**SUPPLEMENTARY**

**Supplementary Table S1. Primers used in the amplification reactions for locus and amplification programs.** 1F = forward and R = reverse primers. 2 PCR program annealing temperature (AT) for each gene.

**Table S1.** Primers used in amplification reactions for locus and amplification programs.

|  |  |  |
| --- | --- | --- |
| **Locus and primer name1** | **Sequence (5'-3')** | **PCR program2** |
| **Actin** |  | Initial denaturation at 94 °C for 5 min, followed by 35 cycles of 94 °C for 30 s, 63,8 °C (AT) for 20 s, 72 °C for 30 s and finally an additional 10 min at 72 °C. |
| ACT-512 F | ATG TGC AAG GCC GGT TTC GC |
| ACT-783 R | TAC GAG TCC TTC TGG CCC AT |
| **Calmodulin** |  | Initial denaturation at 94 °C for 5 min, followed by 35 cycles of 94 °C for 30 s, 55°C (AT) for 20 s, 72 °C for 30 s and finally an additional 10 min at 72 °C. |
| CAL-228 F | GAG TTC AAG GAG GCC TTC TCC C |
| CAL-737 R | CAT CTT TCT GGC CAT CAT GG |
| **ITS** |  | Initial denaturation at 94 °C for 5 min, followed by 35 cycles of 94 °C for 30 s, 57,8 °C (AT) for 20 s, 72 °C for 30 s and finally an additional 10 min at 72 °C. |
| ITS4 R | TCC TCC GCT TAT TGA TAT GC |
| V9G F | TTA CGT CCC TGC CCT TTG TA |
| **LSU** |  | Initial denaturation at 94 °C for 5 min, followed by 35 cycles of 94 °C for 30 s, 51,1 °C (AT) for 20 s, 72 °C for 30 s and finally an additional 10 min at 72 °C. |
| LR5 R | TCC TGA GGG AAA CTT CG |
| NL1 F | GCA TAT CAA TAA GCG GAG GAA AAG |
| **RPB1** |  | Initial denaturation at 94 °C for 5 min, followed by 35 cycles of 94 °C for 30 s, 57 °C (AT) for 20 s, 72 °C for 30 s and finally an additional 10 min at 72 °C. |
| RPB1 F | AGA CGA TYG AGG AGA TCC AGT T |
| RPB1 R | ART CCA CAC GCT TAC CCA TC |

1 F = forward and R = reverse primers.

2 PCR program annealing temperature (AT) for each gene.

**Supplementary Table S2. Genome assemblies of different *Pyricularia* isolates previously classified into host-specific lineages to use as phylogenetic references (Gladieux et al. 2018).**

**S2 Table.** Genome assemblies of different *Pyricularia* isolates previously classified into host-specific lineages to use as phylogenetic references (Gladieux et al., 2018).

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| ***Isolate ID*** | ***Location*** | ***lineage (Gladieux et al 2018)*** | ***Host*** | ***database/accession*** |
| 87-120 | Unknown, Unknown | Oryza | *Oryza sativa* | SAMN08377452 |
| Arcadia | North America, Kentucky, USA | Setaria | *Setaria viridis* | SAMN08009540 |
| B2 | South America, Bolivia | Triticum | *Triticum aestivum* | SAMN05580113 |
| B51 | South America, Quirusillas, Bolivia | Eleusine1 | *Eleusine indica* | SAMN08009542 |
| B71 | South America, Bolivia | Triticum | *Triticum aestivum* | SAMN04942725 |
| Bd8401 | Asia, Philippines | Brachiaria1 | *Brachiaria distachya* | SAMN08009543 |
| BdBar | Asia, Barisal, Bangladesh | Triticum | *Triticum aestivum* | SAMN04940126 |
| BdJes | Asia, Jessore, Bangladesh | Triticum | *Triticum aestivum* | SAMN04942531 |
| BdMeh | Asia, Mehepur, Bangladesh | Triticum | *Triticum aestivum* | SAMN04942534 |
| Bm88324 | Asia, Philippines | Brachiaria2 | *Brachiaria mutica* | SAMN08009544 |
| Br130 | South America, Mato Grosso do Sul, Brazil | Triticum | *Triticum aestivum* | SAMN08009547 |
| BR29 | South America, Brazil | NA | *Digitaria sanguinalis* | http://genome.jouy.inra.fr/gemo/ |
| BR32 | South America, Brazil | Triticum | *Triticum aestivum* | http://genome.jouy.inra.fr/gemo/ |
| Br7 | South America, Parana, Brazil | Triticum | *Triticum aestivum* | SAMN08009545 |
| Br80 | South America, Brazil | Triticum | *Triticum aestivum* | SAMN08009546 |
| CD156 | Africa, Ferkessedougou, Ivory Coast | Eleusine1 | *Eleusine indica* | http://genome.jouy.inra.fr/gemo/ |
| CHRF | North America, Maryland, USA | Lolium | *Lolium perenne* | SAMN08009548 |
| CHW | North America, Maryland, USA | Lolium | *Lolium perenne* | SAMN08009549 |
| DsLIZ | North America, Kentucky, USA | NA | *Digitaria sanguinalis* | SAMN08009550 |
| EI9411 | Asia, Fujian, China | Eleusine2 | *Eleusine indica* | GCA\_001548775.1 |
| EI9604 | Asia, Fujian, China | Eleusine1 | *Eleusine indica* | GCA\_001548785.1 |
| FH | North America, Maryland, USA | Lolium | *Lolium perenne* | SAMN08009551 |
| FR13 | Europe, France | Oryza | *Oryza sativa* | http://genome.jouy.inra.fr/gemo/ |
| G17 | Asia, Japan | Eragrostis | *Eragrostis curvula* | SAMN08009553 |
| G22 | Asia, Japan | Eleusine2 | *Eleusine coracana* | SAMN08009554 |
| GG11 | North America, Kentucky, USA | Lolium | *Lolium perenne* | SAMN08009555 |
| GrF52 | North America, Kentucky, USA | Setaria | *Setaria viridis* | SAMN08009556 |
| GY11 | South America, French Guyana | Oryza | *Oryza sativa* | http://genome.jouy.inra.fr/gemo/ |
| HO | North America, Pennsylvania, USA | Lolium | *Lolium perenne* | SAMN08009558 |
| IA1 | North America, Arkansas, USA | Oryza | *Oryza sativa* | SAMN08009559 |
| IB33 | North America, Texas, USA | Oryza | *Oryza sativa* | SAMN08009560 |
| IB49 | North America, Arkansas, USA | Oryza | *Oryza sativa* | SAMN08009561 |
| IC17 | North America, Arkansas, USA | Oryza | *Oryza sativa* | SAMN08009562 |
| IE1k | North America, Arkansas, USA | Oryza | *Oryza sativa* | SAMN08009563 |
| LpKY97 | North America, Kentucky, USA | Lolium | *Lolium perenne* | SAMN08009564 |
| ML33 | Africa, Mali | Oryza | *Oryza sativa* | SAMN08009565 |
| P131 | Asia, Japan | Oryza | *Oryza sativa* | GCA\_000292605.1 |
| P28 | Paraguay, Paraguay | Lolium | *Bromus tectorum* | SAMN05864041 |
| P29 | Paraguay, Paraguay | Triticum | *Bromus tectorum* | SAMN05898532 |
| P3 | Canindeyu, Paraguay | Triticum | *Triticum durum* | SAMN08009568 |
| Pg1213-22 | North America, Georgia, USA | Lolium | *Festuca arundinaceum* | SAMN08009569 |
| PH14-rn | Asia, Philippines | Oryza | *Oryza sativa* | http://genome.jouy.inra.fr/gemo/ |
| PH42 | Asia, Philippines | Eleusine1 | *Eleusine coracana* | SAMN08009570 |
| PL2-1 | North America, Kentucky, USA | Lolium | *Lolium multiflorum* | SAMN08009571 |
| PL3-1 | North America, Kentucky, USA | Lolium | *Lolium multiflorum* | SAMN08009572 |
| PM1 | North America, Georgia, USA | NA | *Pennisetum americanum* | SAMN08377453 |
| SSFL02 | North America, Florida, USA | Stenotaphrum | *Stenotaphrum secundatum* | SAMN08009573 |
| SSFL14-3 | North America, Florida, USA | Stenotaphrum | *Stenotaphrum secundatum* | SAMN08009574 |
| Sv9610 | Asia, Zhejian, China | Setaria | *Setaria viridis* | GCA\_001548845.1 |
| Sv9623 | Asia, Zhejian, China | Setaria | *Setaria viridis* | GCA\_001548855.1 |
| T25 | South America, Parana, Brazil | Triticum | *Triticum aestivum* | SAMN08009575 |
| TF05-1 | North America, Kentucky, USA | Lolium | *Festuca arundinaceum* | SAMN08009576 |
| TH12-rn | Asia, Thailand | Oryza | *Hordeum vulgare* | http://genome.jouy.inra.fr/gemo/ |
| TH16 | Asia, Thailand | Oryza | *Hordeum vulgare* | http://genome.jouy.inra.fr/gemo/ |
| US71 | North America, USA | Setaria | *Setaria spp.* | http://genome.jouy.inra.fr/gemo/ |
| VO107 | North America, Texas, USA | NA | *Digitaria sanguinalis* | SAMN08009577 |
| WBKY11 | North America, Kentucky, USA | Lolium | *Triticum aestivum* | SAMN08009578 |
| WHTQ | South America, Brazil | Triticum | *Triticum aestivum* | SAMN08009580 |
| Y34 | Asia, Yunnan, China | Oryza | *Oryza sativa* | GCA\_000292585.1 |

**Supplementary Table S3. Pathogenicity experimental dataset by isolates of *Pyricularia oryzae-Triticum* (n = 5), *Pyricularia oryzae-*Other (n = 4), *P. pennisetigena* (n = 2) and *P. grisea* (n = 2) by cultivar (susceptible and resistant).**

|  |  |  |
| --- | --- | --- |
| **Species** | **Isolate ID** | **Study year** |
| *Pyricularia oryzae-Triticum* | CTae005 | Two |
| CTae009 | Two |
| ATae039 | Two |
| GTae019 | Two |
| GTae021 | Two |
| *Pyricularia oryzae-Other* | CLmi007 | One |
| IRrh54 | One |
| SLSsh051 | One |
| YEih113 | One |
| *Pyricularia pennisetigena* | YCeh55 | One |
| ITCeh117 | One |
| *Pyricularia grisea* | YDih056 | One |
| ITDsph107 | One |

**Supplementary Table S4. Morphological Measurement Data of Pyricularia Isolates**

1. Macroscopic and microscopic characteristics of the *Pyricularia pennisetigena* isolates

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Species** | **Isolate** | **Microscopic caracteristic** | | | | | | | **Macroscopic caracteristic** | | | | | |
| **Conidia** | | **Hilum** | | **Microconidia** | | **Hyphae** | **Oatmeal Agar (OA)** | | | **Potato Dextrose Agar (PDA)** | | |
| **Long** | **wide** | **Long** | **wide** | **Long** | **wide** |  | **R1** | **R2** | **Average** | **R1** | **R2** | **Average** |
| *Pyricularia pennisetigena* | YCeh055 | 32.47 | 9.38 | 1.63 | 2.32 | no | no | 3.95 | 4.5 | 4.75 | **4.625** | 2.5 | 2.75 | **2.625** |
|  | ITCeh117 | 33.64 | 9.42 | 1.71 | 2.25 | no | no | 4.37 | 4.8 | md | **4.8** | 5 | 5.3 | **5.15** |
|  | **Average** | **33.06** | **9.40** | **1.67** | **2.29** |  |  | **4.16** |  |  |  |  |  |  |
|  | **Max** | **33.64** | **9.42** | **1.71** | **2.29** |  |  | **4.37** |  |  |  |  |  |  |
|  | **Min** | **33.06** | **9.40** | **1.67** | **2.29** |  |  | **4.16** |  |  |  |  |  |  |

no = not observed; nd = not determined; md = missing data. Microscopic (µm) = mean, measurements were taken on 10 conidia in triplicate; average calculate per specie. Macroscopic, growth diameter 5 days after inoculation on medium (cm) = R1-R2 (Replicates plate), average calculate per replicate.

1. Macroscopic and microscopic characteristics of the *Pyricularia oryzae* isolates

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Species** | **Isolate** | **Microscopic caracteristic** | | | | | | | **Macroscopic caracteristic** | | | | | |
| **Conidia** | | **Hilum** | | **Microconidia** | | **Hyphae** | **Oatmeal Agar (OA)** | | | **Potato Dextrose Agar (PDA)** | | |
| **Long** | **wide** | **Long** | **wide** | **Long** | **wide** |  | **R1** | **R2** | **Average** | **R1** | **R2** | **Average** |
| *Pyricularia oryzae* | CLmi007 | 31.15 | 10.23 | 1.41 | 2.13 | no | no | nd | 1.575 | 1.6 | **1.5875** | 2 | 2.05 | **2.025** |
|  | CBch004 | 28.18 | 10.01 | 1.24 | 1.80 | no | no | nd | 1.8 | 1.3 | **1.55** | 1.75 | 2.1 | **1.925** |
|  | CTae009 | 28.54 | 9.09 | 1.42 | 2.32 | no | no | 4.00 | 1.95 | 2.25 | **2.1** | 2.9 | 2.95 | **2.925** |
|  | CTae005 | 33.08 | 10.14 | 1.54 | 2.11 | no | no | 4.00 | 2.05 | 2.25 | **2.15** | 2.8 | 2.7 | **2.75** |
|  | IAsh017 | 26.89 | 10.22 | 1.18 | 1.93 | no | no | nd | 1.9 | 2.25 | **2.075** | 2.25 | 2.25 | **2.25** |
|  | GTae019 | 29.02 | 9.10 | 1.30 | 2.40 | no | no | nd | 1.56 | 1.55 | **1.555** | 2 | 1.75 | **1.875** |
|  | GTae021 | 26.18 | 9.43 | 1.37 | 2.20 | no | no | 3.87 | 2.25 | 2.13 | **2.19** | 2.1 | 2 | **2.05** |
|  | SLSsh051 | 33.73 | 8.67 | 1.96 | 3.30 | no | no | 2.82 | 1.8 | 1.95 | **1.875** | 1.8 | 1.9 | **1.85** |
|  | YTae028 | 30.36 | 7.72 | 1.79 | 2.33 | no | no | 3.68 | 2 | 2 | **2** | 2.6 | 2.5 | **2.55** |
|  | IRrh054 | 29.97 | 9.69 | 1.20 | 2.25 | no | no | 3.10 | 4.5 | 4.3 | **4.4** | 5.5 | 5.6 | **5.55** |
|  | YEih113 | 28.57 | 9.29 | 1.49 | 1.69 | no | no | nd | 2.7 | 2.5 | **2.6** | 3.05 | 3 | **3.025** |
|  | ATae039 | 28.20 | 9.63 | 1.49 | 1.71 | 11.11 | 2.04 | 3.87 | 2.25 | 1.35 | **1.8** | 2.35 | 2.35 | **2.35** |
|  | YTae025 | 30.55 | 7.85 | 1.72 | 2.05 | no | no | 4.00 | 2.5 | 2.6 | **2.55** | 2.6 | 2.4 | **2.5** |
|  | IAsh016 | 26.47 | 8.08 | 1.42 | 2.18 | no | no | nd | 1.85 | 1.75 | **1.8** | 2.25 | 2.25 | **2.25** |
|  | ITTae101 | 29.66 | 7.60 | 2.20 | 2.23 | no | no | 4.03 | 4.5 | 4.5 | **4.5** | 4.8 | md | **4.8** |
|  | **Average** | **29.37** | **9.12** | **1.52** | **2.18** |  |  | **3.71** |  |  |  |  |  |  |
|  | **Max** | **33.73** | **10.23** | **2.20** | **3.30** |  |  | **4.03** |  |  |  |  |  |  |
|  | **Min** | **26.18** | **7.60** | **1.18** | **1.69** |  |  | **2.82** |  |  |  |  |  |  |

no = not observed; nd = not determined; md = missing data. Microscopic (µm) = mean, measurements were taken on 10 conidia in triplicate; average calculate per specie. Macroscopic, growth diameter 5 days after inoculation on medium (cm) = R1-R2 (Replicates plate), average calculate per replicate.

1. Macroscopic and microscopic characteristics of the *Pyricularia grisea*  isolates

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Species** | **Isolate** | **Microscopic caracteristic** | | | | | | | **Macroscopic caracteristic** | | | | | |
| **Conidia (n = 10)** | | **Hilum (n = 10)** | | **Microconidia** | | **Hyphae** | **Oatmeal Agar (OA)** | | | **Potato Dextrose Agar (PDA)** | | |
| **Long** | **wide** | **Long** | **wide** | **Long** | **wide** |  | **R1** | **R2** | **Average** | **R1** | **R2** | **Average** |
| *Pyricularia grisea* | YDhh012 | 23.31 | 8.84 | 1.18 | 2.14 | 9.09 | 1.66 | 2.79 | 2.55 | 2.45 | **2.50** | 2.65 | 1.90 | **2.28** |
|  | YBsph044 | 31.98 | 8.62 | 1.11 | 2.19 | no | no | 3.45 | 2.30 | 2.15 | **2.23** | 2.50 | 2.40 | **2.45** |
|  | YDih056 | 32.47 | md | nd | nd | no | no | nd | 4.10 | 3.40 | **3.75** | 5.70 | 5.00 | **5.35** |
|  | ITDsph107 | 29.48 | 8.65 | 1.38 | 2.48 | no | no | nd | 2.10 | 2.70 | **2.40** | 2.35 | 2.80 | **2.58** |
|  | YDhh014 | nd | nd | nd | nd | 7.39 | 2.13 | 2.76 | 1.70 | 1.75 | **1.73** | 1.40 | 1.60 | **1.50** |
|  | YDhh105 | 28.94 | md | nd | nd | 7.39 | 2.13 | 2.76 | 3.50 | 3.50 | **3.50** | 5.50 | 5.49 | **5.50** |
|  | **Average** | **29.31** | **8.70** | **1.22** | **2.27** | **7.96** | **1.97** | **2.94** |  |  |  |  |  |  |
|  | **Max** | **32.47** | **8.84** | **1.38** | **2.48** | **9.09** | **2.13** | **3.45** |  |  |  |  |  |  |
|  | **Min** | **23.31** | **8.62** | **1.11** | **2.14** | **7.39** | **1.66** | **2.76** |  |  |  |  |  |  |

no = not observed; nd = not determined; md = missing data. Microscopic (µm) = mean, measurements were taken on 10 conidia in triplicate; average calculate per specie. Macroscopic, growth diameter 5 days after inoculation on medium (cm) = R1-R2 (Replicates plate), average calculate per replicate.

**Supplementary Figure S1.** **Normality and homoscedasticity of variances according to residual simulation diagnostics.** **Left**: QQ plot of the simulated residuals, comparing observed versus expected values by Kolmogorov-Smirnov (KS) indicate to dispersion and outlier tests. **Right:** Scaled residuals reveal the homoscedasticity of variance across levels of the predictor, confirmed by the Levene Test. **A**. Incidence (I): Tests KS indicate significant deviations from uniformity or issues with over/under-dispersion and with the Levene Test confirming no homogeneity of variances or uniformity within the groups. **B.** Disease severity index (DSI): Tests KS indicate no significant deviations from uniformity or issues with over/under-dispersion but with the Levene Test confirming no homogeneity of variances or uniformity within the groups.

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**Supplementary Data S1. Sequence alignments were concatenated into a single matrix used in Phylogenetic analyses.**

Available: https://1drv.ms/f/s!AlULMAeMaNXegoEgKhMb8M5ZJZ4aNg