




First report of *Lasiodiplodia brasiliense* causing maize stalk rot

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Abstract

Maize stalk rot can lead to major corn yield losses in Brazil and worldwide. Maize stalks exhibiting tiny black specks or light brown lesions were collected in the city of Luís Eduardo Magalhães. Based on morphological characteristics and molecular assays, the fungus was identified as *Lasiodiplodia brasiliense*. This is the first report of maize stalk rot caused by *L. brasiliense*.

Keywords Corn · *Zea mays* · Brazil · Botryosphaeriaceae

In June 2016, samples of maize stalks (*Zea mays*) exhibiting tiny black specks (pynidia) or light brown lesions and dark brown pith tissue in the lower internodes (Fig. 1) were collected in the city of Luís Eduardo Magalhães, state of Bahia, which is an important maize producing area in Brazil. The samples were submitted to the Plant Pathology Laboratory at the National Maize and Sorghum Research Center (EMBRAPA), located in the Brazilian state of Minas Gerais for analysis. The symptoms were typical of maize stalk rot (SR) disease in Brazil, a disease mainly caused by the following fungi: *Colletotrichum graminicola*, *Macrophomina phaseolina*, *Fusarium graminearum*, *F. verticillioides* (Syn. *F. moniliforme*), *Stenocarpella macrospora*, *S. maydis*, and *Phaeocytostroma ambiguum*. For fungal isolation, stalk fragments were removed from the surface bordering the lesions and disinfested in 0.5% sodium hypochlorite for 2 min before plating on an oatmeal agar medium (OMA) with tetracycline (20 mg.L⁻¹). The colonies produced after three weeks were light to dark grey and globose pynidia were easily produced on the OMA medium (Fig. 1). Paraphyses were hyaline, aseptate and rounded at the apex. Conidia were hyaline, aseptate, thick-walled, ellipsoid to ovoid and rounded in the apex during the immature phase, and becoming pigmented, ellipsoid to ovoid, with one single septum with longitudinal striations and sizes ranging from 20 to 30 µm long, and 10 to 16 µm wide ($n = 50$) at the maturity (Fig. 1). These morphological characteristics are very similar to the description of *Lasiodiplodia brasiliense* (Netto et al. 2014). The monoconidial isolate (CFMS 1342) was deposited in the

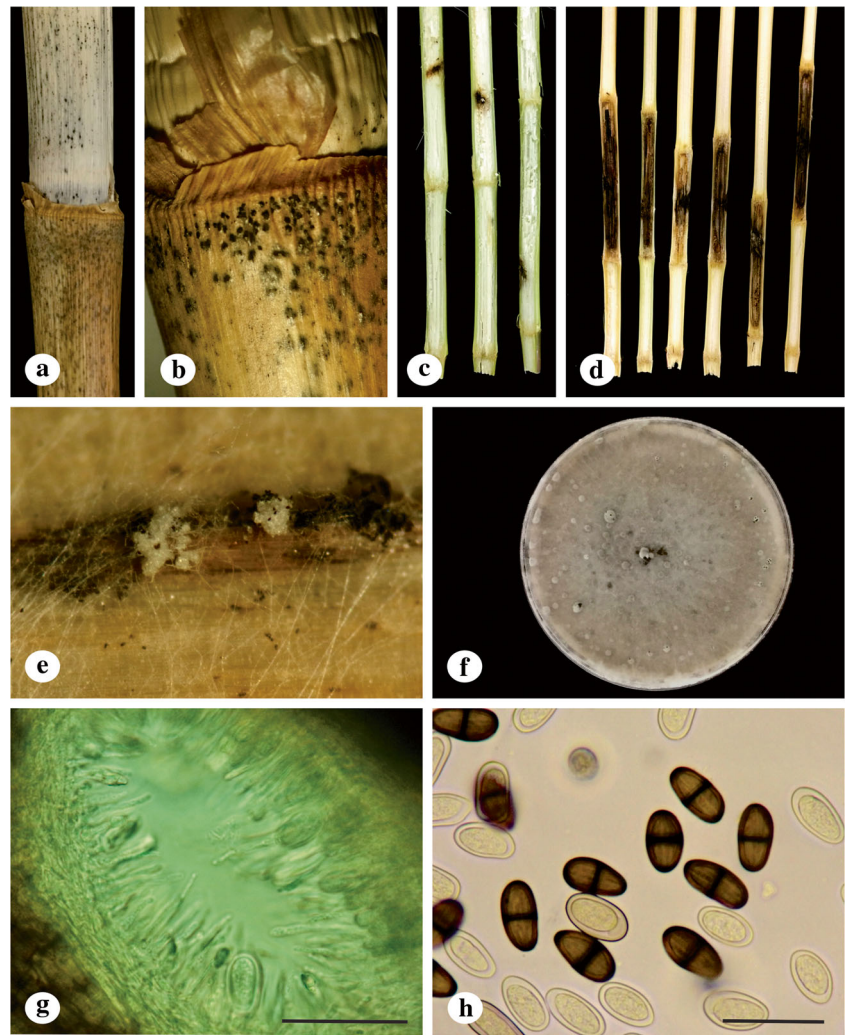
Plant Pathogens Collection of EMBRAPA Maize and Sorghum, and the DNA of the internal transcribed spacer (ITS) region of the nuclear ribosomal DNA and the translation elongation factor 1-alpha gene (TEF1- α) (Carbone and Kohn 1999; White et al. 1990) were partially sequenced and deposited in GenBank with accession numbers MF952733 and MF952734, respectively. The isolate showed 99% identity with the type species of *L. brasiliense* for both the ITS region (accession JX464063) and TEF1- α gene (JX464049).

For the pathogenicity test in maize, a toothpick previously immersed in a spore suspension (10⁶ conidia mL⁻¹) was inserted into the third internode (disinfested with 70% ethanol) of plants at the tasseling stage (VT) (Nicoli et al. 2015). Plants of two maize hybrids, P30F53YH and DKB390PRO, were inoculated with CFMS 1342 isolate, in addition to six plants without the fungus (control). In the evaluation performed after 30 days, the plants inoculated with the fungus showed typical SR symptoms (Fig. 1). Morphological features of the fungus re-isolated were identical to those of *L. brasiliense* used for the inoculation, thus fulfilling Koch's postulates. The experiments were performed twice, and in both cases, the control plants did not exhibit disease symptoms. Although *L. brasiliense* has been reported to cause disease in some crops, mainly on fruit plants (Farr and Rossman 2018), to our knowledge, this is the first report of maize SR caused by *L. brasiliense* in Brazil and in the worldwide. According to IBGE (2018), since 2004 approximately 50% of the area planted with *Carica papaya* in the Brazilian locality of Luís Eduardo Magalhães has been replaced by maize. Since *C. papaya* is the main host of *L. brasiliense*, the present report is of great importance for future research on adaptive behaviour change of *L. brasiliense* to the new host, as well as in the development of adequate management strategies for controlling this pathogen in maize.

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Fig. 1 Maize stalk rot caused by *Lasioidiplodia brasiliense*. **a** and **b**, external disease symptom. **c** and **d**, Pathogenicity test in green house (control and inoculated stalks). **e**, Pycnidia. **f**, Colony in OMA. **g**, Paraphyse rounded at apex. **h**, Hyaline immature conidia and pigmented mature conidia of *L. brasiliense* (scale bars = 40 μ m)



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References

- Carbone I, Kohn LM (1999) A method for designing primer sets for speciation studies in filamentous ascomycetes. *Mycologia* 91:553–556. <https://doi.org/10.2307/3761358>
- Farr DF, Rossman AY (2018) Fungal Databases, Systematic Mycology and Microbiology Laboratory, ARS, USDA. <http://nt.ars-grin.gov/fungalatabases/> Accessed 07 March 2018
- IBGE (2018) Instituto Brasileiro de Geografia e Estatística. <https://cidades.ibge.gov.br/brasil/ba/luis-eduardo-magalhaes/pesquisa/15/11863> Accessed 07 March 2018
- Netto MSB, Assunção IP, Lima GSA, Marques MW, Lima WG, Monteiro JHA, Balbino VQ, Michereff SJ, Phillips AJL, Câmara MPS (2014) Species of *Lasioidiplodia* associated with papaya stem-end rot in Brazil. *Fungal Divers* 67:127–141. <https://doi.org/10.1007/s13225-014-0279-4>
- Nicoli A, Costa RV, Cota LV, Silva DD, Zambolim L, Lanza FE, Guimarães DP, Landau EC (2015) Validação de escala diagramática para quantificação da severidade da antracnose do colmo do milho. *Cienc Rural* 45:1720–1726. <https://doi.org/10.1590/0103-8478cr20141510>
- White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: PCR protocols: a guide to methods and applications. Ed. by Innis MA, Gelfand DH, Sninsky JJ, White TJ. San Diego: Academic Press. 315–322