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CROP PROTECTION

Fungi on irrigated rice seeds produced in the pre-germinated system in the Alto Vale do Itajaí region, Santa Catarina state, Brazil

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ABSTRACT: *The aim of this research was to identify and quantify fungi infecting irrigated rice seeds produced in the 2015/16, 2016/17 and 2017/18 harvests in the pre-germinated system in the Alto Vale do Itajaí Region, Santa Catarina State, Brazil. A total of 479 lots of eight cultivars were analyzed. Seeds were disinfected and sown in potato-sucrose-agar culture medium with incubation for seven days at 25ºC ± 2ºC and photoperiod of 12 hours. The main fungus detected in the three crops season was Microdochium albescens with 54.9% of average incidence, followed by Alternaria padwikii (7.7%) and Bipolaris oryzae (3.3%).*
Key words: *Microdochium albescens, Oryza sativa, seed health*.

Fungos em sementes de arroz irrigado produzidas no sistema pré-germinado na região do Alto Vale do Itajaí, estado de Santa Catarina, Brasil

RESUMO: *O objetivo deste trabalho foi identificar e quantificar fungos infectando sementes de arroz irrigado, produzidas nas safras 2015/16, 2016/17 e 2017/18, no sistema pré-germinado na região do Alto Vale do Itajaí, estado de Santa Catarina. Foram analisados 479 lotes de oito cultivares, sendo as sementes desinfestadas e semeadas em meio de cultura de batata-sacarose-ágar com incubação por sete dias a 25ºC ± 2 ºC e fotoperíodo de 12 horas. O principal fungo detectado nas três safras foi Microdochium albescens com 54,9% de incidência média, seguido de Alternaria padwikii (7,7%) e Bipolaris oryzae (3,3%).*
Palavras-chave: *Microdochium albescens, Oryza sativa, sanidade de sementes.*

In the state of Santa Catarina (SC), irrigated rice (*Oryza sativa* L.) is produced in a pre-germinated system widely sown in monoculture areas and in regions where long periods of wetness occur, factors that favor the occurrence of diseases (SOSBAI, 2018). In this crop system the pathogenic fungus inoculum is easily dispersed to the panicles with subsequent infection of grains or seeds, which can cause stains and their reduction (MIURA, 2002).

Fungi *Alternaria padwickii* (Ganguly), *Bipolaris oryzae* (Haan's Breda), *Cercospora janseana* (Racib.), *Fusarium moniliforme* (Sheldon), *Microdochium albescens* (Thüm.) syn. *Microdochium oryzae*; *Gerlachia oryzae* (Hashioka & Yokogi), *Pyricularia oryzae* (Cav.) and *Sarocladium oryzae* (Saw) some of the main pathogens associated with rice seed (MEW & GONZALES, 2002; SOSBAI, 2018). Infected seeds may affect physiological quality and constitute an important agent of pathogen dissemination (OU, 1972).

Rice seed production in SC focuses mainly on the pursuit of high physiological quality. There is no obvious concern for seed production with high health quality or low incidence of pathogens. The aim of this research was to identify and quantify fungi infecting seeds of irrigated rice cultivars produced in the pre-germinated system in the Alto Vale do Itajaí (AVI), SC, Brazil.

Seed samples produced in the 2015/16, 2016/17 and 2017/18 crops season, benefited by Cooperative Cravil, were analyzed in a total of 479 lots of SCS121 CL (165 lots), SCS122 Miura (85), SCS116 Satoru cultivars. (79), EPAGRI 109 (58),

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SCSBRS Tio Taka (49), SCS118 Marquês (22), SCS117 CL (11) and Primoriso CL (10), from the AVI region, SC. Disease management in these crops followed the indications of the cooperative. For chemical control, a mixture of strobirulin + triazole + benzothiazole was sprayed at the stages of booting and flowering and / or grain formation (SOSBAI, 2018). In the cooperative the seeds were stored in polypropylene bags with humidity and room temperature. Seeds were collected and sent to the Plant Pathology Laboratory Santa Catarina State University (UDESC). Detection of fungi was done in PSA + A culture medium (Potato-Sucrose-Agar + Antibiotic = 200 mg L^{-1} streptomycin sulfate). Seeds were disinfected with sodium hypochlorite solution (1%) for two minutes and then washed with sterile water. For each lot, 200 seeds were analyzed, four replicates of 50, sown in acrylic Petri dishes and kept in growth chambers for seven days at 25 ºC and 12 hours photoperiod. For detection of *P. oryzae* the paper substrate incubation method (blotter test) was used. Six lots were randomly selected from each cultivar, totaling 48 lots. The seeds were arranged in a gerbox acrylic box containing two layers of moistened filter paper and incubated in a continuous light growth chamber at 27 ºC. The evaluation was performed at four and seven days. A completely randomized experimental design was used for both detection methods. Were considered infected in which it was possible to identify the colony and / or structures of the fungi, confirming the identification with microscope (BARNETT & HUNTER, 1998).

The main pathogenic fungus associated with seeds were *M. albescens* with 54.9% of the average incidence and 100% of the prevalence, respectively followed by *A. padwikii* (7.7% / 90.6%), *B. oryzae* (3.3% / 59.9%), *Curvularia* sp. (3.1% / 66.5%) and *S. oryzae* (2.7% / 49.4%) (Table 1 and

Table 1 - Average incidence of fungi associated with seeds of irrigated rice cultivars produced in the Alto Vale do Itajaí Region, Santa Catarina State. Lages, 2019.

Cultivar	Nº Lotes	Incidence of fungi (%)										
		Mic	*Alt*	*Cur*	*Bip*	*Sar*	*Nig*	*Pen*	*Asp*	*Altsp*	*Cer*	*Fus*
Safra 2015 / 2016												
SCS121 CL	57	61.9	6.0	6.6	7.3	2.2	0.8	1.1	0.7	0.1	0.2	0.0
SCS116 Satoru	33	72.3	2.9	1.7	1.6	2.2	1.6	1.3	1.2	0.4	0.3	0.0
SCSBRS Tio Taka	17	55.6	7.7	5.7	7.3	2.7	3.0	0.5	0.7	0.9	0.4	0.4
Epagri 109	11	68.1	2.0	2.0	2.5	3.9	1.2	2.1	2.3	1.0	0.0	0.0
SCS117 CL	11	60.5	9.0	3.9	5.1	2.5	1.4	0.4	0.3	0.5	0.4	0.0
SCS118 Marquês	10	57.2	7.6	9.6	3.3	3.7	0.7	1.8	1.8	0.0	0.2	0.0
Average (%)		62.6	5.8	4.9	4.5	2.8	1.4	1.2	1.2	0.5	0.3	0.1
Safra 2016 / 2017												
SCS121 CL	55	530	7.4	3.0	2.7	4.2	0.9	0.6	1.0	0.7	1.2	0.1
SCS116 Satoru	33	506	9.1	2.4	2.8	2.7	0.7	1.3	1.9	0.9	0.8	0.2
Epagri 109	31	455	11.3	3.0	3.2	3.9	0.6	0.8	1.3	1.5	0.6	0.3
SCSBRS Tio Taka	26	627	6.5	2.5	3.4	3.4	0.7	0.5	0.7	1.0	0.5	0.3
SCS122 Miura	12	431	11.7	5.6	5.1	7.1	0.0	1.2	2.1	2.3	1.1	0.0
SCS118 Marquês	12	453	9.3	3.0	3.3	4.8	1.6	0.4	0.8	1.6	1.1	0.0
Average (%)		500	9.2	3.2	3.4	4.3	0.7	0.8	1.3	1.3	0.9	0.2
Safra 2017 / 2018												
SCS122 Miura	73	535	7.2	1.7	1.6	1.1	0.6	0.1	0.0	0.3	1.2	0.0
SCS121 CL	53	516	5.9	0.8	3.3	0.5	0.8	0.1	0.2	0.4	1.0	0.1
Epagri 109	16	446	14.3	1.7	1.9	0.3	0.7	0.1	0.0	0.8	1.2	0.0
SCS116 Satoru	13	489	8.9	1.2	2.6	0.3	0.0	0.2	0.4	0.7	2.5	0.1
Primoriso CL	10	577	7.2	1.0	0.9	0.9	0.5	0.2	0.0	0.4	2.8	0.0
SCSBRS Tio Taka	6	562	4.5	0.7	2.5	2.2	0.3	0.3	0.0	0.3	1.2	0.0
Average (%)		521	8.0	1.2	2.1	0.9	0.5	0.2	0.1	0.5	1.6	0.0
Overall Average (%)		550	7.7	3.1	3.3	2.7	0.9	0.7	0.9	0.8	0.9	0.1

Mic - *Microdochium albescens*; Alt - *Alternaria padwiicki*; Cur - *Curvularia sp.*; Bip - *Bipolaris oryzae*; Sar - *Sarocladium oryzae*; Nig - Nigrospora oryza; Pen - *Penicillium* spp.; Asp - *Aspergillus flavus*; Alt_{sp} - *Alternaria* sp.; Cer - *Cercospora janseana* e Fus - *Fusarium* sp.

table 2). Similar results were reported in irrigated rice cultivars in 350 samples analyzed in Rio Grande do Sul State (RS), from 1993 to 1998, also detecting *G. oryzae* (18.0%), *Alternaria* sp. (6.3%), *C. lunata* (4.9%) and *B. oryzae* (2.6%) (FRANCO et al., 2001). In another study also in RS, in 162 plots of rice from the 2005/06 crop season, *Alternaria* sp. (9.6%), *Bipolaris sp*. (9.3%), *Gerlachia* sp. (4.9%) and *Curvularia* sp. (3.8%) (FARIAS et al., 2007).

The fungus *M. albescens* was the only with 100% prevalence (Table 2). This pathogen causes scalding in rice leaves, a growing disease in crop in the AVI, SC. The fungus was reported in RS association with seeds ranging from 6 to 31% (average 18%) (FRANCO et al., 2001) and 1 to 33% (average 4.9%) (FARIAS et al., 2007), but with a lower incidence than that obtained in this study (range 43.1 to 72.3% and average 54.9%). This may be related to the detection method, reaction of rice genotypes and cropping systems. MALAVOLTA (2007), using filter paper method detected an average incidence of *M. oryzae* of 24.6 and 29.8%, respectively in 2003 and 2004, in rice genotype seeds in the of São Paulo (SP) state. PSA medium is more efficient in detecting *M. albescens* when compared to the filter paper method (GUTIÉRREZ et al. 2009). Analyzing genotypes, a higher incidence was observed in SCS116 Satoru (72.3% in 2015/16 crop) and lower in SCS122 Miura (43.1% in 2016/17 crop). These values are considered high for a possible obtaining of healthy seed for pathogen management, since there is no information on resistance reaction of cultivars for *M. albescens* in the states of SC and RS (SOSBAI, 2018).

The fungus *A. padwickii*, included in the group of grain spotters, obtained average incidence in the three harvests of 7.7% and prevalence of

Table 2 - Average prevalence of fungi associated with seeds of irrigated rice cultivars produced in the Alto Vale do Itajaí Region, Santa Catarina State. Lages, 2019.

Cultivar	Nº Lotes	Prevalence of fungi (%)										
		Mic	*Alt*	*Cur*	*Bip*	*Sar*	*Nig*	*Pen*	*Asp*	*Altsp*	*Cer*	*Fus*
Safra 2015 / 2016												
SCS121 CL	57	100	84.7	66.7	39.4	63.2	36.4	33.3	48.5	21.2	9.1	0.0
SCS116 Satoru	33	100	100	70.6	76.5	57.6	64.7	29.4	23.5	17.7	17.7	0.0
SCSBRS Tio Taka	17	100	100	90.9	45.5	64.7	54.6	36.4	9.1	18.2	18.2	0.4
Epagri 109	11	100	36.4	45.5	54.6	81.8	27.3	54.6	63.6	27.3	9.1	0.0
SCS117 CL	11	100	89.5	89.5	77.2	72.7	26.3	45.6	22.8	8.8	14.0	0.0
SCS118 Marquês	10	100	90.0	100	50.0	70.0	40.0	50.0	40.0	0.0	100	0.0
Average (%)		100	83.4	77.2	57.2	68.3	41.6	41.6	34.6	15.5	13.0	0.1
Safra 2016 / 2017												
SCS121 CL	55	100	89.1	80.0	63.6	63.6	23.6	27.3	36.4	32.7	40.0	3.6
SCS116 Satoru	33	100	90.9	69.7	60.6	45.5	21.2	48.5	42.4	36.4	21.2	6.1
Epagri 109	31	100	92.3	69.2	53.8	38.5	19.2	21.9	19.2	46.2	34.6	7.7
SCSBRS Tio Taka	26	100	96.8	77.4	67.7	58.1	16.1	32.3	45.2	54.8	32.3	9.7
SCS122 Miura	12	100	100	91.7	100	91.7	0.0	58.3	50.0	75.0	33.3	16.7
SCS118 Marquês	12	100	100	91.7	75.0	50.0	50.0	16.2	25.0	50.0	25.0	16.7
Average (%)		100	94.8	79.9	70.1	57.9	21.7	34.1	36.4	49.2	31.1	10.1
Safra 2017 / 2018												
SCS122 Miura	73	100	868	26.4	66.0	18.9	22.6	5.7	7.6	18.9	32.1	5.7
SCS121 CL	53	100	100	53.1	69.2	15.4	23.1	15.4	15.4	30.8	53.8	0.0
Epagri 109	16	100	100	16.6	50.0	0.0	16.7	16.7	0.0	16.7	50.0	0.0
SCS116 Satoru	13	100	937	50.0	56.3	12.5	18.8	6.3	0.0	31.3	50.0	0.0
Primoriso CL	10	100	918	57.5	45.2	34.3	12.3	6.9	1.4	10.9	49.3	2.7
SCSBRS Tio Taka	6	100	90.0	50.0	30.0	50.0	0.0	10.0	0.0	20.0	90.0	0.0
Average (%)		100	93.7	42.3	52.8	21.9	15.6	10.2	4.1	21.4	54.2	1.4
Overall Average (%)		100	90.6	66.5	59.9	49.4	26.3	28.6	25.0	28.7	32.8	11.6

Mic - *Microdochium albescens*; Alt - *Alternaria padwiicki*; Cur - *Curvularia sp.*; Bip - *Bipolaris oryzae*; Sar - *Sarocladium oryzae*; Nig - Nigrospora oryza; Pen - *Penicillium* spp.; Asp - *Aspergillus flavus*; Alt_{sp} - *Alternaria* sp.; Cer - *Cercospora janseana* e Fus - *Fusarium* sp.

90.6%. In Egypt, India, Korea, Nepal and Thailand, the fungus was detected in 282 lots of 388 analyzed (MATHUR et al., 1972). In Brazil, in SP in 2003 and 2004, the average incidence of *A. padwickii* was 8.2% and 1.6%; respectively, in cultivation of genotypes in flood irrigated system (MALAVOLTA, 2007). In RS, FRANCO et al. (2001), detected values from 0.4% to 20.6%, but do not describe the *Alternaria* species.

The fungus *B. oryzae* presented higher average incidence in cultivar SCS121 CL and SCSBRS Tio Taka (7.3%, 2015/16 crop), and lower average value in cultivar Primoriso CL (0.9%, 2017/18 crop). For this fungus, we highlighted a prevalence of 100% detected in the 2016/17 crop in the cultivar SCS122 Miura. In SP was detected an average incidence of 13.6%, higher than that found in this study (MALAVOLTA, 2007).

The fungus *Curvularia* sp. was detected with incidence and prevalence values similar to *B. oryzae*. It was not possible to identify the species of *Curvularia* since no morphological and molecular characterization was performed. The species *C. eragrostidis* (P. Henn) and *C. lunata* (Wakker) Boedijn, are detected in rice seeds (LIMA & FURTADO, 2007). Other fungi were detected with lower average incidence, such as: *S. oryzae* (2.7%), *Nigrospora oryzae* (0.9%), *Penicillium* spp. (0.7%), *Aspergillus flavus* (0.9%), *Alternaria* sp. (0.8%), *C. janseana* (0.9%) and *Fusarium* sp. (0.1%) (Table 1). These are also reported in seeds in RS (FRANCO et al., 2001; FARIAS et al., 2007). The fungus *P. oryzae* was not detected by the tested. In RS *P. oryzae* was detected with low incidence (0.04%) in 350 seed samples (FRANCO et al., 2001), but later was not detected in 162 samples analyzed (FARIAS et al., 2007).

This was the first survey of fungi associated with irrigated rice seeds in the pre-germinated cultivation system carried out in Santa Catarina State, serving as a basis for future research related to the epidemiological importance of infected seeds.

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DECLARATION OF CONFLICT OF INTERESTS

The authors declare no conflict of interest. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results.

AUTHORS' CONTRIBUTIONS

BTS and RTC conceived and designed experiments. BTS, VR and FCM performed the experiments, BTS carried out the lab analyses. BTS, JALVJ e JB performed statistical analyses of experimental data. BTS, JB, RTC, FCM, WR, VR and JALVJ prepared the draft of the manuscript. All authors critically revised the manuscript and approved of the final version.

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