

ANNUAL REPORT  
COMPREHENSIVE RESEARCH ON RICE  
January 1, 1993 - December 31, 1993

PROJECT TITLE: Cause and Control of Rice Diseases

STATUS OF PROPOSAL: Continuing

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LEVEL OF 1992 FUNDING: 43,438

OVERALL OBJECTIVES AND STATUS OF THE PROJECT:

The primary objective of this project is to understand the biology of rice diseases that occur in California and to develop methods for their control. Previous work has clearly shown the relationship of rice residue management methods and the epidemiology of rice diseases, mainly stem rot and aggregate sheath spot. Both of these diseases have disease cycles that are dependant on overwintering inoculum in the form of sclerotia to initiate disease the following year. Also, both pathogens, [Sclerotium oryzae and R.oryzae sativae] are known to overwinter and increase in rice residue. These facts have been the basis for the beneficial aspects of burning rice residue in the past in regard to rice disease management.

Also, extensive studies have shown that stem rot and aggregate sheath spot severity is affected by nitrogen fertilization practices, stand density and cultivar selection. Although manipulation of these can minimize loss to diseases, the most effective control has been the destruction of overwintering inoculum in the residue by open field burning.

It is now apparent that burning of residue will not be available to all growers. As a result, growers are exploring different methods and schedules for managing the straw after harvest. We have been studying many of these approaches with the primary objectives of determining their affects on epidemiology of rice diseases and also on changes in the populations of other organisms that affect either the viability of rice pathogen inoculum or their persistance in incorporated rice straw.

We have collected microrganisms from straw and sclerotia in the various management treatments compared to determine if there are differences in survival and build up of rice pathogens. Past studies have identified organisms that are mycoparasitic to

sclerotia of S. oryzae and greenhouse studies have shown that some of these organisms are effective in limiting stem rot disease severity in greenhouse tests. Field tests where these organisms were added in attempts to control disease were not encouraging. The status of many of these ongoing studies is summarized in our past reports.

The specific objectives for this year are derived from the previous results that we view as the most promising avenues of continued study in attempts to devise disease management strategies.

#### OBJECTIVES FOR 1993 AND EXPERIMENTS TO ACCOMPLISH OBJECTIVES:

- [1] (a) Continued monitoring populations of fungi known to reduce viability of sclerotia of pathogens and disease incidence in various residue management systems being attempted in various growers fields. These studies are necessary to determine if shifts in microorganism populations are occurring due to various management practices.
- (b) We continued to study the occurrence and severity of stem rot (S. oryzae) and aggregate sheath spot (R. oryzae sativae) under various residue management treatments. Data was collected from experimental sites in Butte, Colusa, and Sutter Counties where effects of incorporation of residue are being studied.
- [2] Further screening and greenhouse experiments to test rates and time of application of microorganisms showing potential for biomanagement of diseases. S. hydrophylum, R. oryzae sativae and the Ascochyta species have shown effective control of stem rot in the greenhouse but attempts in the field have been disappointing. We studied different methods of formulating and applying these to attempt to find ways that may enhance their effectiveness.
- [3] Evaluate disease reactions of different cultivars in fields where residue has been incorporated.
- [4] Evaluate germplasm collections in an attempt to identify new, more disease resistant, germplasm for use by breeders in efforts to produce increased resistance in cultivars to stem rot and aggregate sheath spot.

Field research in 1993 was conducted in growers fields in Butte, Colusa and Sutter Counties. Laboratory and greenhouse studies were conducted in University of California facilities at Davis.

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**SUMMARY OF 1993 RESEARCH (MAJOR ACCOMPLISHMENTS) BY OBJECTIVE:**

Objective 1a. We continued monitoring of populations of fungi known to reduce viability of sclerotia (overwintering inoculum) of pathogens in various residue management systems being tried in growers fields. We also attempted to obtain yield estimates and disease levels in the fields studied to determine the effects of the practices.

Soil samples were obtained prior to flooding of the fields for inoculum level determination and nutrient status (N,P,K,Zn,Fe,S), Soil pH, EC, and CEC for analysis and determination of possible correlations between residue management practice, disease level and yield. The fields being studied were selected to allow continuation of the monitoring of various fungal populations and a comparison of the effects of the practices being tried on occurrence of rice diseases in those fields.

(a) In previous years we reported isolation of large numbers of fungi from straw in soil contact, standing stubble, and from sclerotia of S. oryzae from different systems of residue management. The basic objective being to determine what effects the significant changes in the rice culture system, incorporation of the straw, have on the populations of organisms that participate in the decomposition of straw and the viability and overwintering of pathogen inoculum levels. These studies have shown that certain fungi are more frequently associated with decomposing residue and sclerotia. Samples were collected again this year from most of the fields studied in the previous years to determine if changes in the microflora were occurring. Mostly those fields in which rice is being continuously grown were included in this years survey. A range of systems was surveyed including burned in the fall, chopped and incorporated in the fall, fall incorporated and flooded.

Results of these studies were very similar to those obtained last year. The majority of fungal species recovered from decomposing stubble continued to be Hyphomycetes, a result similar to previous years. A succession of fungi from the early fall isolations to those in the spring continued to be observed. Ubiquitous epiphytes of the late growing season rice plant were again the most frequently isolated from the stubble at the beginning of the decomposition period (early fall). The majority of these were Hyphomycetes and the rice pathogens S. oryzae and R. oryzae-sativae.

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Hyphomycetes in the genera Cladosporium, Acremonium, Epicoccum and Alternaria were isolated frequently throughout the winter depending on the system. These resident fungi are apparently active throughout the decomposition period on rice residue regardless of temperature.

Acremonium and Alternaria were more sensitive to management system changes. Their frequency of isolation was lower in tilled systems supporting our earlier conclusion of the importance of soil contact in initiating the succession of residue decomposing fungi. In flooded systems, changes in frequency of isolation of most fungal species recovered was reduced as also was the severity of stem rot and aggregate sheath spot. The general reduction in fungal activity (including pathogens) under these conditions (flooded) suggests both temperature and lower oxygen availability may be responsible for their lower occurrence in isolation attempts.

Samples from systems where residue was incorporated or "rolled" providing early soil contact of the residue in the fall supported an earlier and more diverse succession of fungi. Zygomycetes such as Mucor species were usually more frequently recovered from these systems. They were recovered less frequently latter in the decomposition period.

Later in the overwintering process (March-April), several Basidiomycetes were more common in the stubble samples. These are considered the secondary colonizers of decomposing organic matter. Predominant genera again included Waitea, Coprinus, Rhizoctonia and Humicola.

Some Gelasinospora species were isolated only from systems where the residue had been burned in the fall. These were usually not recovered later in the winter samplings.

These results are very similar to those obtained in previous years. Most differences in fungi isolated from rice residue in various residue management treatments are minor and vary almost as much between years as they do between treatments with the exception of those treatments where residue is burned in the fall. There does not appear to be major changes in the microflora on rice residue on the surface. A lower level of fungal diversity and frequency was again observed when the residue is incorporated in the fall and the field then flooded.

The fungi that continued to be most frequently isolated from sclerotia of S. oryzae were again mostly Hyphomycetes.

The Ascochyta species shown previously to be the most promising for its potential as a means of biological control of the stem rot pathogen did not increase in frequency of isolation over that of earlier years.

(b) Correlations between nutritional and soil factors with disease level and yield were determined for the sites under study. Ranges of the factors determined from soil samples from (U.C. DANR Analytical Laboratory) the 23 sites studied were:

soil pH - - - - -	4.3 - 5.7
EC (milli-mhos/cm) - -	0.23 - 0.59
CEC (meq/100g) - - - -	11.4 - 52.8
SO <sub>4</sub> -S (ppm) - - - - -	13.9 - 68.0
N (%) - - - - -	0.076 - 0.143
P (ppm) - - - - -	8 - 48
K (ppm) - - - - -	54 - 199
Zn (ppm) - - - - -	1.4 - 11.0
Fe (ppm) - - - - -	71 - 336

No consistant pattern of differences between the levels of the factors and the various methods of residue management were apparent.

The most interesting correlations observed were between levels of Fe, S, and the severity of Aggregate Sheath Spot when compared in fields in which M201 and M202 were grown. On both cultivars, Aggregate Sheath Spot severity was highest when soil F levels exceeded 200 ppm F and soil S exceeded 50 ppm. Although these were the strongest coorelations observed, more study is needed to understand their significance. There was also an association between higher Aggregate Sheath Spot severities and yields in the fields studied. Whether this positive association is due to both the disease and the rice responding to a common factor (S) requires further study.

A coorelation between higher soil K levels and lower stem rot severities was detected in these data. There are reports from other countries suggesting benifits of K in reducing stem rot disease. When trials designed to explore this possibility were done in the past benifits were not observed. Nevertheless the question should be readdressed under conditions of residue incorporation.

Aggregate Sheath Spot was lowest and yields higher in fields where residue had been incorporated and then flooded in the fall than in fields where residue was incorporated and not flooded.

Objective 1b. Study of the occurrence, severity and interaction of Stem Rot (*S. oryzae*) and Aggregate Sheath Spot (*R. oryzae sativae*) under various residue management treatments was continued at experimental sites in Butte, Colusa and Sutter Counties in cooperation with the nitrogen studies being conducted by S. Pettygrove (RM-6).

The experimental design for the continuous trial in Sutter County at the Sills Ranch is described under project RM-6. The main block comparisons are: Fall Burn (FB), Fall Incorporation by discing (FI), Spring Incorporation by discing (SI), Fall Burn plus Vetch (FBV), Fall Incorporation plus Vetch (FIV) and Spring Incorporation plus Vetch (SIV), each replicated 5 times. Subplots of seven nitrogen rates, 0, 30, 60, 90, 120, 150 and 180 lbs N per acre are superimposed on each of the 30 main plots.

Disease ratings for Stem Rot and Aggregate Sheath Spot were made for each of the 210 subplots. Average values for subplot (N level) comparing effects of Stem Rot, Aggregate Sheath Spot and the interaction of the two diseases is shown in the figures Disease Interaction A, Aggregate Sheath Spot & Yield Interaction A and Stem Rot & Yield Interaction A.

These figures demonstrate the complexity of the relationship between the two diseases and also that of various nitrogen fertilization levels. For example: Aggregate Sheath Spot incidence is higher at low N levels where lowest yields were obtained (0 & 30 lbs N),  $R = 0.616$ . Stem Rot severity was generally highest at N levels above 60 lbs N. The lower level at 180 N observed in this data set is not consistent with observations in previous years or in other trials this year for stem rot. The R value between disease severity and yield was .655. I interpret this to indicate that disease is contributing to lower yields but probably not as significantly as is excess N level in these comparisons.

The inoculum level of S. oryzae (stem rot) was determined for each of the main plots. Viable sclerotia per gram soil in the seed bed ranged from: FB=3.52, FBV=3.80, FI=4.74, FIV= 5.38, SI=10.06, SIV=9.24. All of these levels are well above the point where inoculum level differences ( approximately 1.0 viable\scleroyia\gram soil and above) have been observed to result in differences in increasing severity of stem rot (i.e. levels of 1.0 are capable of producing a maximum amount of disease).

The negative correlation between Aggregate Sheath Spot incidence and Stem Rot severity was  $R=0.940$  and generally follows the relationship of N level to the two diseases.

Similar comparisons were made at 3 other sites, B, C and D in Butte County. The plot designs were four replications of N at the same levels in site A. Site B was excluded due to a failure to control weeds in the trial area. Sites C and D were located on the Skinner Ranch in adjacent fields. M-202 was grown in both. The residue was burned in the fall 1992 in field C and incorporated in the fall in field D.

In field C (residue burned) yields were significantly higher than those obtained in field D (residue incorporated). The inverse relationship between Aggregate Sheath spot and Stem Rot was not as strong ( $R=.320$ ) and correlations between disease levels and severity were somewhat low indicating that factors in addition to disease were involved in the yield differences.

Inoculum levels from the seedbed were quite high in both fields with that in C where the residue was burned only slightly lower than where it was incorporated for R. oryzae sativae. S. oryzae inoculum level in both fields was near that expected where disease differences would not be further affected.

The interactions of Stem Rot, Aggregate Sheath Blight and yield for these sites are shown in figure series C and D.

Overall there is the same trend in the field data as that seen for greenhouse experiments regarding the interaction of S. oryzae and R. oryzae sativae. Whether the increases in Aggregate Sheath Spot observed in recent years have \are contributing to lower levels of stem rot damage or if these lower levels are due to the improved resistance in our cultivars can only be speculative. In most cases that been have measured Aggregate Sheath Blight does not affect yields as severely as stem rot. Whether this would be the case on all cultivars should be watched closely as new cultivars are developed.

Objective 2. In field and greenhouse tests where both S. oryzae and Rhizoctonia oryzae-sativae are established, stem rot often appears to occur at a level less than would be expected from the known S. oryzae inoculum level. Other studies in the laboratory have shown that R. oryzae-sativae, Sclerotium hydrophylum and the Ascochyta sp. isolated from sclerotia are mycoparasites of the sclerotia of S. oryzae.

Greenhouse experiments to determine the optimum inoculum levels and methods for applying these fungi as potential biological control agents of the stem rot disease were carried out. These consisted mainly as co-inoculation onto rice plants with sclerotia of S. oryzae, the cause of stem rot. The test organisms were added to the experiments both in pelleted form and as free propagules.

Rates tested for S. hydrophulum and R. oryzae-sativae ranged from 10 - 300 viable sclerotia per plant. These higher rates are much more than have been observed in the field.

In all tests with S. hydrophylum, stem rot was minimized to insignificant levels by all concentrations tested. S. hydrophylum is not an aggressive pathogen of rice but it was recovered from sloughed leaves and sheathes of 100% of the test plants.

R. oryzae-sativae coinoculations resulted in significant stem rot reduction only when inoculation levels exceeded 25 sclerotia per plant. Levels of R. oryzae sativae often exceed this in the field when calculated on a sclerotia/gram soil/plant.

Coinoculation experiments with S. oryzae with conidia of the Ascochyta species that is strongly mycoparasitic on S. oryzae sclerotia did not result in stem rot reduction in the greenhouse tests regardless of number of conidia added. This result may have been expected since this mycoparasite occurs on and in the sclerotia during the overwintering season and is not generally an epiphyte. Thus its effect on viability of stem rot sclerotia occurs during the soil and saprophytic stages of the stem rot pathogen.

Results of these tests were similar regardless of the rice cultivar upon which they were conducted.

These and earlier results from both field and greenhouse tests suggest that field applications of these organisms may not be practical as an approach to controlling stem rot. Since all three occur naturally in rice fields, methods to enhance their natural populations may be more productive.

Objective 3: The UC Cooperative Extension regional variety trial for Butte County was located in a field where residue has been incorporated in the fall for the past three years. This allowed for an evaluation of disease occurrence on different cultivars under conditions of fairly high disease pressure. The disease ratings were made at the time the field was drained. Results for the currently grown cultivars evaluated during 1992 season and 1993 season at this location are compared below. They are listed in order of increasing severity of stem rot as observed each year. Aggregate Sheath Spot incidence observed is also given in order of increasing incidence.

Stem Rot		Aggregate Sheath Spot	
(1992)	(1993)	(1992)	(1993)
(Disease Severity)	(Disease Severity)	(Disease Incidence)	(Disease Incidence)
Ranking	Ranking	Ranking	Ranking
M-204	M-202	L-202	M-203
L-203	M-201	M-203	L-202
M-202	M-202	M-103	M-103
M-201	M-204	Calmo.101	S-201
M-103	L-203	M-201	Calmo.101
L-202	Calmo.101	S-201	M-201
M-203	M-203	M-202	M-202
Calmo.101	L-202	M-204	L-203
S-201	S-201	L-203	M-204

The relative rankings differ to some degree between years for the specific diseases but in most cases not significantly.

There is also a trend that those cultivars on which lower stem rot severity occurs sustain higher incidences of Aggregate Sheath spot. This is consistent with the observations and results reported regarding the interaction of various organisms earlier in this report.

Objective 4. The changes in our California rice culture system suggest that efforts to improve disease resistance should be increased. Breeding has increased Stem Rot resistance in many of our presently grown cultivars over that in the older tall cultivars. At the same time these are more susceptible to Aggregate Sheath Spot than were the older cultivars.

Germplasm collections on which disease evaluations had been reported in other rice growing areas were surveyed and accessions requested for evaluation with California isolates of S. oryzae and R. oryzae sativae with the objective of identifying possible new sources of resistance for use by the plant breeders. Particular emphasis this year was on collections that had been reported to have resistance to rice sheath blight caused by R. solani. This was based on suggestions and results by some rice researchers that resistance to R. solani may also be effective against R. oryzae sativae. The two diseases are similar in life cycle and symptoms. The pathogens are similar in morphology but one is bi-nucleate and the other is multi-nucleate.

Seed of 43 wild rice accessions representing 10 species of Oryza were screened in greenhouse tests for disease reaction to both S. oryzae and R. oryzae sativae including: O. ridleyi, O. latifolia, O. minuta, O. nivara, O. eichingeri, O. rhizomatis, O. alta, O. officinalis, O. australiensis and O. rufipogon. Also tested were lines Ta-Poo-Cho and Pankaj from the IRGC series. These later two are reported to be resistant R. solani. California cultivars M-201 and M-202 were used as comparison cultivars in all tests.

One accession of each of O. alta, O. latifolia, O. rufapogon and 2 of O. minuta were more resistant to stem rot than M-201 and M-202 but none of these showed a promising level of Aggregate Sheath Blight resistance.

Ta-Poo-Cho, Pankaj, and the O. rhizomatis, O. ridleyi, O. alta, O. nivara, O. latifolia and O. officinalis showed a higher level of Aggregate Sheath Spot resistance than M-201 and M-202 but were more susceptible to Stem Rot.

None of the lines tested thus far showed acceptable levels of resistance to both Stem Rot and Aggregate Sheath Blight. Additional accessions will be obtained and evaluated this year.

#### PUBLICATIONS OR REPORTS:

Webster, Robert K. Report to the California Rice Research Board: Project RP-2. Cause and Control of Rice Diseases, pp. 77-87. In: Annual Report of Comprehensive Rice Research. 1992. University of California and U.S Dept. of Agriculture.

#### CONCISE GENERAL SUMMARY OF CURRENT YEAR'S RESULTS:

Continued study of fields in which various methods of residue incorporation are being practiced revealed similar population frequencies of organisms seen in previous years. Fungi isolated from fields where residue is incorporated are more frequently

recovered and are more diverse than in fields where the residue is burned. In the fields where the residue was incorporated and then flooded, frequency of recovery of most of the species being monitored is reduced. This years results confirm oue earlier observations that incorporation of the residue is affecting the populations of mycoflora that occur in rice fields. What the long term effects of these shifts will be on rice culture and rice disease severity is not yet known.

Preliminary observations for possible differences in nutritional factors between fields where residue is incorporated or burned revealed no consistant pattern between levels of soil factors and disease with the possible exception of potassium, iron and sulfur. These observations need further study.

An inverse relationship between Stem Rot severity and Aggregate Sheath Spot incidence observed in field trials suggests that AGSS is affecting the level of incidence and severity of Stem Rot similar to results in greenhouse studies. The magnitude of this interaction varies by degree in the field when different cultivars are grown. This interaction probably contributes to the relative differences in disease susceptibility measured between cultivars in the field under heavy disease preasure.

Attempts to identify new sources of disease resistant germplasm for use by breeders will be continued. Sources studied this year showing the highest level of AGSS resistance were highly susceptible to stem rot. No sources studied this year had promising levels of resistance to both AGSS and Stem Rot.





