy compared to the single variables models (Table 4.8). Combining SumPrec_05 and SumT0525RH87_03 into a linear combination, Model K, resulted in the lowest AIC and misclassification rate based on the selected YI. Models J and K had the same misclassification rate but Model K had a slightly lower AIC and higher ROC. Overall the accuracy of phase two multivariate models were between 72% and 79% resulting in an improvement of accuracy compared to the single variable models.

Discussion

The results of this research demonstrated that soil moisture indices are positively associated with leaf rust epidemics over a 20-year time period. If an epidemic occurs, precipitation coupled with temperature and relative humidity combination variables are highly associated with severe leaf rust epidemics in Kansas.

In this analysis, variables summarizing soil moisture conditions in the state and region were positively associated with the ZNDX in the spring (April) and the PDSI in winter (December) and were the most highly associated variables with leaf rust epidemics (Phase One). This is consistent with Eversmeyer and Kramer (1998) who noted the importance of snow cover, temperature and precipitation for the overwintering and survival of leaf rust prior to winter dormancy. Soil moisture in December corresponds to the period of time when winter wheat in

Kansas, Oklahoma and Texas is established and entering dormancy. Soil moisture during this period may stimulate canopy development resulting in microenvironments that could favor the development of water vapor that forms dew from soil moisture, a process known as distillation (Jacobs et al. 1990). When winter wheat is resuming growth, soil moisture may influence leaf rust development in the spring. Moisture during this time period could favor infection processes. Once the canopy closes, the main source of dew is from atmospheric water vapor. The average Palmer moisture anomaly index for Kansas in April (ZNDXKS_04) was the most highly associated variable with leaf rust epidemics of all the variables considered (Table 4.5). The increase in ZNDXKS_04 is indicative of increased moisture conditions across the state. The presence of free moisture has been widely documented in literature to favor leaf rust infections (Chester 1946; deVallielle-Pope et al. 1995; Hogg et al. 1969). Precipitation in October (SumPrec_10) was also highly associated with epidemics and had a high ROC (Table 4.5). October corresponds to the optimum planting date and subsequent establishment of wheat in Kansas. Combining soil moisture conditions and the mean temperature in December resulted in the model with the highest accuracy and best fit for Winter variables (Model F, Table 4.6). In the Fall, Model C (Table 4.6) including the Southern Oscillation Index in September and precipitation in October was the model with the highest accuracy and the best model fit. While climate indices variables have not been traditionally used in plant prediction and forecast model, these variables have been shown to be associated with the wheat diseases Fusarium head blight and stem rust epidemics in the U.S. (Kriss et al. 2012; Scherm and Yang 1995). The SOI in September was negatively associated with leaf rust epidemics. A negative SOI is indicative of below normal sea level pressure between Tahiti and Darwin, Australia and is associated with warmer ocean waters and El Niño patterns if sustained for long periods of time. Warm El Niño phases have been associated with above average fall precipitation in Mexico and along the Gulf Coast of Texas, U.S. (Ropelewski and Halpert 1986). The above normal precipitation associated with the SOI in August and September could influence the oversummering potential of leaf rust in the southern U.S. Soil moisture and relative humidity in April and optimum temperatures in May (Model H, Table 4.6) were the most highly associated variables and resulted in the highest accuracy with leaf rust epidemics. Temperatures between 15 to 20°C have been considered the optimum range for germination and appressorial development of leaf rust (Chester 1946; de Vallavieille-Pope et al. 1995; Hogg et al. 1969).

In contrast to the “critical month” model developed by Chester, we did not find an association of leaf rust epidemics with average monthly temperature greater than 10°C or with the accumulation of hours with temperatures greater than 10°C in any month. One reason for this is that from March to May cooler temperatures may rarely restrict leaf rust development.

For phase two of the analysis, May local weather conditions were the most highly associated variables with severe leaf rust epidemics including SumPrec_05 and SumT1520RH87_05 (Table 4.7). Previous research had identified snow cover, precipitation and temperature variables to be associated with leaf rust epidemics in Kansas (Eversmeyer and Kramer 1998; Eversmeyer and Burleigh 1970; Burleigh et al. 1972). In Europe, temperature and moisture variables were also associated with final leaf rust disease severity (Wiik and Ewaldz 2009).

Single and multivariate models could be useful for developing a disease forecasting system for leaf rust epidemics in Kansas. The variables and models from each time period could be deployed sequentially as a preliminary forecast of leaf rust epidemics based on the fall and winter conditions. The forecast could be updated with the spring models. If regional observations of disease are reported, then Phase two models could be used to monitor the month of May until the model threshold is met for a severe leaf rust epidemic. This could be accomplished by monitoring the accumulation of precipitation and optimum temperatures for leaf rust throughout the critical time for fungicide applications. The variables and models identified in this research, regional communication of disease observations and knowledge of varietal resistance could be used as components for a leaf rust disease forecasting system and aid in fungicide decision making by growers.

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Figures and Tables

Table 4.1 Phase one fall environmental variables selected from boosted regression trees

Group[a]	Variable Code[b]	Description
CLI	NINO4.0A_08	Nino 4.0 temperature anomaly in August
CLI	SOI_08	Southern Oscillation Index in August
CLI	SOI_09	Southern Oscillation Index in September
DRT	SPICRD_11	SPI per CRD in Kansas in November
DRT	ZNDXCRD_10	ZNDX per CRD in Kansas in October
DRT	ZNDXCRD_11	ZNDX per CRD in Kansas in November
DRT	ZNDXKS_11	Average ZNDX in Kansas in November
P	SumPrec_10	Sum of precipitation (mm) in October
P	SumPrecip_10	Number of hours in October with measureable precipitation (mm)
P	SumPrecip_11	Number of hours in November with measureable precipitation (mm)
T	SumT0525_10	Number of hours in October with temperatures between 5 and 25°C
T	SumT1025_10	Number of hours in October with temperatures between 10 and 25°C
T	SumT1030_09	Number of hours in September with temperatures between 10 and 30°C
T	SumT1520_10	Number of hours in October with temperatures between 15 and 20°C
T	SumTG35_08	Number of hours in September with temperatures greater than 35°C
TP	SumT1025Precip_10	Number of hours in October with temperatures between 10 and 25°C and measureable precipitation (mm)
TP	SumT1520Precip_08	Number of hours in August with temperatures between 15 and 20°C and measureable precipitation (mm)
TRH	SumT1520RH87_08	Number of hours in August with temperatures between 15 and 20°C and relative humidity greater than 87%
TRH	SumT1520RH87_09	Number of hours in September with temperatures between 15 and 20°C and relative humidity greater than 87%

[a] Variables were grouped by type of information represented: (DRT) soil moisture indices, (CLI) climate indices, (T) temperature, (RH) relative humidity, (P) precipitation, (TP) combinations of temperature and precipitation and (TRH) combinations of temperature and RH.

[b] Acronyms describe the variables summarizing regional soil moisture indices (ZNDX, PDSI, and SPI), temperature (T), average temperature (MeanT), relative humidity (RH), average relative humidity (MeanRH), and precipitation (P). The number after the variable acronym specifies the value or range used, and "_number" indicates the month (ranging from January (01) to December (12)). For example, SumT0525RH87_05 signifies the sum of hours in May (_05) with temperatures (T) between 5 and 25°C and relative humidity (RH) greater than 87%.

Table 4.2 Phase one winter environmental variables selected by boosted regression trees

Group[a]	Variable Code[b]	Description
CLI	SOI_12	Southern Oscillation Index in December
DRT	PDSIKS_01	Average PDSI in Kansas in January
DRT	PDSIKS_02	Average PDSI in Kansas in February
DRT	PDSIKS_12	Average PDSI in Kansas in December
DRT	PDSIKSOKTX_01	Average PDSI in Kansas, Oklahoma, and Texas in January
DRT	PDSIKSOKTX_02	Average PDSI in Kansas, Oklahoma, and Texas in February
DRT	PDSIKSOKTX_12	Average PDSI in Kansas, Oklahoma, and Texas in December
DRT	SPIKSOKTX_12	Average SPI in Kansas, Oklahoma, and Texas in December
DRT	ZNDXCRD_01	ZNDX per CRD in Kansas in January
LW	SumDewPresence_01	Leaf wetness in January
NDVI	NDVIOK_02	NDVI departure from normal in Oklahoma in February
P	SumPrec_01	Sum of precipitation (mm) in January
P	SumPrec_02	Sum of precipitation (mm) in February
P	SumPrec_12	Sum of precipitation (mm) in December
P	SumPrecip_01	Number of hours in January with measureable precipitation(mm)
T	SumT0525_02	Number of hours in February with temperatures between 5 and 25°C
T	SumT1025_01	Number of hours in January with temperatures between 10 and 25°C
T	SumT1025_02	Number of hours in February with temperatures between 10 and 25°C
T	SumT1520_01	Number of hours in January with temperatures between 15 and 20°C
T	MeanT_02	Mean temperature in February
T	MeanT_12	Mean temperature in December
TP	SumT0525Precip_02	Number of hours in February with temperatures between 5 and 25°C and measureable precipitation (mm)
TRH	SumT1025RH87_02	Number of hours in February with temperatures between 10 and 25°C and relative humidity greater than 87%

[a] See Table 4.1, footnote a.

[b] See Table 4.1, footnote b.

Table 4.3 Phase one spring environmental variables selected by boosted regression trees

Group[a]	Variable Code[b]	Description
DRT	PDSICRD_04	PDSI per CRD in Kansas in April
DRT	PDSIKS_04	Average PDSI in Kansas in April
DRT	PDSIKSOKTX_06	Average PDSI in Kansas, Oklahoma, and Texas in June
DRT	SPICRD_03	SPI per CRD in Kansas in March
DRT	SPICRD_05	SPI per CRD in Kansas in May
DRT	ZNDXCRD_03	ZNDX per CRD in Kansas in March
DRT	ZNDXCRD_04	ZNDX per CRD in Kansas in April
DRT	ZNDXKS_04	Average ZNDX in Kansas in April
DRT	ZNDXKSOKTX_03	Average ZNDX in Kansas, Oklahoma, and Texas in March
NDVI	NDVIKS_06	NDVI departure from normal in Kansas in June
NDVI	NDVIKSOKTX_06	NDVI departure from normal averaged over Kansas, Oklahoma, and Texas in June
NDVI	NDVIOK_03	NDVI departure from normal in Oklahoma in March
NDVI	NDVIOK_06	NDVI departure from normal in Oklahoma in June
NDVI	NDVITX_06	NDVI departure from normal in Texas in June
P	SumPrec_05	Sum of precipitation (mm) in May
RH	MeanRH_04	Mean relative humidity in April
T	SumT0525_05	Number of hours in May with temperatures between 5 and 25°C
T	SumT1520_05	Number of hours in May with temperatures between 15 and 20°C
T	SumTG30_06	Number of hours in June with temperatures greater than 30°C
T	SumTG30_07	Number of hours in July with temperatures greater than 30°C
TP	SumT0525Precip_04	Number of hours in April with temperatures between 5 and 25°C and measureable precipitation (mm)

[a] See Table 4.1, footnote a.

[b] See Table 4.1, footnote b.

Table 4.4 Phase two environmental variables selected by boosted regression trees

Group[a]	Variable Code[b]	Description
P	SumPrec_05	Sum of precipitation (mm) in May
T	SumT1030_05	Number of hours in May with temperatures between 10 and 30°C
T	SumT0525_03	Number of hours in March with temperatures between 5 and 25°C
T	SumT0525_04	Number of hours in April with temperatures between 5 and 25°C
T	SumT1520_04	Number of hours in April with temperatures between 15 and 20°C
T	SumTG10_05	Number of hours in May with temperatures greater than 10°C
TRH	SumT1520RH87_05	Number of hours in May with temperatures between 15 and 20°C and relative humidity greater than 87%
TRH	SumT1025RH87_03	Number of hours in March with temperatures between 10 and 25°C and relative humidity greater than 87%
TRH	SumT0525RH87_03	Number of hours in March with temperatures between 5 and 25°C and relative humidity greater than 87%
TRH	SumT1025RH87_05	Number of hours in May with temperatures between 10 and 25°C and relative humidity greater than 87%

[a] See Table 4.1, footnote a.

[b] See Table 4.1, footnote b.

Table 4.5 Phase one single variable logistic regression models for classifying leaf rust epidemics and non-epidemics in Kansas

Variable[a]	Season	Group	AIC[b]	ROC	Youden Index[c]	TP[d]	TN	FP	FN	Misclassification Rate[e]
SOI_09	Fall	CLI	213.18	0.71	0.55	24	103	10	43	0.29
PDSIKSOKTX_12	Winter	DRT	211.06	0.69	0.32	58	68	45	9	0.30
SPIKSOKTX_12	Winter	DRT	226.76	0.67	0.45	26	97	16	41	0.32
ZNDXKS_04	Spring	DRT	196.69	0.82	0.41	48	85	28	19	0.26
SumPrecip_10	Fall	P	209.92	0.75	0.38	41	86	27	26	0.29
SumPrec_12	Winter	P	231.98	0.64	0.41	25	94	19	42	0.34
MeanRH_04	Spring	RH	202.68	0.77	0.34	53	67	46	14	0.33
SumT0525_05	Spring	T	203.30	0.76	0.50	38	98	15	29	0.24
SumT1520_05	Spring	T	208.51	0.75	0.30	52	63	50	15	0.36
MeanT_12	Winter	T	229.97	0.64	0.39	37	67	46	30	0.42
SumT1025Precip_10	Fall	TP	227.29	0.68	0.31	45	61	52	22	0.41
SumT0525Precip_02	Winter	TP	232.65	0.67	0.35	38	77	36	29	0.36

[a] See Table 4.1, footnote a.

[b] The AIC statistic is used to compare different logistic models. Logistic regression models with smaller values are preferred.

[c] The Youden Index was used as the cut-point for converting model-generated probabilities to the classification of an observation as a leaf rust epidemic or non-epidemic.

[d] There were 180 observations total. TN = true negatives (number of non-epidemics correctly classified) (113 cases); TP = true positives (number of epidemics correctly classified) (67 cases); FN = false negatives (number of epidemics incorrectly classified as non-epidemics); FP = false positives (number of non-epidemics incorrectly classified as epidemics).

[e] The proportion of the total observations that were incorrectly classified.

Table 4.6 Phase one multivariate logistic regression models for classifying leaf rust epidemics and non-epidemics in Kansas

Model ID	Season	Variables[a]	AIC[b]	ROC	Youden Index[c]	TP[d]	TN	FP	FN	Misclassification Rate[e]
A	Fall	SumT1025Precip_10 PDSIKSOKTX_12	211.32	0.72	0.33	57	71	42	10	0.29
B	Fall	SumPrecip_10 SPIKSOKTX_12	202.81	0.78	0.37	45	82	31	22	0.29
C	Fall	SOI_09 SumPrecip_10	195.78	0.81	0.32	49	83	30	18	0.27
D	Winter	PDSIKSOKTX_12 SumT0525Precip_02	207.89	0.76	0.29	61	63	50	6	0.31
E	Winter	MeanT_12 SumPrec_12	227.06	0.69	0.36	43	73	40	24	0.36
F	Winter	PDSIKSOKTX_12 MeanT_12	191.05	0.81	0.37	52	84	29	15	0.24
G	Spring	ZNDXKS_04 SumT0525_05	181.58	0.83	0.33	52	87	26	15	0.23
H	Spring	ZNDXKS_04 SumT1520_05	179.47	0.85	0.35	53	90	23	14	0.21
I	Spring	SumT0525_05 MeanRH_04	188.08	0.81	0.62	36	107	6	31	0.21

[a] See Table 4.1, footnote a.

[b] See Table 4.5, footnote b.

[c] See Table 4.5, footnote c.

[d] See Table 4.5, footnote d.

[e] See Table 4.5, footnote e.

Table 4.7 Phase two single variable logistic regression models for classifying moderate and severe leaf rust epidemics in Kansas

Variable[a]	Group	AIC[b]	ROC	Youden Index[c]	TP[d]	TN	FP	FN	Misclassification Rate[e]
SumPrec_05	P	80.73	0.74	0.53	14	37	3	13	0.24
SumT1030_05	T	74.08	0.79	0.40	18	31	9	9	0.27
SumT1520RH87_05	TRH	74.22	0.80	0.41	18	33	7	9	0.24
SumT1025RH87_03	TRH	74.87	0.80	0.36	18	31	9	9	0.27
SumT0525RH87_03	TRH	76.91	0.77	0.27	21	21	19	6	0.37

[a] See Table 4.1, footnote a.

[b] See Table 4.5, footnote b.

[c] See Table 4.5, footnote c.

[d] There were 67 observations total. TN = true negatives (number of moderate epidemics correctly classified) (40 cases); TP = true positives (number of severe epidemics correctly classified) (27 cases); FN = false negatives (number of severe epidemics incorrectly classified as moderate epidemics); FP = false positives (number of moderate epidemics incorrectly classified as severe epidemics).

[e] See Table 4.5, footnote e.

Table 4.8 Phase two multivariate logistic regression models for classifying moderate and severe leaf rust epidemics in Kansas

Model ID	Variables[a]	AIC[b]	ROC	Youden Index[c]	TP[d]	TN	FP	FN	Misclassification Rate[e]
J	SumPrec_05 SumT1520RH87_05	73.68	0.81	0.40	20	33	7	7	0.21
K	SumPrec_05 SumT0525RH87_03	70.30	0.82	0.51	18	35	5	9	0.21
L	SumPrec_05 SumT1025RH87_03	71.30	0.82	0.46	19	29	11	8	0.28
M	SumT1030_05 SumPrec_05	71.43	0.83	0.44	18	33	7	9	0.24

[a] See Table 4.1, footnote a.

[b] See Table 4.5, footnote b.

[c] See Table 4.5, footnote c.

[d] See Table 4.7, footnote d.

[e] See Table 4.5, footnote e.

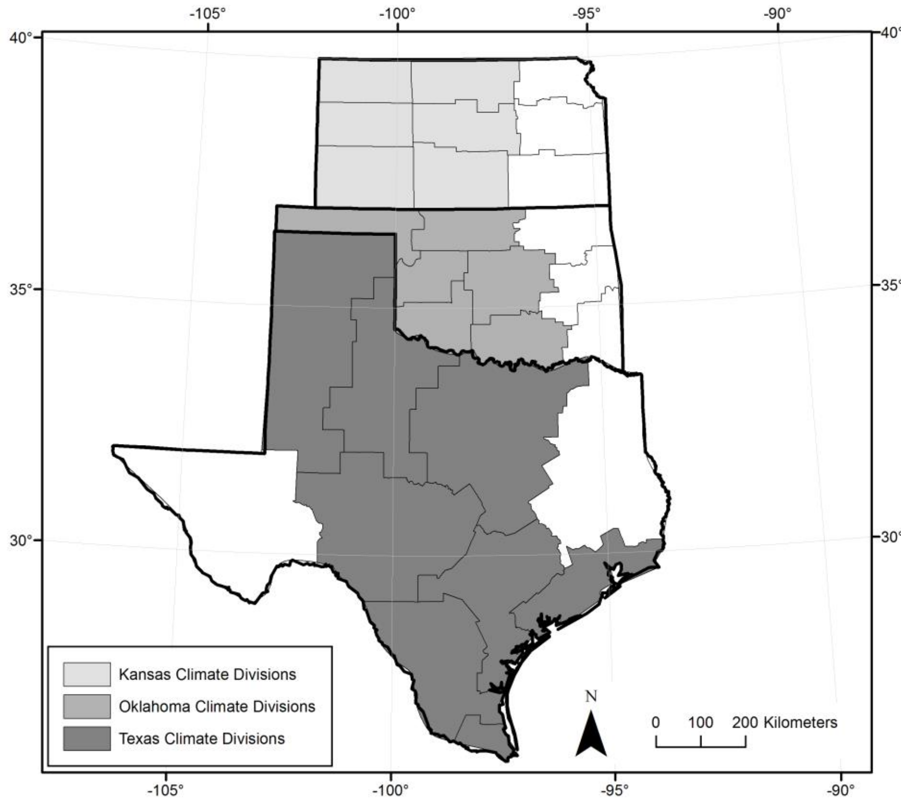

Figure 4.1 Climate divisions in Kansas, Oklahoma and Texas. Within Kansas, crop reporting districts and climate divisions are the same spatially. Gray areas indicate climate divisions over which regional soil moisture conditions and NDVI were averaged.

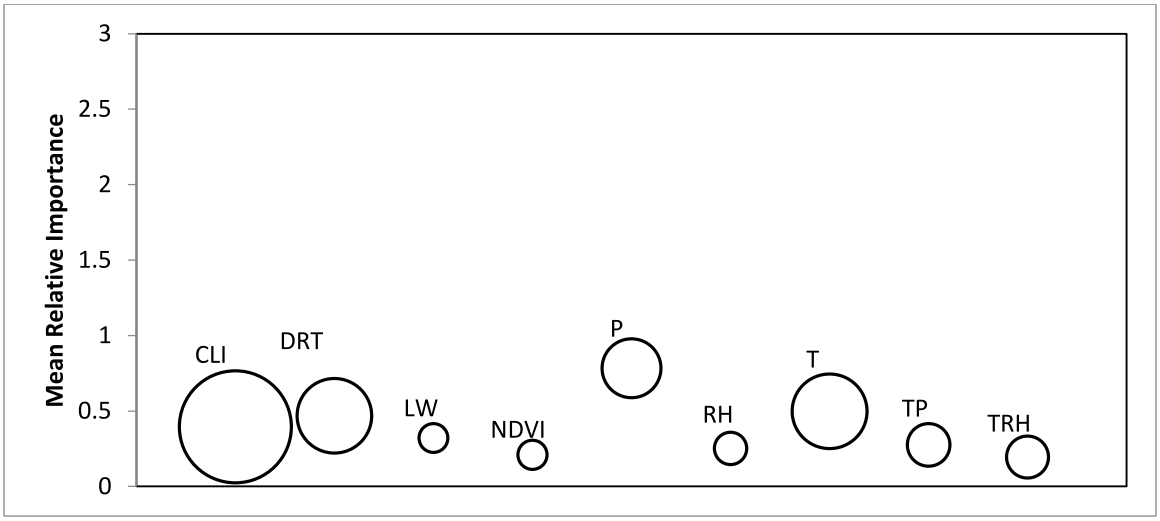

Figure 4.2 The fall mean relative influence and variable groups most associated with leaf rust epidemics and non-epidemics in Kansas. The size of the bubble indicates maximum relative influence from the boosted regression tree analysis per variable group. Groups represent climate indices (CLI), soil moisture indices (DRT), leaf wetness (LW), normalized difference vegetation index (NDVI), precipitation (P), relative humidity (RH), temperature (T), combined temperature and precipitation conditions (TP), and combined temperature and relative humidity conditions (TRH).

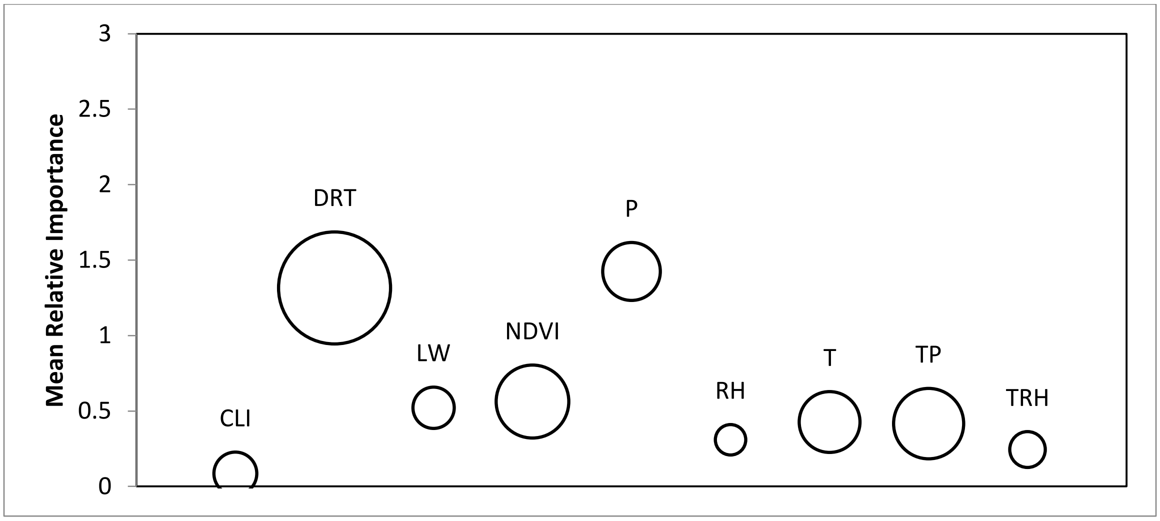

Figure 4.3 The winter mean relative influence and variable groups most associated with leaf rust epidemics and non-epidemics in Kansas. The size of the bubble indicates maximum relative influence from the boosted regression tree analysis per variable group. Groups represent climate indices (CLI), soil moisture indices (DRT), leaf wetness (LW), normalized difference vegetation index (NDVI), precipitation (P), relative humidity (RH), temperature (T), combined temperature and precipitation conditions (TP), and combined temperature and relative humidity conditions (TRH).

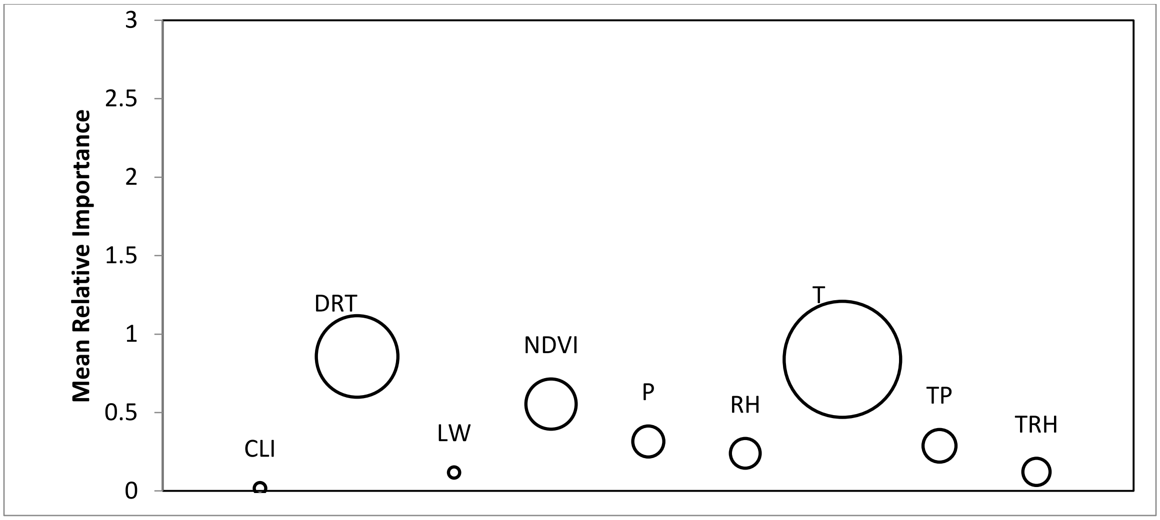

Figure 4.4 The spring mean relative influence and variable groups most associated with leaf rust epidemics and non-epidemics in Kansas. The size of the bubble indicates maximum relative influence from the boosted regression tree analysis per variable group. Groups represent climate indices (CLI), soil moisture indices (DRT), leaf wetness (LW), normalized difference vegetation index (NDVI), precipitation (P), relative humidity (RH), temperature (T), combined temperature and precipitation conditions (TP), and combined temperature and relative humidity conditions (TRH).

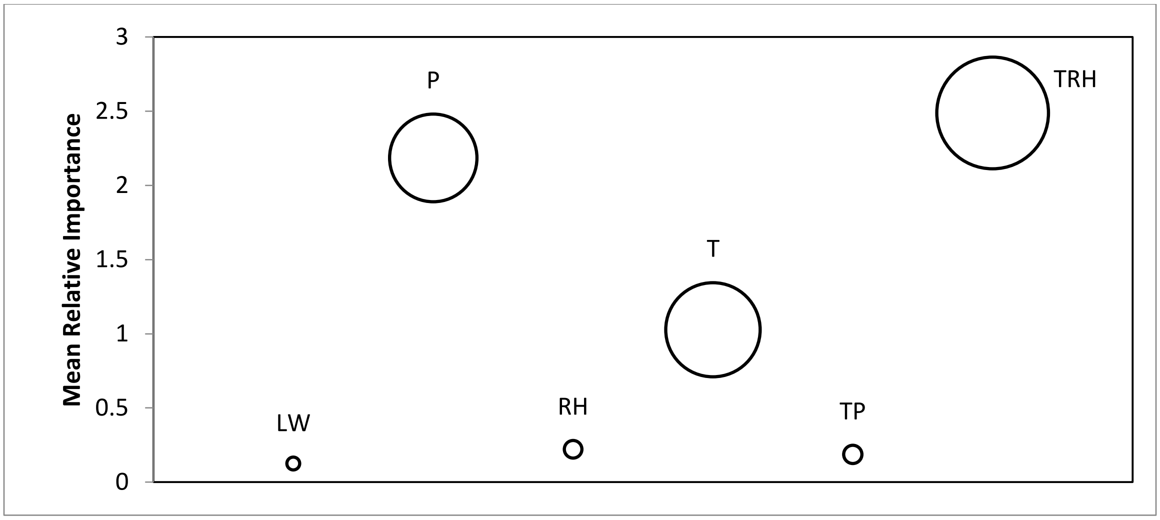

Figure 4.5 The mean relative influence and variable groups most associated with severe and moderate epidemics in Kansas. The size of the bubble indicates maximum relative influence from the boosted regression tree analysis per variable group. Groups represent leaf wetness (LW), precipitation (P), relative humidity (RH), temperature (T), combined temperature and precipitation conditions (TP), and combined temperature and relative humidity conditions (TRH).

Chapter 5 - Conclusions

The overall objectives of this research were to identify environmental conditions that favor or restrict leaf rust and stripe rust epidemics and to begin evaluating these variables in a predictive framework. The variables identified through this analysis could be used to predict epidemics and severe epidemics in combination with disease observations in the southern Great Plains and the growers' knowledge of varietal resistance in a disease forecast system.

In Chapter 2, local weather conditions that favor leaf rust infection events were evaluated in an outdoor field environment. Prior to this research, these conditions had never been explored in ambient field conditions. The results of this research demonstrated that leaf rust infection events are correlated with the number of hours at optimum temperatures and high relative humidity and leaf wetness duration over the exposure period. This research was used to develop variables for evaluating the association of environmental conditions with regional leaf rust yield loss in Kansas. An additional experiment could include evaluating more isolates on susceptible varieties in ambient field environments.

Monthly environmental conditions that are associated with stripe rust and leaf rust epidemics were identified in Chapters 3 and 4. The results of this research found soil moisture conditions were highly associated with stripe rust and leaf rust epidemics. Soil moisture in April was the most highly influential variable with leaf rust epidemics. Soil moisture conditions in the fall and winter, which corresponds to the planting, establishment and dormancy for winter wheat in Kansas, were strongly associated with stripe rust epidemics. Optimal temperatures in May were associated with severe stripe rust and leaf rust epidemics. For stripe rust the optimum temperature range was 7 to 12°C. Temperatures between 15 to 20°C were associated with severe leaf rust epidemics when coupled with high relative humidity (>87%). The single variable and multivariable models could be combined with field-level disease observations and an understanding of varietal reaction to stripe rust and leaf rust as part of an operational disease forecasting system in Kansas. However, before this can happen it will be important to test and validate the potential models with additional data. The models presented in this project were validated using cross validation techniques that are useful for testing models with limited data. More confidence in the functionality of the models could be established by testing with new data. In addition to validating the models with untested data, an analysis of the costs of misclassification errors could provide insight to the risk associated with incorrect predictions. This analysis focused on yield losses in susceptible varieties. In the future, it may be possible to incorporate genetic resistance into the models to determine yield loss by varietal resistance. Resistance could be incorporated as a continuous or ordinal variable to further define regional yield losses.

Overall the variables and models identified in this research are associated with leaf rust and stripe rust epidemics and can potentially be released in a disease forecasting system after further validation. These models could help inform growers of their risk of the diseases in their region and encourage timely scouting and fungicide decisions.