

Blast fungus inoculation reduces accumulation and remobilization of pre-anthesis assimilates to rice grains

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Summary. The importance of stored assimilates for grain development in cereals has been widely recognized, particularly in cereals exposed to stress during the grain filling period. This study was conducted to investigate the effect of the blast fungus *Magnaporthe grisea* on dry matter accumulation, remobilization and utilization in rice (*Oryza sativa* L.) under field conditions. Four rice varieties with various levels of susceptibility to rice blast were grown in 2003 and 2004 under high and low disease intensity, caused by early inoculation and natural infection respectively. High disease intensity reduced plant height, dry matter accumulation, the harvest index and grain yield. Plants grown under high disease intensity translocated fewer stored assimilates to the grain than plants grown under low disease intensity. Dry matter translocation from the vegetative parts to the grain was significantly correlated with dry matter at anthesis ($r=0.65$, $P<0.01$). High stress from early leaf blast caused by inoculation with the blast fungus affected overall plant growth and lowered the production of pre-anthesis assimilates, as well as causing severe levels of neck blast that resulted in a low translocation rate during the filling period, and therefore a lower grain yield than in naturally infected plants.

Key words: dry matter, translocation, *Oryza sativa*, *Pyricularia oryzae*.

Introduction

Grain filling of cereals depends on carbon from the current assimilates being transferred directly to the grain, and assimilates being redistributed from reserve pools in vegetative tissues either pre- or post-anthesis (Kobata *et al.*, 1992; Schnyder, 1993). The importance of each of these resources is dependent on many factors, including plant species, cultivar, cultural practices and growth conditions. The proportion of grain yield provided by the

translocation of pre-anthesis assimilates accounts for 6–73% in wheat (Austin *et al.*, 1977; Bidinger *et al.*, 1977; Papakosta and Gagianas, 1991), 4–74% in barley (Gallagher *et al.*, 1975; Przulj and Momcilovic, 2001) and 46–65% in triticale (Royo *et al.*, 1999). In rice, various studies have shown that the contribution of remobilized carbohydrates to grain yield ranges from 20 to 42% (Cock and Yoshida, 1972; Ramasamy *et al.*, 1997; Ntanos and Koutroubas, 2002).

The role of stem reserves in grain development may become crucial under conditions of severe abiotic stress, such as water deficit or heat, which diminishes the assimilation process during the filling period. Early plant senescence induced by water deficit during grain filling in rice may facilitate

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remobilization of carbon reserves, accelerate grain filling, and increase grain yield under drought conditions (Yang *et al.*, 2003; Kumar *et al.*, 2006). In addition, the contribution of dry matter partitioning from the stems and leaves increases with the severity of drought stress (Kumar *et al.*, 2006).

The importance of pre-anthesis assimilates for grain development has also been reported for plants grown under biotic stress. The post-anthesis tolerance of wheat to speckled leaf blotch (*Septoria tritici*) has been attributed to the storage of stem reserves and the capacity of plants to translocate reserves to the growing kernel when the photosynthetic source is limited (Zilberstein *et al.*, 1985).

Blast disease, caused by *Magnaporthe grisea* (Hebert) Barr (anamorph: *Pyricularia oryzae* Cavara), is one of the most important diseases of rice due to its destructive and widespread nature (Ou, 1985). The blast fungus can infect rice plants at any stage of their development. It occurs early as whitish to grayish and brownish spots or lesions, as nodal rot, and as a neck blast, which causes necrosis and frequent breakage of the panicle (Agarwal *et al.*, 1989). Leaf blast lesions reduce the net photosynthetic rate of individual leaves in a leaf area extending far beyond the leaf portion showing visible disease (Bastiaans and Roumen, 1993). Neck blast is considered the most important symptom of rice blast because it is more closely related to yield loss, and severe neck blast sometimes occurs without being preceded by severe leaf blast (e.g., Zhu *et al.*, 2005).

Blast disease epidemics are affected by weather, varietal susceptibility and crop management practices (e.g., Suzuki, 1975; Ou, 1985; Nyvall, 1999). Several studies have examined yield loss from blast disease (Goto, 1965; Padmanabhan, 1965; Bastiaans, 1993a). Most of these studies were conducted in tropical and subtropical areas. In Mediterranean conditions, Katsantonis *et al.* (2008) reported a greater tissue nitrogen concentration and nitrogen translocation from the vegetative parts to the grain in rice plants grown under high disease pressure than in plants grown under low disease pressure. However, only limited information is available on how rice blast affects carbohydrate storage in the grain filling of rice. The purpose of this study was to assess, under field conditions, the pre-anthesis dry matter accumulation, remobilization and utilization of vari-

ous rice varieties in response to blast disease in inoculated and naturally infected plants grown in a Mediterranean-type environment.

Materials and methods

Field experiments

The experiments were carried out at the Experimental Station of the Cereal Institute in Kalochori (latitude 40°33' N, longitude 23°00' E, altitude 0 m), Thessaloniki, Greece for two growing seasons, in 2003 and 2004. Four rice varieties, in 2003 and 2004. Four rice varieties, Maratelli and Selenio from Italy, Senia from Spain, and L-202 from Greece, were tested. Apart from Maratelli, these varieties are currently being cultivated in Europe. The varieties were chosen after a preliminary study to have a wide degree of response to the rice blast (Ntanos *et al.*, 2002). L-202, Selenio and Senia are moderately resistant to blast. Maratelli is a susceptible variety and was used as a susceptible control (Araujo *et al.*, 1998).

Two plots of rice plants were grown in a field and each plot was given a particular disease treatment. In the first treatment, conidia of *M. grisea* were inoculated into the plants, while in the second plot, located 300 m away so as to avoid any cross contamination with the fungus from the first plot, the rice plants were left to develop infection naturally. These two treatments corresponded to a high and a low disease intensity respectively.

Seeds were sown in pots on 6 May 2003 and 13 May 2004. The seedlings were transplanted by hand to the field at the 5- to 6-leaf stage, on 12 June 2003 and 24 June 2004. With both treatments, the seedlings were arranged in rows using the adjacent control design with three replications (Briggs and Shebeski, 1968; Katsantonis *et al.*, 2007). Selenio, Senia and L-202 were used as the varieties for testing, and Maratelli as the adjacent control variety. According to this arrangement, rows of the testing varieties were always separated from each other by two rows of the control variety. The rows were 2 m long and 0.15 m apart, and the plant-to-plant distance within rows was 0.10 m.

The field was flooded from the day of transplanting and the water was maintained between 5 and 10 cm deep until the grains reached physiological maturity. The field was fertilized with 55 kg N ha⁻¹ as ammonium sulfate, 75 kg P₂O₅ ha⁻¹ as super phosphate and 75 kg K₂O ha⁻¹ as potassium sulfate,

all applied by hand before transplanting. A further 145 kg N ha⁻¹ was applied when the rice reached the tillering stage, 50 kg N ha⁻¹ at stem elongation, and finally 50 kg N ha⁻¹ before panicle initiation. Weeds were controlled by hand weeding.

Inoculum preparation – Disease assessment

Stock cultures were created from seeds of infected plants of the Greek variety Roxani. The fungus was isolated in 9-cm Petri dishes, on a layer with three filter papers soaked in sterile water. The Petri dishes were incubated for 7 days at 22°C with a 12 hour day. When the fungus was grown from infected seeds, it was identified microscopically. The conidia of *P. oryzae* were harvested by touching the conidiophores with a small wire loop containing a 0.025% Tween 20 solution and cultured monoconidially using the serial dilution method, on a rice flour agar (RFA) medium (15 g l⁻¹ rice flour, 20 g l⁻¹ agar, 2.5 g l⁻¹ yeast extract and 40 mg l⁻¹ streptomycin). Permanent stock cultures were prepared by inoculating the fungus on RFA dishes and then a sterile filter paper disk (Whatman no. 4, 70 mm) was placed over each medium. Cultures were incubated at 27°C with a 12-hour day for 10 days. After the fungus fully penetrated the filter paper, the paper was peeled away from the dish and air-dried at 30°C. The papers were then enclosed in sterile water-proof boxes and stored at -20°C (Valent *et al.*, 1986, Agarwal *et al.*, 1989).

To prepare the inoculum, 0.5 cm-long segments of the stored filter paper were placed on RFA and incubated for 10 days at 27°C with a 12 hour day. The conidia were diluted to a final concentration of 5×10⁴ per ml of tap water with 0.5% gelatine. The conidial suspension was prepared from pre-inoculated dishes. Ten ml of sterile distilled water was poured on the overgrown cultures. Conidia were then dislodged into a suspension using a metal rod. The suspension was filtered through cheesecloth to remove all mycelium and measured using a hemocytometer. Inoculation was performed during the evening hours at the 6- to 7-leaf stage by spraying 3 ml of the conidial suspension on each rice plant using a fine air sprayer (Crown, Humbrol spray attached to Camlab aerosol). After inoculation with the blast fungus, free water was sprinkled on the leaves three times a day

between 11:00 and 16:00, in order to facilitate blast development.

Blast was assessed on individual plants 20, 40, 60 and 80 days after inoculation (DAI), using a lesion-type rating scale from 1 to 6, where 1 = completely resistant plants, with no symptoms; 2 = resistant plants, with small brown spots of pin-point size or larger brown spots without a sporulating center; 3 = partially resistant plants, with small roundish to slightly elongated necrotic sporulating spots; 1-2 mm diameter spots with a brown margin or yellow border; 4 = susceptible plants, with narrow gray or elliptical lesions, 1–2 mm wide and 3 mm long lesions surrounded by a brown border; 5 = highly susceptible plants, showing wide spindle-shaped gray lesions with a brown or chlorotic border; and 6 = highly susceptible plants, with coalescent greyish to whitish lesions (dead leaves) (Notteghem, 1981; Silué *et al.*, 1992). Neck blast was assessed 55 days after 50% heading by counting the infected panicles and dividing this by the total number of panicles per line to calculate the percentage of infection.

Morpho-physiological data

The dates of anthesis and maturity were recorded. Anthesis was recorded when the anthers in the central spikelets of 50% of the panicles in a row had extruded, and maturity when most of the panicles in a plot had completely lost their green colour. Plant height and panicle length were measured at maturity, and were both based on individual measurements of the main stems of five plants selected at random from each replication. Plant height was measured from the soil surface to the tip of the panicle. Plant samples were taken at anthesis and maturity from a 1 m segment of each row. The plants were cut at ground level, the stems in the sample were counted and the plants were separated into leaf+stem and panicle. At maturity, panicles were further separated into the panicle vegetative components (branches and rachis) and the grain. All plant samples were oven-dried at 70°C until constant weight, and weighed.

The parameters referring to dry matter movement within the rice plant that are discussed in this paper were evaluated as follows (Papakosta and Gagianas, 1991):

1. Dry matter translocation (kg ha⁻¹) = Total above-ground dry matter at anthesis – [(leaf+stem)

+ panicle vegetative components] dry matter at maturity.

2. Dry matter translocation efficiency (%) = (Dry matter translocation/total above-ground dry matter at anthesis) × 100.

3. Contribution of pre-anthesis assimilates to the grain (%) = (Dry matter translocation/grain dry matter at maturity) × 100.

Post-anthesis dry matter accumulation (PAD-MA) was calculated as the difference between the total above-ground dry matter at maturity and that at anthesis. The harvest index (HI) was calculated as the ratio of grain dry weight to the total above-ground dry weight at maturity.

Statistical analysis

All data were subjected to analysis of variance (ANOVA) by GenStat (2005) using a completely randomized design with unequal replications. Means between disease treatments in each year were compared using the least significant difference (LSD) test as calculated for unequal replications (Gomez and Gomez, 1984). Standard statistical procedures were used for calculating simple correlation coefficients and linear regression equations (Steel and Torrie, 1980).

Results

Weather conditions and disease development

Weather conditions were typical for the area in both years of the experiment; however, there were some differences between years (Katsantonis *et al.*, 2008). Small rain events in the summer of 2003 maintained the temperature at moderate levels and strongly raised the relative humidity. During the period of disease development the mean temperatures ranged from 24.6 to 26.2°C in 2003, and from 23.6 to 29.6°C in 2004, and the relative humidity from 42 to 76% in 2003, and from 27 to 53% in 2004.

Leaf blast ratings in the inoculated experiments at the various disease assessment dates were similar in both years. At the final annual assessment (80 DAI), leaf blast ratings ranged from 3.33 (Senia) to 4.95 (Maratelli) in 2003 and from 3.33 (Senia) to 5 (L-202) in 2004 (Fig. 1). Across varieties, mean neck blast in 2003 was almost double that of 2004 (71.6% vs. 39.4%) (Fig. 2). Natural infection in the non-inoculated rice plants was high in 2003; at the final annual assessment it ranged, for leaf blast, from 2 (Senia) to 3.3 (Maratelli) (Fig. 1) and, for neck blast, from 48.8% (L-202) to 71.8%

Fig. 1. Ratings of leaf blast from *Magnaporthe grisea* in four rice varieties, inoculated (solid lines) and non-inoculated (dotted lines) with the fungus in field experiments conducted in 2003 and 2004. Leaf blast was assessed 20, 40, 60 and 80 days after inoculation. Vertical bars show the mean standard error.

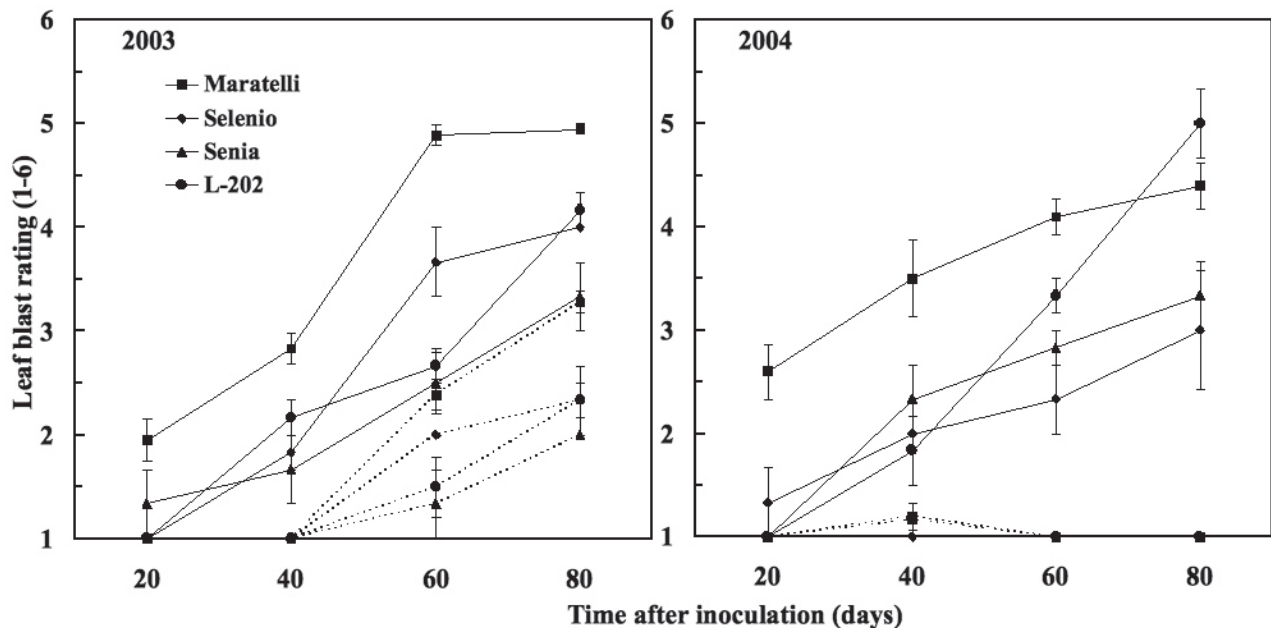
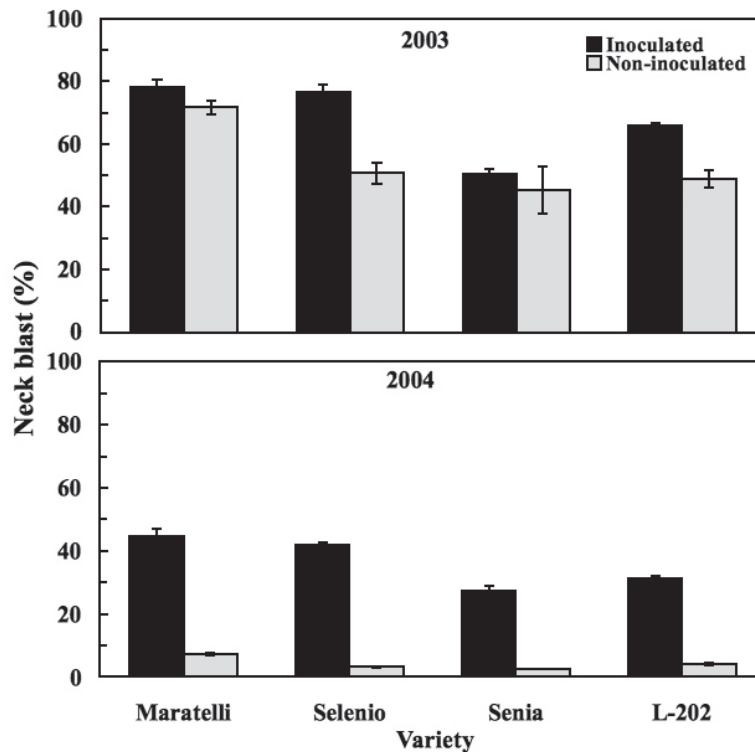


Fig. 2. Neck blast from *Magnaporthe grisea* 55 days after the time of 50% heading of four rice varieties, inoculated and non-inoculated with the fungus in field experiments conducted in 2003 and 2004. Error bars show the mean standard error.



(Maratelli) (Fig. 2). Infection of the non-inoculated plants on the other hand was negligible in 2004, with its highest value of 1.2 (Maratelli) for leaf blast and 7.1% (Maratelli) for neck blast. Leaf blast ratings at 80 DAI were significantly correlated with neck blast in both years ($r=0.86$, $P<0.01$ in 2003, and $r=0.879$, $P<0.01$ in 2004).

Dry-matter accumulation and partitioning

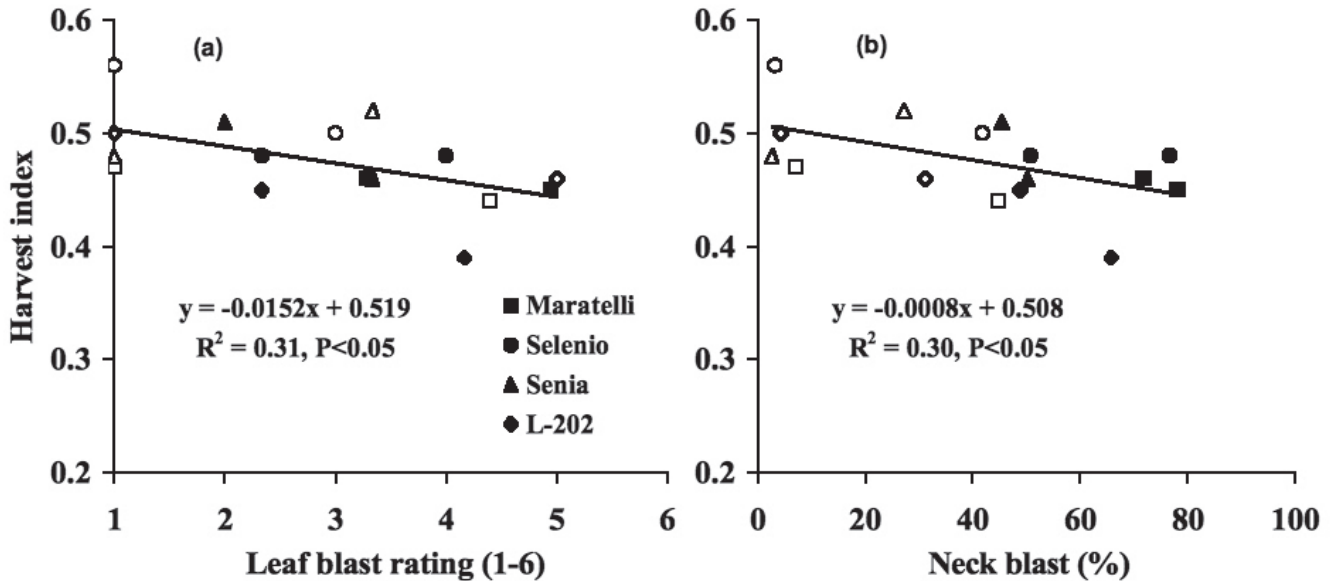
Blast disease affected the agronomical characteristics of the plants, to an extent that depended on the rice variety and on the year of experimentation (Table 1). Across varieties, inoculation with the fungus reduced mean plant height and panicle length by 3.7 and 6.2% respectively in 2003, and by 3.3 and 2.1% in 2004. Plant height was positively correlated with total dry matter at anthesis ($r=0.74$, $P<0.01$) and with total dry matter at maturity ($r=0.72$, $P<0.01$). Inoculation reduced the HI from 2% (Maratelli) to 13% (L-202) in 2003, and from 6% (Maratelli) to 11% (Selenio) in 2004

(Table 1). The HI was negatively correlated with both leaf blast ($r=-0.56$, $P<0.05$) and neck blast ($r=-0.55$, $P<0.05$) (Fig. 3).

Dry-matter accumulation was in most cases lower in 2004 than in 2003 with both treatments (Table 2). On average, total above-ground dry matter at anthesis and maturity in 2004 were 20.2% and 24.6% lower respectively than in 2003. Inoculation of the plants reduced overall dry matter at anthesis by 13.9% in the first year, and by 7.2% in the second year. Selenio was most strongly affected in both years, showing a decrease in total above-ground dry matter of 38% in 2003, and of 33% in 2004. Varieties differed in their dry matter at anthesis; when disease intensity was negligible, in non-inoculated plants in 2004, Maratelli produced the highest dry matter ($10,135 \text{ kg ha}^{-1}$) and Senia the lowest (6649 kg ha^{-1}) (Table 2).

Total above-ground dry matter after anthesis increased with both treatments. Averaged across varieties, post-anthesis dry matter levels were

Fig. 3. Relationship (a) between leaf blast rating 80 days after inoculation and the harvest index; (b) between neck blast infection 55 days after the time of 50% heading and the harvest index in rice. Each relationship was based on the means derived from four rice varieties, two disease treatments and two years (2003, filled symbols; 2004, empty symbols) of experimentation (n=16).



higher for non-inoculated plants than for inoculated plants; (8091 vs 6949 kg ha⁻¹ in 2003, and 6513 vs 4946 kg ha⁻¹ in 2004). Inoculation caused a decrease in the total dry matter at maturity that was similar in the two years (-14.3% in 2003 and -14.5% in 2004). The dry matter of the vegetative parts of inoculated plants at maturity was 12% less than that of non-inoculated plants in 2003, and 8.1% less in 2004. The corresponding decrease in grain yield was 17% in 2003, and 21% in 2004.

Dry matter translocation

The mean amount of dry matter translocated from the vegetative parts of the plants to the grains during the filling period in 2003 was significantly lower for inoculated plants than for non-inoculated plants (644 vs 1099 kg ha⁻¹), whereas in 2004 there were no significant differences between treatments (1109 vs 1153 kg ha⁻¹) (Table 3). Dry matter translocation was positively correlated with total above-ground dry matter at anthesis ($r=0.64$, $P<0.01$) (Fig. 4). Dry matter translocation efficiency ranged from 2.7 to 17.8%. Mean dry matter translocation efficiency across varieties was lowered significantly (31.2%) by inoculation

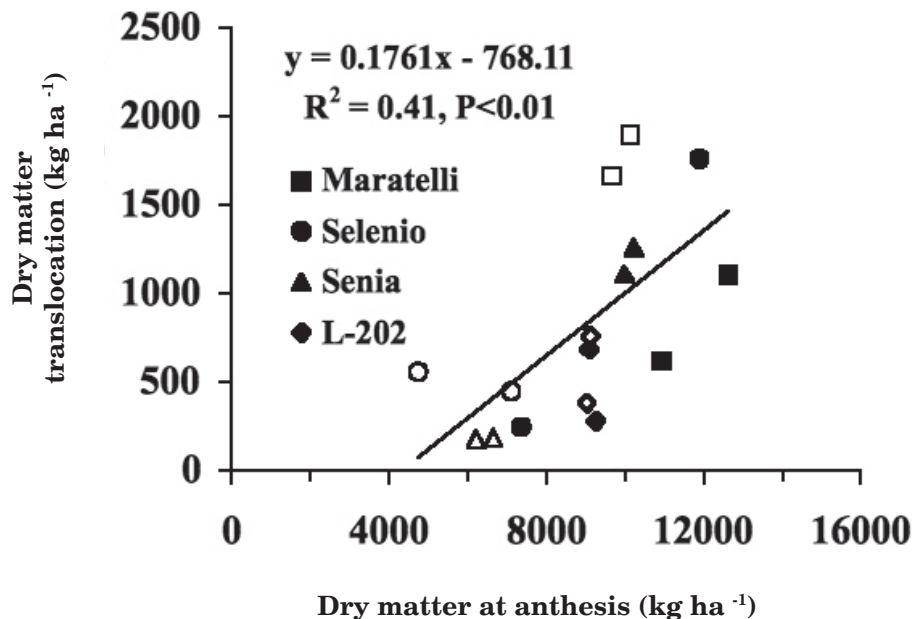
in 2003, but in 2004 these decreases were similar in inoculated and non-inoculated plants. Inoculation caused differences between varieties in their dry-matter translocation and in their dry-matter translocation efficiency only in 2003. Both dry matter-related parameters were significantly lowered by inoculation in Maratelli and Selenio, but they were not affected in Senia and L-202. The contribution of pre-anthesis assimilates to grain dry matter at maturity ranged from 2.8 to 27.4% (Table 3). Overall it was 27.1% less in the inoculated plants than in the non-inoculated plants in 2003 (8.6 vs 11.8%), while the difference in 2004 was not significant (18.6 vs 15.4%).

Discussion

Yearly differences

The differences in the rates of rice infection between the two years of the experiment could be explained by differences in the weather between years. Rice blast was more severe in 2003, when the temperature was lower and the relative humidity higher than in 2004. It is well documented that environmental conditions, and especially relative

Fig. 4. Relationship between dry matter accumulated at anthesis and dry matter translocation in rice. The relationship was based on the means derived from four rice varieties, two disease treatments and two years (2003, filled symbols; 2004, empty symbols) of experimentation (n=16).



humidity, strongly affect the sporulation, release and germination of blast conidia (Ou, 1985). In the inoculated plants, leaf blast infection at the final annual assessment (80 DAI) was similar in the two years, indicating that inoculation caused relatively high and uniform levels of rice blast in the leaves. Neck blast on the other hand was greatly affected by variations in the weather. In 2004, when the summer was drier, inoculated plants had on average 23% fewer infected panicles than in 2003. The non-inoculated plants seemed to be more weather dependent, because both types of blast symptoms varied greatly between years. The hotter and drier growing period of 2004 produced very low levels of infection, suppressing disease development. The positive correlation between leaf blast and neck blast in both years suggests that a high disease level in the leaves probably also caused severe blast on the neck.

Weather differences between years probably also caused the lower amounts of dry matter accumulation seen with both disease treatments in 2004 as compared with 2003 (Table 2). The high

temperatures (above 35°C) that prevailed during the vegetative growth of plants in 2004 may have increased maintenance respiration, leading to a greater loss of assimilates (Murata, 1964; Yoshida, 1981).

Inoculation and plant growth parameters

The greater disease severity of inoculated plants reduced plant height and dry matter accumulation compared with non-inoculated plants (Tables 1 and 2). The extent of the reduction, however, depended on the rice variety and on the year of experiment, reflecting variations in genotype response to blast disease and the level of disease intensity achieved in a particular year. These results are consistent with Torres and Teng (1993), who reported that leaf blast had a strong negative effect on plant height, and that this effect that was proportional to disease severity. The shortening of rice plants from rice blast found in our study was probably associated with the adverse effect of blast on dry mat-

Table 1. Plant height, panicle length and harvest index of four rice varieties inoculated and non-inoculated with *Magnaporthe grisea* in field experiments conducted in 2003 and 2004.

Year	Treatment	Variety	Plant height (cm)	Panicle length (cm)	Harvest index
2003	Inoculated	Maratelli	109.3	18.1	0.45
		Selenio	94.5	19.9	0.48
		Senia	93.5	15.7	0.46
		L-202	86.6	20.1	0.39
		Overall mean	100.9	18.3	0.45
	Non-inoculated	Maratelli	114.0	19.1	0.46
		Selenio	96.8	20.5	0.48
		Senia	97.2	17.2	0.51
		L-202	89.5	22.4	0.45
		Overall mean	104.8	19.5	0.47
		LSD ($P=0.05$) ^a	3.1	0.9	0.03
		LSD ($P=0.05$) ^b	5.7	1.6	0.05
	LSD ($P=0.05$) ^c	2.3	0.6	0.02	
2004	Inoculated	Maratelli	94.0	18.3	0.44
		Selenio	83.0	19.0	0.50
		Senia	89.0	16.7	0.52
		L-202	87.0	21.3	0.46
		Overall mean	90.4	18.6	0.46
	Non-inoculated	Maratelli	97.0	18.3	0.47
		Selenio	86.0	19.7	0.56
		Senia	93.0	17.3	0.48
		L-202	90.0	22.0	0.50
		Overall mean	93.5	19.0	0.50
		LSD ($P=0.05$) ^a	3.9	0.7	0.02
		LSD ($P=0.05$) ^b	7.1	1.3	0.04
	LSD ($P=0.05$) ^c	2.8	0.5	0.01	

LSD, Least significant difference at the 0.05 probability level.

^a Means of the control variety (Maratelli) between treatments.

^b Means of the main varieties (Selenio, Senia, L-202) between treatments.

^c Overall treatment means in each year.

ter, as suggested by the positive correlation between plant height and levels of dry matter. The lower levels of dry matter in inoculated plants were mainly due to a lower crop growth rate, because the time to anthesis was similar with both treatments (data not shown). The crop growth rate is a function of canopy gross pho-

tosynthesis and crop respiration (Evans, 1993), processes that are strongly influenced by the environment (Loomis and Connor, 1992; Akita, 1993). Blast severity affects both these processes in rice. Bastiaans (1993a) reported that leaf blast reduced the photosynthetic rate of rice leaves not only in the green leaf area itself,

Table 2. Dry-matter accumulation and partitioning at anthesis and maturity of four rice varieties inoculated and non-inoculated with *Magnaporthe grisea* in field experiments conducted in 2003 and 2004.

Year	Treatment	Dry matter (kg ha ⁻¹)										PADMA ^d (kg ha ⁻¹)
		Anthesis					Maturity					
		Leaf + stem	Panicle vegetative components	Total	Leaf + stem	Panicle vegetative components	Total	Leaf + stem	Panicle vegetative components	Total	Grain	
2003	Inoculated	Maratelli	10491	432	10923	9798	510	10308	8560	18868	7945	
		Selenio	7080	291	7371	6698	430	7128	6537	13665	6294	
		Senia	9571	404	9975	8561	307	8868	7621	16489	6514	
		L-202	8777	318	9095	8018	396	8414	5400	13814	4719	
		Overall mean	9537	387	9924	8832	447	9279	7593	16873	6949	
	Non-inoculated	Maratelli	12033	604	12637	10981	557	11538	9591	21129	8492	
		Selenio	11332	570	11902	9410	736	10146	9447	19593	7691	
		Senia	9790	432	10222	8507	457	8964	9301	18265	8043	
		L-202	8914	353	9267	8562	425	8987	7484	16471	7204	
		Overall mean	11076	532	11608	9960	549	10509	9190	19699	8091	
LSD (<i>P</i> =0.05) ^a LSD (<i>P</i> =0.05) ^b LSD (<i>P</i> =0.05) ^c		950	84	1005	865	90	901	630	1001	732		
		1735	153	1835	1579	164	1644	1150	1827	1336		
		690	61	729	627	65	653	457	726	531		
2004	Inoculated	Maratelli	9299	361	9660	7594	402	7997	6193	14189	4529	
		Selenio	4556	191	4747	3914	280	4195	4577	8772	4025	
		Senia	6040	169	6209	5741	294	6036	6141	12176	5968	
		L-202	8805	318	9124	7879	491	8369	6986	15355	6232	
		Overall mean	7957	297	8254	6766	380	7146	6055	13200	4946	
	Non-inoculated	Maratelli	9682	453	10135	7833	413	8246	7437	15683	5548	
		Selenio	6889	223	7112	6320	350	6669	8623	15292	8180	
		Senia	6472	177	6649	6107	358	6465	5885	12350	5701	
		L-202	8689	337	9026	8131	519	8650	9250	17900	8874	
		Overall mean	8578	355	8932	7368	411	7779	7665	15445	6513	
LSD (<i>P</i> =0.05) ^a LSD (<i>P</i> =0.05) ^b LSD (<i>P</i> =0.05) ^c		788	77	817	420	71	451	385	633	1128		
		1439	141	1492	766	130	824	703	1156	2059		
		572	56	593	304	52	327	279	459	818		

LSD See Table 1.
^{a,b,c} See Table 1.
^d PADMA, post-anthesis dry matter accumulation.

but also in the green leaf tissue surrounding the lesion. Moreover, Toyoda and Suzuki (1957) and Bastiaans (1993a) reported an increase in respiration as a result of blast infection.

Inoculation reduced post-anthesis dry-matter accumulation in both years. Both grain and vegetative dry matter contributed to this reduction. However, the reduction in grain dry matter due to inoculation was much greater than the reduction in vegetative dry matter, suggesting that blast disease had a stronger effect on the grains than on the vegetative plant parts. These results could be explained by considering the specific nature of blast disease in rice. Blast infection during the generative growth stages mainly results in panicle or neck infections that cause necrosis of the plant neck and incomplete grain filling. Candole *et al.* (2000) reported that rough rice from blast-infected panicles was drier and thinner than rice from blast-free panicles. In our study, rice blast altered dry matter partitioning at maturity, as indicated by the decrease in the HI due to inoculation. Leaf and neck blast were both linked to the HI because of their significant correlation (Fig. 3). Bastiaans (1993b) found that the HI was similar in healthy and inoculated rice plants. This finding, contradicting the present study, may have been due to the genotype being differently expressed in different conditions. A reduction of the HI due to disease stress was also reported by Tivoli *et al.* (1996) for pea plants (*Pisum sativum*) with ascochyta blight (*Mycosphaerella pinodes*).

Inoculation and dry matter translocation

Dry matter translocation was greater in plants grown under low disease stress (non-inoculated plants) than in plants grown under high disease stress (inoculated plants) (Table 3). By contrast, previous research on various plant species reported that the translocation of pre-anthesis assimilates increased in plants grown under stress (Blum *et al.*, 1997; Ramasamy *et al.*, 1997; Yang *et al.*, 2001; Kumar *et al.*, 2006). The inconsistency between our findings and those of other researchers could be explained by the fact that neck blast stress has a strong effect on the translocation process and may completely block the transport routes towards the grains in infected plants. Another explanation could be that the time of stress

initiation differed in the various studies. In most studies, the factor that caused the stress was applied after anthesis. In the present study, disease stress was maintained throughout the growth cycle of the plants and reduced the dry matter at anthesis and also the dry matter that was potentially available for translocation. A lower amount of dry matter at anthesis resulted in a lower translocation of dry matter during the grain filling period, as confirmed by the positive correlation between the two parameters (Fig. 4).

Differences in dry matter translocation between the disease treatments were greater in 2003 than in 2004, although the difference in disease level between inoculated and non-inoculated plants was smaller in 2003. This was probably because, as has already been mentioned, weather conditions in 2004 suppressed plant growth, reversing the difference in dry matter accumulation between disease treatments as compared with 2003.

The differences in dry matter translocation and dry matter translocation efficiency between rice varieties were mainly associated with the susceptibility of these varieties to blast disease. When a high disease level happened in Maratelli and Senlenio in 2003, inoculated plants had significantly lower values of both dry matter-related parameters than non-inoculated plants. On the contrary, in Senia and L-202, where the disease level was lower, differences caused by the disease treatments were not significant.

Conclusions

Early inoculation of rice plants with *M. grisea* caused stress and affected overall plant growth, with a significant reduction in grain yield as compared with non-inoculated plants. Inoculated plants, grown under high disease intensity, accumulated less dry matter up to anthesis and translocated less dry matter from the vegetative parts of the plants to the grains than naturally infected plants, grown under low disease intensity. Leaf blast and neck blast were correlated, and it is therefore concluded that high leaf blast levels caused the low production of pre-anthesis assimilates, while at the same time high infection also caused severe levels of neck blast, resulting in a low translocation rate during the grain filling period.

Table 3. Dry matter translocation, dry matter translocation efficiency and contribution of pre-anthesis assimilates to the grain of four rice varieties inoculated and non-inoculated with *Magnaporthe grisea* in field experiments conducted in 2003 and 2004.

Year	Treatment	Variety	Dry matter translocation (kg ha ⁻¹)	Dry matter translocation efficiency (%)	Contribution of pre-anthesis assimilates to the grain (%)	
2003	Inoculated	Maratelli	615	5.6	7.2	
		Selenio	243	3.5	3.7	
		Senia	1107	11.0	14.5	
		L-202	681	7.4	12.1	
		Overall mean	644	6.4	8.6	
		Overall mean				
	Non-inoculated	Maratelli	1099	8.6	11.8	
		Selenio	1756	14.8	18.6	
		Senia	1258	12.3	13.5	
		L-202	278	2.9	3.6	
		Overall mean	1099	9.3	11.8	
		Overall mean				
	LSD ($P=0.05$) ^a			309	2.8	4.5
	LSD ($P=0.05$) ^b			564	5.1	8.2
LSD ($P=0.05$) ^c			224	2.0	3.2	
2004	Inoculated	Maratelli	1663	15.9	27.4	
		Selenio	552	10.2	12.8	
		Senia	173	2.8	2.8	
		L-202	754	8.1	10.9	
		Overall mean	1109	11.7	18.6	
		Overall mean				
	Non-inoculated	Maratelli	1889	17.8	25.6	
		Selenio	443	6.8	5.1	
		Senia	184	2.7	3.1	
		L-202	376	4.1	4.0	
		Overall mean	1153	11.4	15.4	
		Overall mean				
	LSD ($P=0.05$) ^a			1014	10.1	15.8
	LSD ($P=0.05$) ^b			1852	18.3	28.9
LSD ($P=0.05$) ^c			736	7.3	11.5	

^{a,b,c} See Table 1.

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