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COMPREHENSIVE RESEARCH ON RICE
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PROJECT TITLE: Investigations on Rice Blast Disease in California

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OBJECTIVES AND EXPERIMENTS CONDUCTED BY LOCATION TO ACCOMPLISH
OBJECTIVES:

The major objective of this project is to gain an understanding of the biology of Rice Blast disease in California and develop information that will be critical in developing disease management strategies. During the 1998 season we emphasized studies on pathogen diversity, identification of possible overwintering inoculum sources, monitoring spore production in fields, analysis of weather data and use of fungicides for control of blast.

Objectives of the proposed research were:

- 1) Identify genetic and pathotype diversity of Pyricularia grisea isolates present in California
- 2) Investigate and identify possible overwintering sources for P. grisea in California
- 3) Determine relative abundance and temporal presence of airborne inoculum and identify field conditions under which airborne inoculum is produced
- 4) Determine effects of grower practices on rice blast disease development in California

Objective1 - Identify genetic and pathotype diversity of Pyricularia grisea isolates present in California. DNA fingerprinting was conducted using the MGR-586 probe in RFLP (random fragment length polymorphism) analysis to determine the variation in the genetic structure of the California population of P. grisea.

Pathotype testing was done by inoculating differential cultivars with isolates of P. grisea to determine their race. The international standard cultivars were used with a few accepted substitutions which give the same disease reactions. Cultivars used include Katy, Zenith, NP125, Usen, Dular, Jefferson, CI 8970-S and M-201. Lemont and Mars were also included because of

their ability to separate out races which occur in the southern U.S. but are not differentiated by the international standards.

Distribution of the blast disease was monitored by field examination and laboratory confirmation of samples collected that were suspected to be blasted throughout the 1998 rice growing season. P. grisea isolates were collected during the 1998 growing season and single spore isolations were made for future DNA fingerprinting and race typing. These isolates were collected throughout the geographic distribution of the rice blast disease in Colusa, Glenn and Sutter Counties.

Objective 2 - Investigate and identify possible overwintering sources for P. grisea in California. Surveys for possible infections of alternate hosts around and in rice fields were conducted prior to and throughout the 1998 growing season. Suspect lesions on weeds were collected and taken to the laboratory for culturing and identification of the cause of the lesions. Residue was collected from fields where blast was known to have occurred in 1997 prior to preparation of the ground for the 1998 growing season and examined for the blast pathogen in the laboratory by standard culture techniques. Panicles and tiller nodes were collected for culturing in a Colusa County field from areas that were stripper harvested and conventionally harvested.

Objective 3 - Determine relative abundance and temporal presence of airborne inoculum and identify field conditions under which airborne inoculum is produced. Airborne inoculum was monitored at four sites (1-Colusa Co., 2-Glenn Co., 1-Butte Co.) during the 1998 growing season by placing spore trapping equipment at the edge of the field. Spores were sampled for 30 minutes each day at either 2 am or 6 am. Sampling rods were changed every two days and brought to the lab for microscopic observation. The presence and relative density of spores at the time of sampling were determined from this observation.

Microclimate data including temperature, relative humidity, leaf wetness, precipitation, wind speed and wind direction were collected in six rice fields in Colusa, Glenn and Butte Counties. Data were collected by in-field stations, manufactured by Adcon Telemetry, Inc., that were solar powered and transmitted data as a fifteen minute average to a base station by radio signal. One weather station was located at each of the sites where spore trapping equipment was operating.

Objective 4 - Determine effects of grower practices on rice blast disease development in California. Chemical control of blast with fungicides is a common practice in many areas where the blast disease occurs. Small field trials with 10'x20' ft plots and three replications were carried out to determine the efficacy of Quadris at three sites in Colusa Co. and three sites in Glenn Co. Two rates of Quadris at one and or two applications at two maturity stages were tested. Evaluation of the efficacy of Quadris was also made in one Colusa Co. field in which a commercial application was made by the grower. Percent neck blast was determined by random samples and counts within each plot. Yields and milling quality were determined and compared between treatments where possible.

The effect of different rates of nitrogen on the incidence of blast disease was tested at two different sites within one field in Colusa Co. Each plot was 10'x40' and each treatment was replicated four times. Nitrogen was applied as pre-plant ammonium sulfate at rates of 100, 125, 150, 175 and 200 lbs N/ac. Percent neck blast was determined by random samples and counts within each plot. Yields were determined and compared between treatments.

The relative susceptibility of California cultivars to blast disease was compared in one of the UCCE regional trials in Colusa Co. by taking random samples to determine an estimate for the percent neck blast in each replication for each cultivar.

SUMMARY OF 1998 RESEARCH (MAJOR ACCOMPLISHMENTS) BY OBJECTIVE:

Objective1 - Identify genetic and pathotype diversity of Pyricularia grisea isolates present in California. A small amount of seed of each of the international standard differentials was obtained and planted in the greenhouse for production of seed which could be use for pathotype testing. Seed was harvested at the end of the summer and pathotype testing was conducted on over 50 P. grisea isolates from the 1996 and 1997 growing season and is being continued with other isolates. Pathotype testing indicates that each of these isolates belongs to the race IG-1. DNA fingerprinting of approximately 60 isolates from the 1996 growing season indicate that these isolates are very closely related genetically suggesting that they may all be from the same source. IG-1 was the predominant race in Arkansas and Texas about 20 years ago, but the DNA fingerprinting patterns for the California isolates are not closely related to the reference type isolates from that era.

DNA has been extracted from isolates collected in 1996 and 1997 and is currently being extracted from over 100 isolates collected from 24 fields in 1998. DNA fingerprinting and pathotype testing of these isolates is continuing through the winter and spring. All have been individually purified genetically by isolating single spores and growing stock cultures for these studies. This process is very labor intensive but when completed should provide an accurate determination of the race situation in California.

The blast disease appeared to have a much later start in 1998 than in the previous year. Leaf blast was first observed around mid-July in 1998 as compared to mid-June in 1997. The overall distribution of the disease was about the same as in 1997 with disease being observed in fields in Colusa, Glenn and Sutter Counties. We observed and confirmed the disease in fields West of Interstate 5 in Glenn and Colusa Counties, East of the Sacramento River in Glenn County and North of Hahn Road in Colusa County. Blast was observed in Sutter County in the same areas it was confirmed in 1997. Leaf blast did not seem to be as severe in 1998 as it has been in the previous two years. There were no large areas seen within fields where plants had been killed by leaf blast early in the season. Blast was much more difficult to spot early in the season because of the lack of these obvious signs and the sporadic nature of the disease. The overall severity of blast during 1998 appears to have been less than in previous years but the number of fields affected did not seem to be reduced. This may have been related to the late planting time of many fields, cool spring temperatures, effects of the wet winter on overwintering inoculum or the lack of plantings of the quite susceptible cultivar M-201. Since the discovery of blast in 1996, the acreage planted to M-201 in areas affected by blast has dramatically decreased. The observations of blast disease in California over the last three years are giving us a demonstration of the unpredictable and temperamental nature of this disease.

Objective 2 - Investigate and identify possible overwintering sources for P. grisea in California. Leaf spots from many weeds and grass species around rice fields were examined for the presence of P. grisea. The blast pathogen was not found to be causing leaf spots or lesions on any of the non-rice plant material examined prior to or throughout the 1998 growing season.

The blast pathogen is easily isolated from crop residue soon after harvest. The fungus sporulates readily in culture from infected nodes of stems and panicles when subjected to conditions that favor sporulation. When residue was collected in the Spring of 1998 from a field severely affected by rice blast in 1997 the pathogen was easily recovered from this material. Symptomatic panicle and tiller nodes often yielded sporulation when placed in humid chambers. Residue from stripper harvested areas and conventionally harvested areas both harbored the rice blast fungus prior to preparation of the ground for planting.

Objective 3 - Determine relative abundance and temporal presence of airborne inoculum and identify field conditions under which airborne inoculum is produced. Airborne inoculum was monitored at four sites (1-Colusa Co., 2-Glenn Co., 1-Butte Co.) during the 1998 growing season. No blast was found at two of the sites in Glenn and Butte County and no P. grisea spores were trapped at either of these sites. The other site in Glenn County had very little blast pressure but there were fields in the area where the disease was present. Spores were only trapped at very low concentrations between the period of 8/2/98 through 8/6/98 at this site. The concentration of spores during that period was approximately 1 spore/m³ which is basically our lower detection limit. The results of the spore trapping at this site would indicate that there was not enough inoculum in the immediate vicinity for a severe outbreak of blast in this field. That, in fact, was what happened with disease incidence remaining close to 0 throughout the season.

The field in Colusa County was a different story. Spore trapping began in mid-July and low spore densities were being detected at 2am in that area. On 8/4/98 we began sampling spores at 6am and found that more spores were being released at this time. Throughout the season, spore densities were generally much higher at 6am than at 2am at this site. Spores were trapped during each sampling period between 8/4/98 and 9/24/98 at the 6am sampling time. Spore concentrations ranged from 0.77 to 279 spores/m³ with a mean of 106.6 spores/m³ and a median of 101.2 spores/m³. This information indicated that there was sufficient inoculum in the air to result in a significant amount of rice blast disease in the immediate area. Early leaf blast and fairly heavy neck blast incidence at the end of the season were observed in this field. In fact, this spore trap was positioned between the checks which contained the Site 1 nitrogen trial in Table 7 and the fungicide trial in Table 2.

When blast was first found in California there was considerable speculation that it was due to unusual weather during the 1996 season. After living with rice blast in California for three years it is apparent that this is not the case. Microclimate data including temperature, relative humidity, leaf wetness, precipitation, wind speed and wind direction were collected in six rice fields in Colusa, Glenn and Butte Counties. Data were collected by in-field stations, manufactured by Adcon Telemetry, Inc., that were solar powered and transmitted data as a fifteen minute average to a base station by radio signal. Four of these weather stations were located at each of the sites where spore trapping equipment was operating. There does not seem to be any great differences in weather data among sites during the spore trapping periods that would explain why we have disease in some areas and not in others. The weather data do

indicate that conditions are for the most part permissive for blast disease development during our growing season. Most days had high relative humidity periods and leaf wetness periods that would be more than adequate for disease development. Our cool night temperatures may slow disease development and it is unclear how the cool wet spring affected disease incidence and overwintering inoculum. Weather stations were not operational until well after planting so it is unclear whether early climatic conditions were favorable or unfavorable to blast. Regardless of the weather conditions, there is still the biological requirement that the pathogen be present to cause the disease. There must be favorable climatic conditions, a susceptible host and the pathogen all acting in concert to bring about the development of disease.

Objective 4 - Determine effects of grower practices on rice blast disease development in California. Although overall blast disease pressure appeared to be lighter in 1998, the selection of locations for fungicide trials could not have been better. Small field trials with 10'x20' ft plots and three replications were carried out to determine the efficacy of Quadris at three sites in Colusa Co. and three sites in Glenn Co. Two rates of Quadris at one and or two applications at two maturity stages were tested. Evaluation of the efficacy of Quadris was also made in one Colusa Co. field in which a commercial application was made by the grower.

There was a good response to Quadris with respect to neck blast incidence in all treatments with highest yields in treatments 2 and 4 at Site 1 (Table 1). There were no significant differences in milling quality among the treatments.

Table 1 - Effect of Application Time and Rate of Quadris for Control of Rice Blast Disease - Site 1 - CW, Colusa County - 1998

Treatment - Rate -lbs ai/ac BS = Boot Split; H = Heading	Percent Neck Blast	Yield 14% lbs/ac	% Total Rice	% Head Rice
1 - 0.2 - 50% BS	0.64 c	8012 ab	72.3	68.4
2 - 0.2 - 60-90% H	2.49 b	8563 a	73.0	69.9
3 - 0.2 - 50% BS + 60-90% H	1.01 c	8005 ab	72.5	69.2
4 - 0.3 - 50% BS	0.52 c	8546 a	71.8	68.1
5 - 0.3 - 60-90% H	1.86 bc	7632 b	72.0	68.7
6 - 0.3 - 50% BS + 60-90% H	0.73 c	8117 ab	72.7	69.6
7 - 0.0 - Control	4.00 a	7933 ab	72.1	68.4

Means with the same letter within each column are not significantly different
Cultivar M-202; Three replications per treatment

There was a good response to Quadris with respect to neck blast incidence with highest reductions in the two application treatments 3 and 6 at Site 2 (Table 2). There were some large yield differences although these were not statistically significant due to variation between replications. There were some slight milling quality differences among the treatments in this trial.

Table 2 - Effect of Application Time and Rate of Quadris for Control of Rice Blast Disease - Site 2 - CE, Colusa County - 1998

Treatment - Rate -lbs ai/ac BS = Boot Split; H = Heading	Percent Neck Blast	Yield 14% lbs/ac	% Total Rice	% Head Rice
1 - 0.2 - 50% BS	2.69 ab	7429	71.2 a	67.2 a
2 - 0.2 - 60-90% H	2.82 ab	8569	72.1 bc	68.1 abc
3 - 0.2 - 50% BS + 60-90% H	0.73 b	7839	71.8 ab	68.7 bcd
4 - 0.3 - 50% BS	2.09 b	7843	71.8 ab	67.7 ab
5 - 0.3 - 60-90% H	2.09 b	7984	72.3 bc	69.1 cd
6 - 0.3 - 50% BS + 60-90% H	0.00 b	7533	72.7 c	69.8 d
7 - 0.0 - Control	5.25 a	7316	71.9 ab	68.0 abc

Means with the same letter within each column are not significantly different
Cultivar M-202; Three replications per treatment

There was a good reduction in neck blast with the application of Quadris and yields were slightly higher when compared to the control at Site 3 (Table 3).

Table 3 - Effect of Application Time and Rate of Quadris for Control of Rice Blast Disease - Site 3 - ST, Colusa County - 1998

Treatment - Rate -lbs ai/ac BS = Boot Split; H = Heading	Percent Neck Blast	Yield 14% lbs/ac
1 - 0.2 - 50% BS	0.75 b	8164
2 - 0.2 - 60-90% H	2.02 ab	7867
3 - 0.2 - 50% BS + 60-90% H	1.00 b	7807
4 - 0.3 - 50% BS	0.84 b	8134
5 - 0.3 - 60-90% H	2.02 ab	8141
6 - 0.3 - 50% BS + 60-90% H	0.80 b	8019
7 - 0.0 - Control	3.41 a	7647

Means with the same letter within each column are not significantly different
Cultivar M-202; Three replications per treatment

The largest reduction in incidence of neck blast was gained with the two applications of Quadris in treatments 3 and 6 at Site 4 (Table 4). There appears to have been some beneficial effects to yield with the fungicide application, especially at the higher rates. Although there are some major differences in milling quality, these were not statistically significant due to large variations among the replications.

Table 4 - Effect of Application Time and Rate of Quadris for Control of Rice Blast Disease - Site 4 - SP, Glenn County - 1998

Treatment - Rate -lbs ai/ac BS = Boot Split; H = Heading	Percent Neck Blast	Yield 14% lbs/ac	% Total Rice	% Head Rice
1 - 0.2 - 50% BS	5.10 ab	7208	70.9	66.6
2 - 0.2 - 60-90% H	3.58 ab	7243	70.8	61.2
3 - 0.2 - 50% BS + 60-90% H	2.36 b	7204	69.8	55.5
4 - 0.3 - 50% BS	5.35 ab	6818	69.5	57.9
5 - 0.3 - 60-90% H	5.98 ab	7471	69.2	59.2
6 - 0.3 - 50% BS + 60-90% H	3.78 ab	7516	70.7	54.6
7 - 0.0 - Control	7.72 a	6979	70.6	62.2

Means with the same letter within each column are not significantly different
Cultivar Cal Mochi-101; Three replications per treatment

Site 5 had a very low incidence of neck blast and there were no significant differences between treatments with regard to percent neck blast, yield or milling quality (Table 5).

Table 5 - Effect of Application Time and Rate of Quadris for Control of Rice Blast Disease - Site 5 - CE, Glenn County - 1998

Treatment - Rate -lbs ai/ac BS = Boot Split; H = Heading	Percent Neck Blast	Yield 14% lbs/ac	% Total Rice	% Head Rice
1 - 0.2 - 50% BS	0.92 a	7092 a	70.9	64.8
2 - 0.2 - 60-90% H	0.32 a	7086 a	69.4	66.1
3 - 0.2 - 50% BS + 60-90% H	0.00 a	7665 a	71.1	66.2
4 - 0.3 - 50% BS	0.57 a	7294 a	70.0	66.6
5 - 0.3 - 60-90% H	1.73 a	7741 a	70.4	65.6
6 - 0.3 - 50% BS + 60-90% H	0.00 a	7436 a	70.6	65.9
7 - 0.0 - Control	0.25 a	7700 a	68.7	64.9

Means with the same letter within each column are not significantly different
Cultivar M-202; Three replications per treatment

There was a very low level of neck blast at Site 6 and the trial was harvested by the grower before the small plot harvested could be scheduled (Table 6). With the low level of disease pressure we probably did not lose more than time and effort at this site.

Table 6 - Effect of Application Time and Rate of Quadris for Control of Rice Blast Disease - Site 6 - ZO, Glenn County - 1998

Treatment - Rate -lbs ai/ac	Percent Neck Blast
BS = Boot Split; H = Heading	
1 - 0.2 - 50% BS	0.47
2 - 0.2 - 60-90% H	1.20
3 - 0.2 - 50% BS + 60-90% H	0.80
4 - 0.3 - 50% BS	0.55
5 - 0.3 - 60-90% H	1.54
6 - 0.3 - 50% BS + 60-90% H	1.86
7 - 0.0 - Control	1.13

Means with the same letter within each column are not significantly different

Cultivar M-202; Three replications per treatment

The grower applied fungicide trial consisted of two checks totaling approximately 33 acres being treated at 0.2 lbs a.i./ac Quadris at 50% heading. The cultivar was M-401 and treated checks were harvested separately to allow comparisons of yield and milling quality among treated and untreated areas. The incidence of neck blast was very low throughout the entire field. Plants in treated checks appeared to be greener and Aggregate Sheath Spot severity was less than in untreated checks. The leaves of untreated plants senesced more rapidly than those that were treated with Quadris. The treated portion of the field yielded 980 lbs/ac more than the untreated portion and there was no significant difference in milling quality.

The effects of nitrogen rate on neck blast were evaluated in two different checks within the same field in Colusa County. Higher rates of nitrogen resulted in higher incidences of neck blast and lower yields in each of these trials (Table 7). Rice plants are known to be more susceptible to the blast pathogen in the presence of excess nitrogen. The differences in percent neck blast between the two checks demonstrates the erratic and unpredictable nature of this disease.

Table 7 - Effects of Nitrogen Rate on Neck Blast - Colusa Co.

N lbs/ac	Site 1		Site 2	
	% Neck Blast	Yield lbs/ac	% Neck Blast	Yield lbs/ac
100	32	6943	7.8	7537
125	46	6750	7.7	7540
150	47	6367	10.7	7696
175	46	6416	12.9	7069
200	47	6349	12.9	7035

N applied as pre-plant ammonium sulfate

Cultivar M-202; 3 replications per treatment

Sites 1 & 2 were located in different checks of the same field in Colusa Co.

Relative susceptibility of California cultivars to blast disease was investigated at the statewide cultivar evaluation trial in Colusa County where blast was present. This trial provided the opportunity to determine the relative susceptibility of the currently grown cultivars and advanced lines from the breeding program. The percent neck blast of currently grown California cultivars from the trial in 1998 and the two trials evaluated in 1997 are given in Table 8.

Table 8 - Relative Susceptibility of California Cultivars to Blast Disease Over Two Seasons (Percent Neck Blast)

Cultivar	Colusa 1998	Colusa 1997	Glenn 1997
M-201	6.10 bcde	32.38 a	6.29 bcd
M-202	4.20 bcde	10.39 defgh	4.56 bcdef
M-204	3.30 bcde	17.96 cd	4.05 bcdef
L-203	2.60 cde	20.59 bc	3.60 bcdef
S-102	2.20 de	16.64 cde	3.37 bcdef
CM-101	2.00 de	4.25 gh	0.37 f
S-201	1.70 de	2.80 h	1.64 def
M-103	1.50 de	4.16 gh	2.99 bcdef
L-204	0.90 e	21.13 bc	4.52 bcdef

Means with the same letter within each column are not significantly different
Four replications per cultivar

The disease pressure in the 1998 variety trial was similar to that of the 1997 Glenn County trial. The rankings of the cultivars based on relative susceptibility to rice blast were quite consistent over the three trials and two years. The only deviation between years was that L-204 appeared to be quite susceptible in the 1997 trials whereas it ranked lowest in the 1998 trial based on percent neck blast. Data gathered from these variety trials over the last two years agree with observations in growers' fields that M-201 appears to be our most susceptible cultivar. The fact that the percent neck blast differed between the sites and years on the same cultivars demonstrates that even though a cultivar is susceptible, it must be in a vulnerable stage of development at the same time conditions allow formation and movement of spores to infect the neck nodes. Since all were planted on the same date, some of the differences noted may be due to different stages of maturity at the time factors were favorable for infection and some lines may have escaped infection in these trials.

PUBLICATIONS OR REPORTS:

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Scardaci, S.C., R.K. Webster, C.A. Greer, J.E. Hill, J.F. Williams, R.G. Mutters, D.M. Brandon, K.S. McKenzie and J.J. Oster. 1997. Rice Blast: A New Disease in California. Agronomy Fact Sheet 1997-2. University of California, Davis.

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CONCISE GENERAL SUMMARY OF CURRENT YEAR'S RESULTS:

Studies to date on the Blast disease show that there is only one known race of *P. grisea* currently in California, IG-1. DNA fingerprints indicate that the isolates typed thus far are remarkably homogeneous from a genetic standpoint. IG-1 was common in Texas and Arkansas about 20 years ago. It is also known to occur in Japan. A knowledge of the race or races of *P. grisea* that occur in California is essential for the rice breeders in selecting parents for crosses and screening in attempts to produce cultivars with improved resistance to blast.

The blast disease distribution area in 1998 was similar to that of 1997 but with severity appearing to be less in most fields. Blast was observed in Colusa, Glenn and Sutter Counties. Our first observation of leaf blast was in mid-July 1998 which was about one month later than observed in 1997. It appears that the blast disease got a later start in 1998 which may account for the lower apparent disease incidence and lack of severe leaf blast outbreaks.

The blast pathogen is easily recovered from residue during the fall after harvest. As the overwintering season progresses, saprophytes on the residue complicate isolation of *P. grisea* but it still can be recovered in the spring just prior to planting. *P. grisea* was recovered from residue in areas which had been stripper harvested as well as conventionally harvested just prior to preparation of the ground for planting. Observations in the field, particularly of leaf blast, suggested that residue is the primary source of initial inoculum for early occurrences of leaf blast.

Leaf spots from many weeds and grass species around and in rice fields were examined for the presence of *P. grisea*. The blast pathogen was not found to be causing leaf spots or lesions on any of the non-rice material examined prior to and throughout the 1998 season.

Examination of weather data indicated that the initial occurrence of blast in the Sacramento Valley was not due solely to unusual weather conditions during the 1996 season. Weather in the Sacramento Valley appears to be permissive for the occurrence of blast and there are a few periods of time during the growing season that it becomes more favorable. Besides favorable conditions and a susceptible host, there must be inoculum of the pathogen present for the occurrence of disease. The spore trapping experiments showed that determining the inoculum pressure may be an effective way of predicting the risk of neck blast under conducive climatic conditions.

Trials to determine the efficacy of Quadris, for the control of the neck blast phase of the disease indicate that Quadris can effectively minimize losses in total yield. Additional study is

needed to assure that this expensive control option is used at the proper times and rates to assure that benefits are realized.

Nitrogen fertilization trials reiterated the importance of using only the necessary amount of nitrogen for optimizing yield of the crop. Too much nitrogen results in a plant that is much more susceptible to blast and the probability of reduction in yields.

Evaluation of regional cultivar trials showed that of our California cultivars, M-201 is the most susceptible to the blast disease with the others showing a range of susceptibility. The results of this trial were quite similar to those evaluated in 1997. These results are encouraging since they indicate that there are differences regarding susceptibility to the blast disease in the genetic base of our California breeding program and that improvements in resistance are possible. These results also suggest that the lack of M-201 plantings may be partially responsible for less severe blast outbreaks over the last year.