

Multiplex amplicon sequencing dataset for genotyping pandemic populations of the wheat blast fungus

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We report genotyping data for 186 *Magnaporthe oryzae* (Syn. *Pyricularia oryzae*) isolates from different geographical regions obtained using multiplex amplicon sequencing. The dataset consists of 84 single nucleotide polymorphisms (SNPs) selected from transcript sequences to distinguish between the clonal lineage of the wheat blast fungus that emerged in South East Asia in 2016 and other *M. oryzae* genotypes. The sample coverage includes pandemic wheat blast isolates from Bangladesh (N=75, 2016-2020) and the newly reported Zambian outbreak (N=13, 2018-2020). The dataset is provided as an open science resource as part of the OpenWheatBlast initiative. We welcome analyses and feedback from the community.

Introduction

Given the propensity of the blast fungus *Magnaporthe oryzae* (syn. *Pyricularia oryzae*) to jump hosts and spread to new geographic regions, reliable and cost-effective diagnostic tools are needed to monitor this pandemic pathogen at the genotype level. This is not necessarily a straightforward problem considering the relatively low genetic diversity and potential for gene flow between lineages of *M. oryzae*. Here, we apply multiplex amplicon sequencing to rapidly genotype *M. oryzae* strains at low cost. We used a set of 84 single nucleotide polymorphisms (SNPs) that were designed to distinguish between the pandemic clonal lineage of the wheat blast fungus that reached South East Asia in 2016 from other *M. oryzae* genotypes (Islam et al., 2016). We applied this method to a panel of 186 isolates, including 75 wheat isolates from Bangladesh (2016-2020) (Islam et al. 2016) and 13 from the newly reported Zambian outbreak (2018-2020) (Tembo et al. 2020). Analyses of the SNP dataset should demonstrate whether the method can robustly discriminate between different lineages of *M. oryzae* (Gladieux et al. 2018). The dataset is provided as an open science resource as part of the [OpenWheatBlast](#) initiative. We welcome analyses and

feedback from the community. Whole genome shotgun sequencing of the Zambian wheat blast isolates is in progress.

Results and Conclusions

Selection of SNP panel. We selected 84 SNPs based on the transcriptomics data of Islam et al. (2016) and other blast fungus genome sequences available at the time (May 2017) (Table 1, Supplemental Dataset 1). The panel of 84 SNPs was primarily designed to discriminate between the wheat blast clonal lineage of *M. oryzae* in Bangladesh (BD lineage) from other genotypes. The targeted sequences and SNPs are provided in Supplemental Dataset 1 with the SNP indicated between brackets.

***M. oryzae* isolates.** The sample collection is listed in Table 2 and summarized in Table 3. It includes epidemic wheat blast isolates from Bangladesh (N=75) and Zambia (N=13) as well as wheat blast isolates from Brazil (N=18) and a worldwide collection of *M. oryzae* isolates representative of the main genetic lineages and isolated from an additional 12 grass hosts (Table 3). The collection dates span 1948 to 2020 (Table 3). The DNA was obtained from multiple sources. A subset of DNA samples was extracted in Norwich from cultures of isolates obtained from Bangladesh (N.U. Mahmud and T. Islam) and Zambia (B. Tembo) as well as from our own isolate collection (N. Talbot and S. Kamoun labs). In addition, we processed DNA samples directly provided by N.U. Mahmud, T. Islam and their co-workers (Bangladesh rice and wheat blast) and S. Asuke and Y. Tosa (global collection). Two transcript samples of wheat blast lesions collected in Bangladesh and processed in 2016 (Islam et al. 2016) were included as controls. Most isolates were previously described as indicated in Table 2 (Chiapello et al., 2015; Farman et al. 2002; Gladieux et al. 2018; Islam et al. 2016; Hau et al. 2007; Yoshida et al. 2016).

Genotyping matrix. We used ‘MonsterPlex’ amplification, a well-established method of multiplex amplicon sequencing as performed by Floodlight Genomics LLC (Knoxville, TN). We applied ‘MonsterPlex’ and Illumina DNA sequencing to the 186 isolates of *M. oryzae* (Table 2). We combined all 84 primer pairs into a single well for multiplex amplification of each sample and extracted the SNP identity from the Illumina sequence data. The matrix of 186 isolates x 84 SNPs is provided for further analysis (Supplemental Dataset 2). Missing data reflects partial coverage possibly because the isolates may have failed to amplify with a subset of the amplicons due to genetic divergence or other reasons. The two transcript samples also yielded amplicons with only a subset of the primers.

Data availability. The MonsterPlex method resulted in a cost-effective matrix of 84 SNPs for the collection of 186 *M. oryzae* samples (Supplemental Dataset 2). We provide the dataset as an open science resource as part of the [OpenWheatBlast](#) initiative (Kamoun et al. 2019) and through the [Zenodo OpenWheatBlast Community](#).

Statement on continuing work. The datasets are being shared prior to formal publication and should be considered preliminary. We encourage and welcome analyses and feedback from the community. Please use the [Zenodo OpenWheatBlast Community](#) to share any analysis reports or get in touch with [JW](#) and [SK](#). We will post our own analyses of this dataset as soon as completed.

Supplemental Datasets.

Supplemental Dataset 1. Targeted sequences flanking the 84 SNPs (in brackets).

Supplemental Dataset 2. SNP data for 186 samples.

References

- Chiapello H., Mallet L., Guérin C., Aguilera G., Amselem J., Kroj T., et al. 2015. Deciphering Genome Content and Evolutionary Relationships of Isolates from the Fungus *Magnaporthe oryzae* Attacking Different Host Plants. *Genome Biol Evol*, 7:2896–912.
- Farman, M.L., Eto, Y., Nakao, T., Tosa, Y., Nakayashiki, H., Mayama, S., and Leong, S.A. 2002. Analysis of the Structure of the AVR1-CO39 Avirulence Locus in Virulent Rice-Infecting Isolates of *Magnaporthe grisea*. *MPMI* 15:1, 6-16.
- Gladieux P., Condon B., Ravel S., Soanes D., Maciel J.L.N., Nhani A., et al. Gene Flow between Divergent Cereal- and Grass-Specific Lineages of the Rice Blast Fungus *Magnaporthe oryzae*. 2018. *mBio*, 9:e01219–17.
- Islam, T., Croll, D., Gladieux, P., Soanes, D., Persoons, A., Bhattacharjee, P., Hossain, S., Gupta, D., Rahman, Md.M., Mahboob, M.G., Cook, N., Salam, M., Surovy, M.Z., Bueno Sancho, V., Maciel, J.N., Nani, A., Castroagudin, V., de Assis Reges, J.T., Ceresini, P., Ravel, S., Kellner, R., Fournier, E., Tharreau, D., Lebrun, M.-H., McDonald, B., Stitt, T., Swan, D., Talbot, N., Saunders, D., Win, J., and Kamoun, S. 2016. Emergence of wheat blast in Bangladesh was caused by a South American lineage of *Magnaporthe oryzae*. *BMC Biology*, 14:84.
- Kamoun, S., Talbot, N.J., and Islam, M.T. 2019. Plant health emergencies demand open science: Tackling a cereal killer on the run. *PLOS Biology*, 17:e3000302.
- Maciel, J.L., Ceresini, P.C., Castroagudin, V.L., Zala, M., Kema, G.H., and McDonald, B.A. (2014). Population structure and pathotype diversity of the wheat blast pathogen *Magnaporthe oryzae* 25 years after its emergence in Brazil. *Phytopathology* 104, 95-107.
- Tembo, B., Mulenga, R.M., Sichilima, S., M'siska, K.K., Mwale, M., Chikoti, P.C., Singh, P.K., Pedley, K.F., Peterson, G.L., Singh, R.P., Braun, H.J. 2020. Detection and characterization of fungus (*Magnaporthe oryzae* pathotype Triticum) causing wheat blast disease on rain-fed grown wheat (*Triticum aestivum* L.) in Zambia. *PLOS ONE*, 15, e0238724.
- Hau, V., Hirata, K., Murakami, J., Nakayashiki, H., Mayama, S., and Tosa, Y. 2007. Rwt4, a wheat gene for resistance to Avena isolates of *Magnaporthe oryzae*, functions as a gene for resistance to Panicum isolates in Japan. *J Gen Plant Pathol* 73, 22–28.
- Yoshida, K., Saitoh, H., Fujisawa, S., Kanzaki, H., Matsumura, H., Yoshida, K., Tosa, Y., Chuma, I., Takano, Y., Win, J., Kamoun, S., and Terauchi, R. (2009). Association genetics reveals three novel avirulence genes from the rice blast fungal pathogen *Magnaporthe oryzae*. *Plant Cell* 21, 1573-1591.
- Yoshida K., Saunders D.G.O., Mitsuoka C., Natsume S., Kosugi S., Saitoh H., et al. 2016. Host specialization of the blast fungus *Magnaporthe oryzae* is associated with dynamic gain and loss of genes linked to transposable elements. *BMC Genomics*, 17:370.

Table 1. Single nucleotide polymorphisms (SNP) used to genotype host-specific lineages of *Magnaporthe oryzae*.

¹ Chromosome assignments, SNP positions, and gene descriptions are based on *M. oryzae* genome assembly and locus tags of annotation version MG8 (GenBank accession GCA_000002495.2).

² GenBank accession for chromosome

Index	Chromosome ¹	GenBank Accession ²	SNP Position ¹	Locus tag for gene1	Gene description ¹	Forward primer (5' - 3')	Reverse primer (5' - 3')
1	chr01	NC_017844.1	3938418	MGG_09255	kinesin-II 85 kDa subunit	TGGTCTTTTCGACAAAGAGG	CGGTATTCCAAGTCTGTG
2	chr01	NC_017844.1	5865304	MGG_05616	pre-mRNA-splicing factor prp46	GTTCTGTGATCTTGGAT	AGAGGAGAAGTCCCGCAAGT
3	chr02	NC_017850.1	1760459	MGG_08820	hypothetical protein	ACGACGATGGCCGTTTC	CTATTTCACTCCGGTCAGC
4	chr02	NC_017850.1	1766158	MGG_08821	hypothetical protein	TTGGGTTTGCTCAGCATCT	TGTTTGACTTATCCTCGTCCA
5	chr02	NC_017850.1	1905333	MGG_08863	hypothetical protein	GGCCACACCGTAACTGTCAT	AGTGCCGCAAGCTGGATA
6	chr02	NC_017850.1	1915505	MGG_11326	NAK protein kinase	GACCGCCTCAGTTCCAGTC	TCTGTTTCCTTGGGCTTAGG
7	chr02	NC_017850.1	2085434	MGG_08911	hypothetical protein	ATAGACATGTGCCTCACG	CCTCGAGGTGATGAGCAAA
8	chr02	NC_017850.1	2091939	MGG_08913	DNA polymerase gamma	GCCGAAACCTCGAAAATG	CAGTCTTGCCATCGGGATA
9	chr02	NC_017850.1	2143567	MGG_08925	hypothetical protein	AGAGGGCTGGGAGGAGTCT	CATGATGCTTCCAGGTAGGG
10	chr02	NC_017850.1	5630812	MGG_08084	cell wall biogenesis protein phosphatase Ssd1	GCGGCCGTCACCAGGA	TTTGTCGGTAGGCTTGAACC
11	chr02	NC_017850.1	5646459	MGG_08088	hypothetical protein	TTACCACTGGAAGCCTCGTC	AGGATGTGCGAACACCTAGC
12	chr02	NC_017850.1	5695471	MGG_08098	AP-3 adaptor complex subunit mu	TGTTCTGAGCCGTTTTAGCA	AATCGAAGAAGGCAGACG
13	chr02	NC_017850.1	5817237	MGG_08135	hypothetical protein	GGCGCCTTTCCGTCTATTAC	CCCATCCGTTGATGTTGAA
14	chr02	NC_017850.1	5886105	MGG_08156	kinesin-II 85 kDa subunit	TCAATCTTGCGCTTGAA	TGTCGATGACACATACG
15	chr02	NC_017850.1	6029864	MGG_08198	hypothetical protein	ATCCGTGATCTGGTCTGTTCT	ATCCTCTAGAGCCGGGACAC
16	chr02	NC_017850.1	6034337	MGG_08199	cutinase transcription factor 1 beta	AATCTTCTCCCCACCAC	TTCAATCCCCCGTGTATG
17	chr02	NC_017850.1	6104737	MGG_12321	hypothetical protein	AACCGACGCAAGATTTTTTA	CCTTGTCGAGACAAAACCT
18	chr02	NC_017850.1	6251845	MGG_07067	FAD binding domain-containing protein	GCCCTGCGCAATAGTGTA	ACATCGCCGTGCACCT
19	chr02	NC_017850.1	6342032	MGG_07095	hypothetical protein	ACCAGCAGACGAGACAGGAT	TCACCGATTGACTTCTTCA
20	chr02	NC_017850.1	6360844	MGG_07099	hypothetical protein	AGCATCAAGGCCATCATCA	CAAAGTCTGGTGGCTTCTT
21	chr02	NC_017850.1	6389194	MGG_12345	Sec1 family superfamily protein	TATGGGGAAGAGTGGAGCTG	TTGCTGGGTGCCACTACTA
22	chr02	NC_017850.1	6392670	MGG_07110	phosphatidylethanolamine N-methyltransferase	GCTAGGTAGCGGCTTGCTG	CACTTGCCCTGGGGTTTAC
23	chr02	NC_017850.1	6434021	MGG_07120	26S proteasome regulatory subunit rpn-1	GGACACAGAGGACCAGGAAC	CCCAGTGATTTGAAGTGCT
24	chr02	NC_017850.1	6462618	MGG_07123	lon protease like protein 2	AGACGCCCGTCTTTTTGAG	TCACCGACGCACGTA
25	chr02	NC_017850.1	6551944	MGG_07149	hypothetical protein	GCTGCTGCATGGTCCAAT	TCGAGTGCCGAGCTTTTTC
26	chr02	NC_017850.1	6641219	MGG_07174	elongator complex protein 1	GAAGCGATGAGTTGCTAT	TGCTGCTGCCAAACACTC
27	chr02	NC_017850.1	6769666	MGG_12377	phytochrome-1	GAGATTGCGGTTTCAGGACAC	CTGCTCAAGGTCACGGAAG
28	chr02	NC_017850.1	6789878	MGG_07213	pre-mRNA-splicing factor prp1	TGGGCAGCTCAACGAG	CTCGTTGTTGGGGTCTGTT
29	chr02	NC_017850.1	6859920	MGG_07222	DNA polymerase epsilon subunit B	GAGAAGAGGCGAGCGACAC	CCATAGGTGTGCTCCAAC
30	chr02	NC_017850.1	6863972	MGG_07224	threonine dehydratase	TGAAGAGGGAGGATGAGCAG	TCGAGGTGAGCCATCTTGT
31	chr02	NC_017850.1	6964720	MGG_07255	E3 ubiquitin-protein ligase hula	ATCGCCCTGTTAGCATGAAT	ATTCCGGTCTGTGTGGAAG
32	chr02	NC_017850.1	7040763	MGG_07273	WD repeat-containing protein 2	GGTAGACGTGCGTGTGCGAG	CGTCACTTCGATAGGGTGG
33	chr02	NC_017850.1	7104143	MGG_07286	hypothetical protein	TATGGCCACAGTCTACCG	AGCAAAGTTTCGGAGTTCGAC
34	chr03	NC_017849.1	70440	MGG_13326	hypothetical protein	TACAAACACGGTGCATGTCG	TCAAAGAGACTCCACCAGCA
35	chr03	NC_017849.1	917101	MGG_07523	hypothetical protein	AGTCACCTATGCCGTGGAAC	CGGGCAATATCTTCACGAAT
36	chr03	NC_017849.1	969690	MGG_13413	hypothetical protein	AGCTGCTCAAGGACTCAT	AGCAGCCGGAGGAGTTG
37	chr03	NC_017849.1	1398869	MGG_07660	hypothetical protein	ACCGAGTCCATCTGACACTG	CTGAGGATCTCAAGGCGATG

38	chr03	NC_017849.1	3007218	MGG_05797	hypothetical protein	GACCTTGCTCCCAACTTTT	CCGGCAACCAGGATAC
39	chr04	NC_017851.1	1805536	MGG_03455	hypothetical protein	ACGAGTTCAGATCAAGG	ATCATGCGGGCGGACGAC
40	chr04	NC_017851.1	2541577	MGG_03252	E3 ubiquitin-protein ligase ptr1 + RNA transporter 1	GCTAATAAGTCCACGCTGTGC	AGCCAAGCACGGTTTTCA
41	chr04	NC_017851.1	2617961	MGG_03230	seryl-tRNA synthetase	ATTCTTGCGCGTTGCT	CCCTGGGACTCAGC
42	chr04	NC_017851.1	2650655	MGG_03220	isoleucyl-tRNA synthetase	GGTCCATCACGTTCTCGTTT	TTGAAGAAGGCCGAAGAAAA
43	chr04	NC_017851.1	2766301	MGG_12761	hypothetical protein	GAAGCTCGGAGACGCCTAAT	CTGAGGATCGGTGTTTCCAT
44	chr04	NC_017851.1	3963393	MGG_06379	midasin	GAGTGTGAGAAACCCCATGC	ACGACGTCGCACCTGTCTAT
45	chr04	NC_017851.1	4581932	MGG_10118	ATPase family AAA domain-containing protein 1	TGTGCCATCTGAAGCAGTCT	AGGAGTCGGCATTACAAACG
46	chr05	NC_017852.1	2567951	MGG_00535	hypothetical protein	TATTGAGGAACCGGCCACT	TTTGGGGTTGAGCTG
47	chr05	NC_017852.1	2688927	MGG_00500	meiotically up-regulated 71 protein	TCGAGCAGTGAATTTTCGT	TCCGTGATGATTTGAGTTTCC
48	chr05	NC_017852.1	2759059	MGG_00477	hypothetical protein	TCGCTCAGTTTGCCTGTAA	CATTGGCAGATGAAACCTT
49	chr05	NC_017852.1	2832832	MGG_00456	BTB domain and ankyrin repeat protein	ACCCTTGATTTCGTTTAC	TGGGCATGACGAAGATGATA
50	chr05	NC_017852.1	2841083	MGG_00454	hypothetical protein	GAAGCTTCCACACCCAAGTC	ACACACAGGCCACGAAATA
51	chr05	NC_017852.1	2865464	MGG_00448	RanGTP-binding protein	ACAGCACGCCGTGAG	CAGATTATGCTGCGTAGCTC
52	chr05	NC_017852.1	2868782	MGG_00447	brefeldin A resistance protein	GAGCTGCAGGTAGGTGAGT	CTTCCCGTCTGTCAACC
53	chr05	NC_017852.1	2923295	MGG_00428	conidial yellow pigment biosynthesis polyketide synthase	GCGTCCAGGCATAAGTCG	ACTGGGATAAATACCACGA
54	chr05	NC_017852.1	3127670	MGG_00361	hypothetical protein	GTCCGAGGCAGACTTTCATGT	ATCACCCCTCCGCATCTTC
55	chr05	NC_017852.1	3205174	MGG_00338	hypothetical protein	GCGGGTAGACAGTGGACTA	ATCGAAAGAAAGGGTTCC
56	chr05	NC_017852.1	3504249	MGG_00258	hypothetical protein	TCACTTCCACAGCATGGTTC	CTAGAAGCCACGGTCAATC
57	chr06	NC_017853.1	2070800	MGG_12141	hypothetical protein	GACCAAAGCTACTGCGGAAC	AGCTGGCTGGGTGAT
58	chr06	NC_017853.1	2200503	MGG_12131	skeletal muscle and kidney-enriched inositol phosphatase	ACGGAGTTTGTGCGATGT	AACCACATCTGGGTGGGATA
59	chr06	NC_017853.1	2255686	MGG_04006	hypothetical protein	TCGGCCAACACTGGAAG	TGAGATCGTCGTCAATTCAAA
60	chr06	NC_017853.1	2277918	MGG_04000	hypothetical protein	GGTCGAATGTGTTTCGCTTCT	AGCGTCCGAGAAACGATGT
61	chr06	NC_017853.1	2284885	MGG_03997	hypothetical protein	GGATCGTGGTAAGACACAC	CCGATGGAGTATCATCATGG
62	chr06	NC_017853.1	2299116	MGG_03994	importin alpha re-exporter	GCCGTCCTTGATGACAATTAG	ACCAAGATGCAGGAGAACGA
63	chr06	NC_017853.1	2311842	MGG_03991	topoisomerase 1-associated factor 1	TCCCAGTCTTTGAACATAGAGGA	CATCCATCTCCTTACAACATGA
64	chr06	NC_017853.1	2379246	MGG_03970	glucan synthesis regulatory protein	ACAAGAGGTGATCCGATGCT	AAGCCCAAAGCTTGAGTCG
65	chr06	NC_017853.1	2386875	MGG_12118	STE/STE20/YSK protein kinase	TGTGGCTCTTGAGGTGAGG	TCGCTCAACCAACTCCA
66	chr06	NC_017853.1	2455997	MGG_12111	hypothetical protein	GCTGCATGTTCCCTCAAGTC	TTGAATAAAGCTTCGCTGA
67	chr06	NC_017853.1	3364033	MGG_09413	CAMK protein kinase	GATGATGCCCGACAAAT	CGGTTTCGAGAGTGCATGT
68	chr06	NC_017853.1	3414515	MGG_09398	hypothetical protein	AGCGGGTCCGTGCTTC	TGCGGTGGCCAGACC
69	chr06	NC_017853.1	3839221	MGG_09869	hypothetical protein	GTCTTGTTGGCGGGAAGTTCT	CCCAGAGTATAGACGATTTCCA
70	chr07	NC_017854.1	784208	MGG_02977	hypothetical protein	AGCCACCTCCTCGTTTCTC	ATGAGGAGCCACAGATTG
71	chr07	NC_017854.1	1385226	MGG_02823	beige/BEACH domain-containing protein	GCGGACAGAGGAAGAGAAGA	AAAGAAGCCGAGAGGACT
72	chr07	NC_017854.1	1454677	MGG_02804	DNA repair protein rad16	GGGCTCATACATGACGACGTA	CCTACGATGGTGACCAGGAC
73	chr07	NC_017854.1	1498220	MGG_02788	hypothetical protein	GATGCTCCGGGGTCACT	CAGAGCCCATACCAGTCGAT
74	chr07	NC_017854.1	1579823	MGG_02771	hypothetical protein	AAATTTGACACGTT	CACCAACAAGGAAGAAGACG
75	chr07	NC_017854.1	1592180	MGG_02767	phospholipid-transporting ATPase 1	TTGTCAACGGATTTGCCTTC	CCGATAGAGCCATATTC
76	chr07	NC_017854.1	1686743	MGG_02744	hypothetical protein	AGAGTCTATGGCGTTCTG	AGGACGATGGTACCAGGAC
77	chr07	NC_017854.1	1845527	MGG_02697	serologically defined colon cancer antigen 1	GCCCTCAGTGACATCTAGCC	TGATTCTGACAACGAAGG
78	chr07	NC_017854.1	2019953	MGG_12631	hypothetical protein	TTGACTCCAAGAAGGTCGTT	AGTTTGTCTCCATGAGC
79	chr07	NC_017854.1	2066602	MGG_02649	hypothetical protein	AAGGCTTGGACCGGTGTAG	TCCAGTGTGCCATTACCT
80	chr07	NC_017854.1	2070964	MGG_02648	interferon-induced GTP-binding protein Mx	TTCGCAAACGCTCTCAGTG	TCCTCGCGGAGACTCG
81	chr07	NC_017854.1	2084582	MGG_02644	hypothetical protein	GGTCGGTCTGTGCTCT	AAGTCAAGCATCCAAGTC
82	chr07	NC_017854.1	2126103	MGG_02631	hypothetical protein	GCCCCTCTTGACACTTCGAG	CAGGGAATTTTCGTCATGT

83	chr07	NC_017854.1	2164719	MGG_12647	Ser/Thr protein phosphatase family protein	GTCGTGATAATCGGGTTGCT	ATGAGCAGGTGGACTGA
84	chr07	NC_017854.1	3245392	MGG_09607	maltose permease MAL31	AGTACCTGCCCAGGACCAAC	CTCACGGCGTTGCTCAG

Table 2. *Magnaporthe oryzae* samples used in this report¹ Samples are sorted by host species from which they are collected from, and then by year collected.² Region (where available) and country where samples were collected.

Sample Name ¹	Host species collected from	Year	Location ²	Reference
Br58	<i>Avena sativa</i>	1990	Parana, Brazil	Yoshida et al., 2016
BR29	<i>Digitaria sanguinalis</i>	1989	Brazil	Chiapello et al., 2015
NP10-28-1-1-1	<i>Eleusine coracana</i>	1975	Pokara, Nepal	Hau et al., 2007
Ken15-15-1	<i>Eleusine coracana</i>	1976	Tokyo, Japan	Hau et al., 2007
MZ5-1-6	<i>Eleusine coracana</i>	1976	Miyazaki, Japan	Hau et al., 2007
G10-1	<i>Eleusine coracana</i>	1977	Tochigi, Japan	Hau et al., 2007
GFEC1-5-1	<i>Eleusine coracana</i>	1977	Gifu, Japan	Hau et al., 2007
IN77-31-1-1	<i>Eleusine coracana</i>	1977	Bangalor, India	Hau et al., 2007
IN77-39-1-2	<i>Eleusine coracana</i>	1977	Bangalor, India	Hau et al., 2007
Z2-1	<i>Eleusine coracana</i>	1977	Nagawa, Japan	Hau et al., 2007
SZEC1-1-1	<i>Eleusine coracana</i>	1978	Shizuoka, Japan	Hau et al., 2007
IN77-36-1-1	<i>Eleusine indica</i>	1977	Mysor, India	Hau et al., 2007
CD156	<i>Eleusine indica</i>	1989	Ferkessedougou, Ivory Cost	Chiapello et al., 2015
Br62	<i>Eleusine indica</i>	1991	Brazil	Gladioux et al., 2017
TH16	<i>Hordeum vulgare</i>	1987	Thailand	Chiapello et al., 2015
WK3-1	<i>Lolium perenne</i>	1996	Yamagushi, Japan	Hau et al., 2007
TP1	<i>Lolium perenne</i>	1997	Tochigi, Japan	Hau et al., 2007
TP2	<i>Lolium perenne</i>	1997	Tochigi, Japan	Hau et al., 2007
AK1	<i>Lolium perenne</i>	1998	Akita, Japan	Hau et al., 2007
FI5	<i>Lolium perenne</i>	1998	Chiba, Japan	Hau et al., 2007
PGPA	<i>Lolium perenne</i>	1998	Pennsylvania, USA	Maciel et al. 2014
LW3	<i>Lolium perenne</i>	1999	Yamanashi, Japan	Hau et al., 2007
PgKY	<i>Lolium perenne</i>	2000	Kentucky, USA	Maciel et al. 2014
Hoku1	<i>Oryza sativa</i>	1948	Japan	Yoshida et al., 2009
P-2b	<i>Oryza sativa</i>	1948	Aichi, Japan	Yoshida et al., 2009
Ken53-33	<i>Oryza sativa</i>	1953	Aichi, Japan	Yoshida et al., 2009
Ken54-04	<i>Oryza sativa</i>	1954	Japan	Hau et al., 2007
Ina72	<i>Oryza sativa</i>	1957	Japan	Yoshida et al., 2009
Ina168	<i>Oryza sativa</i>	1958	Aichi, Japan	Yoshida et al., 2009
PO-02-7306	<i>Oryza sativa</i>	1973	Indonesia	This study
PO-12-7301	<i>Oryza sativa</i>	1973	Indonesia	This study
PO-12-7301-2	<i>Oryza sativa</i>	1973	Indonesia	Farman et al., 2002
PO-02-7501	<i>Oryza sativa</i>	1975	Indonesia	Farman et al., 2002
PO-04-7501	<i>Oryza sativa</i>	1975	Indonesia	Farman et al., 2002
0903-4	<i>Oryza sativa</i>	1976	Japan	This study
1836-3	<i>Oryza sativa</i>	1976	Niigata, Japan	Yoshida et al., 2009
88A	<i>Oryza sativa</i>	1976	Ibaraki, Japan	Yoshida et al., 2009
Jan-03	<i>Oryza sativa</i>	1976	Japan	This study
Jan-12	<i>Oryza sativa</i>	1976	Japan	This study
PH14	<i>Oryza sativa</i>	1980	Los Baños, Philippines	Chiapello et al., 2015
FR13	<i>Oryza sativa</i>	1988	France	Chiapello et al., 2015
GY11	<i>Oryza sativa</i>	1988	French Guyana	Chiapello et al., 2015
CHNOS59-6-11	<i>Oryza sativa</i>	1989	China	Farman et al., 2002
CHNOS60-8-1	<i>Oryza sativa</i>	1989	China	Farman et al., 2002
Br10	<i>Oryza sativa</i>	1990	Brazil	Yoshida et al., 2009
Br13	<i>Oryza sativa</i>	1990	Brazil	Farman et al., 2002
Br15	<i>Oryza sativa</i>	1990	Brazil	Farman et al., 2002
VHG4.5	<i>Oryza sativa</i>	1996	Vietnam	Hau et al., 2007
VHT3.3	<i>Oryza sativa</i>	1998	Vietnam	Hau et al., 2007
VHT6.1	<i>Oryza sativa</i>	1998	Vietnam	Hau et al., 2007
VTB6.1	<i>Oryza sativa</i>	1998	Vietnam	Hau et al., 2007
RB-3a	<i>Oryza sativa</i>	2017	Bangladesh	This study
RBK-1	<i>Oryza sativa</i>	2017	Bangladesh	This study
RB13b	<i>Oryza sativa</i>	2018	Khulna, Bangladesh	This study
RBCh1814-1	<i>Oryza sativa</i>	2018	Chuadanga, Bangladesh	This study
RBD1831	<i>Oryza sativa</i>	2018	Dinajpur, Bangladesh	This study
RBMe1819	<i>Oryza sativa</i>	2018	Meherpur, Bangladesh	This study
RBNi1830-1	<i>Oryza sativa</i>	2018	Nilfamari, Bangladesh	This study
RBRa1834	<i>Oryza sativa</i>	2018	Rangpur, Bangladesh	This study
RBRa1836-3	<i>Oryza sativa</i>	2018	Rangpur, Bangladesh	This study
RBTa1847	<i>Oryza sativa</i>	2018	Tangail, Bangladesh	This study
RBTa1850-13	<i>Oryza sativa</i>	2018	Tangail, Bangladesh	This study
NI922	<i>Panicum bisulcatum</i>	1974	Japan	This study
SZPM1-1-1	<i>Panicum miliaceum</i>	1978	Shizuoka, Japan	Hau et al., 2007
STPM1-3-2	<i>Panicum miliaceum</i>	1981	Saitama, Japan	Hau et al., 2007
STPM4-2-2	<i>Panicum miliaceum</i>	1981	Saitama, Japan	Hau et al., 2007
YNPM4-1-1	<i>Panicum miliaceum</i>	1983	Yamanashi, Japan	Hau et al., 2007

NNPM1-2-1	<i>Panicum miliaceum</i>	1984	Nagano, Japan	Hau et al., 2007
NRPM1-1-1	<i>Panicum miliaceum</i>	1990	Japan	This study
BTRp-5	<i>Panicum repens</i>	2016	Gazipur, Bangladesh	This study
BTRp-6	<i>Panicum repens</i>	2016	Gazipur, Bangladesh	This study
GFS11-7-2	<i>Setaria italica</i>	1977	Japan	Hau et al., 2007
IN77-16-1-1	<i>Setaria italica</i>	1977	India	Hau et al., 2007
IN77-20-1-1	<i>Setaria italica</i>	1977	India	Hau et al., 2007
NRS12-2-2	<i>Setaria italica</i>	1977	Japan	This study
NRS13-1-1	<i>Setaria italica</i>	1977	Japan	Hau et al., 2007
NNS13-2-1	<i>Setaria italica</i>	1984	Japan	Hau et al., 2007
NI913	<i>Setaria viridis</i>	1974	Japan	This study
KANSV1-4-1	<i>Setaria viridis</i>	1975	Kanagawa, Japan	Yoshida et al., 2016
US71	<i>Setaria viridis</i>	1998	Kentucky, USA	Chiapello et al., 2015
Br130.8	<i>Triticum aestivum</i>	1990	Mato Grosso do Sul, Brazil	Gladioux et al., 2017
Br3	<i>Triticum aestivum</i>	1990	Londrina, Brazil	Hau et al., 2007
Br46	<i>Triticum aestivum</i>	1990	Brazil	This study
Br49	<i>Triticum aestivum</i>	1990	Dourados, Brazil	Hau et al., 2007
Br50	<i>Triticum aestivum</i>	1990	Brazil	This study
Br8	<i>Triticum aestivum</i>	1990	B. V. Paraiso, Brazil	Hau et al., 2007
BR32	<i>Triticum aestivum</i>	1991	Brazil	Chiapello et al., 2015
Br117.1	<i>Triticum aestivum</i>	1992	Brazil	This study
Br127.1	<i>Triticum aestivum</i>	1992	Brazil	This study
PY5003	<i>Triticum aestivum</i>	2005	Londrina, PR, Brazil	Maciel et al. 2014
PY5010	<i>Triticum aestivum</i>	2005	Londrina, PR, Brazil	Maciel et al. 2014
PY5033	<i>Triticum aestivum</i>	2005	Londrina, PR, Brazil	Maciel et al. 2014
PY6017	<i>Triticum aestivum</i>	2006	Coromandel, MGG, Brazil	Maciel et al. 2014
PY6025	<i>Triticum aestivum</i>	2006	Coromandel, MGG, Brazil	Maciel et al. 2014
PY6045	<i>Triticum aestivum</i>	2006	Goiania, Brazil	Maciel et al. 2014
PY36.1	<i>Triticum aestivum</i>	2007	Brasilia, DF, Brazil	Maciel et al. 2014
PY86.1	<i>Triticum aestivum</i>	2008	Cascavel, PR, Brazil	Maciel et al. 2014
PY0925	<i>Triticum aestivum</i>	2009	Perdizes, MG, Brazil	Maciel et al. 2014
BTBa-B2	<i>Triticum aestivum</i>	2016	Gazipur, Bangladesh	This study
BTBa-B1	<i>Triticum aestivum</i>	2016	Gazipur, Bangladesh	This study
BTJP-1(a)	<i>Triticum aestivum</i>	2016	Bangladesh	This study
BTJP-4-1	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4(1)	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4(3)	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4c	<i>Triticum aestivum</i>	2016	Bangladesh	This study
BTJP-9c	<i>Triticum aestivum</i>	2016	Bangladesh	This study
BTJP-4-11	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4-12	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4-16	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4-18	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4-3	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4-4	<i>Triticum aestivum</i>	2016	Bangladesh	This study
BTJP-4-5	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4-5	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTJP-4-6	<i>Triticum aestivum</i>	2016	Jhenaidah, Bangladesh	This study
BTMaV-10(a)	<i>Triticum aestivum</i>	2016	Bangladesh	This study
BTMB-5(0)	<i>Triticum aestivum</i>	2016	Bangladesh	This study
BTMP-S13-1	<i>Triticum aestivum</i>	2016	Meherpur, Bangladesh	This study
BTMP-S13-2	<i>Triticum aestivum</i>	2016	Meherpur, Bangladesh	This study
s12_transcripts	<i>Triticum aestivum</i>	2016	Nittanandapur, Bangladesh	Islam et al., 2016
s5_transcripts	<i>Triticum aestivum</i>	2016	Meherpur, Bangladesh	Islam et al., 2016
BTGS-6e	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGP-6(c-4)	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGP-6(c-7)	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGP-6e	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGP1-b	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGS-6(b-2)	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGS-6f	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGS-6g	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTGS-6h	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTMaU-10(a)	<i>Triticum aestivum</i>	2017	Magura, Bangladesh	This study
BTMaU-10(b)	<i>Triticum aestivum</i>	2017	Magura, Bangladesh	This study
BTMP-2g	<i>Triticum aestivum</i>	2017	Meherpur, Bangladesh	This study
BTMP-182-5	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1827-1	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1827-2	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1828-4	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1828-5	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1830-1	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1830-4	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1831-5	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-184-3	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study

BTMP-1843-1	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1845-1	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-1845-3	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-186-3	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-186-4	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-187-3	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-189-1	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
BTMP-189-2	<i>Triticum aestivum</i>	2018	Meherpur, Bangladesh	This study
ZMW18_06	<i>Triticum aestivum</i>	2018	Mpika, Zambia	Tembo et al., 2020
ZMW18_08	<i>Triticum aestivum</i>	2018	Mpika, Zambia	Tembo et al., 2020
ZMW18_10	<i>Triticum aestivum</i>	2018	Mpika, Zambia	Tembo et al., 2020
ZMW18_11	<i>Triticum aestivum</i>	2018	Mpika, Zambia	Tembo et al., 2020
BTMP-191-2	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-1910-3	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-192-3	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-193-3	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-194-1	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-194-2	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-195-1	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-195-2	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-196-2	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-197-3	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-198-1	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-199-3	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
BTMP-199-7	<i>Triticum aestivum</i>	2019	Meherpur, Bangladesh	This study
ZMW19_09	<i>Triticum aestivum</i>	2019	Mt. Makulu, Zambia	This Study
ZMW19_13	<i>Triticum aestivum</i>	2019	Mt. Makulu, Zambia	This Study
ZMW19_17	<i>Triticum aestivum</i>	2019	Mt. Makulu, Zambia	This Study
BTMP-202-1	<i>Triticum aestivum</i>	2020	Kutubpur, Bangladesh	This study
BTMP-202-4	<i>Triticum aestivum</i>	2020	Kutubpur, Bangladesh	This study
BTMP-203-5	<i>Triticum aestivum</i>	2020	Meherpur, Bangladesh	This study
BTMP-204-2	<i>Triticum aestivum</i>	2020	Meherpur, Bangladesh	This study
BTMP-204-9	<i>Triticum aestivum</i>	2020	Meherpur, Bangladesh	This study
BTMP-206-5	<i>Triticum aestivum</i>	2020	Meherpur, Bangladesh	This study
BTMP-208-6	<i>Triticum aestivum</i>	2020	Kutubpur, Bangladesh	This study
BTMP-2801-1	<i>Triticum aestivum</i>	2020	Meherpur, Bangladesh	This study
BTMP-2801-5	<i>Triticum aestivum</i>	2020	Meherpur, Bangladesh	This study
BTMP-2807-4	<i>Triticum aestivum</i>	2020	Meherpur, Bangladesh	This study
ZMW20_03	<i>Triticum aestivum</i>	2020	Mpika, Zambia	This Study
ZMW20_04	<i>Triticum aestivum</i>	2020	Mpongwe, Zambia	This Study
ZMW20_07	<i>Triticum aestivum</i>	2020	Mpongwe, Zambia	This Study
ZMW20_14	<i>Triticum aestivum</i>	2020	Mt. Makulu, Zambia	This Study
ZMW20_15	<i>Triticum aestivum</i>	2020	Mpongwe, Zambia	This Study
ZMW20_16	<i>Triticum aestivum</i>	2020	Mpika, Zambia	This Study

Table 3. Overview of *Magnaporthe oryzae* isolate collection used in this report.

Host of origin	Number of isolates	Country of origin	Year(s) of collection
<i>Avena sativa</i>	1	Brazil	1990
<i>Digitaria sanguinalis</i>	1	Brazil	1989
<i>Eleusine coracana</i>	9	India, Japan, Nepal	1975-1978
<i>Eleusine indica</i>	3	Brazil, India, Ivory Coast	1977-1991
<i>Hordeum vulgare</i>	1	Thailand	1987
<i>Lolium perenne</i>	8	Japan, USA	1996-2000
<i>Oryza sativa</i>	39	Bangladesh, Brazil, China, France, French Guyana, Indonesia, Japan, Philippines, Vietnam	1954-2018
<i>Panicum bisulcatum</i>	1	Japan	1974
<i>Panicum miliaceum</i>	6	Japan	1974-1990
<i>Panicum repens</i>	2	Bangladesh	2016
<i>Setaria italica</i>	6	India, Japan	1977-1984
<i>Setaria viridis</i>	3	Japan, USA	1974-1998
<i>Triticum aestivum</i>	18	Brazil	1990-2009
<i>Triticum aestivum</i>	75	Bangladesh	2016-2020
<i>Triticum aestivum</i>	13	Zambia	2018-2020
Total	186		1954-2020