First report of *Gymnosporangium clavipes* Cooke & Peck affecting *Crataegus mexicana* var. *Chapeado* and *C. gracilior* in Mexico

D. Alvarado-Rosales¹, E. H. Nieto-López¹*, D. Téliz-Ortiz¹, V. Ayala-Escobar¹, H. V. Silva-Rojas¹, R. Nieto-Angel², S. G. Leyva-Mir³, A. Jiménez-Nieto³ and C. Méndez-Inocencio⁴

¹Fitopatología, Instituto de Fitosanidad, Campus Montecillo, Colegio de Postgraduados. Km 36.5 Carretera México-Texcoco. Montecillo, Texcoco, 56230. Estado de México, México
²Departamento de Fitotecnia y Parasitología Agrícola, Universidad Autónoma Chapingo. Km 38.5 Carretera México-Texcoco. Chapingo, Texcoco, 56230. Estado de México, México
³CINVESTAV-IPN Unidad Querétaro. Libramiento Norponiente # 2000 Fraccionamiento Real de Juriquilla Santiago de Querétaro, 76230, Querétaro, México
⁴Departamento de Biotecnología, CIIDIR, IPN Unidad Michoacán, Justo Sierra # 28 Jiquilpan, 59510, Michoacán, México

*Corresponding author
Tel.: +52- 595-109-42-90
E-mail: edgar.nieto@colpos.mx

The tejocote (*Crataegus* spp.) is a tree considered to be native to Mexico. The aim of this study was to identify the causal agent of tejocote rust in the State of Puebla. Tejocote fruits were sampled in 2012 and 2013. The fungus was studied morphologically using light and scanning electron microscopy and molecularly using phylogenetic analysis of 18S and 28S rDNA genes. The fungus was identified as *Gymnosporangium clavipes* on tejocote fruits. To our knowledge, this is the first confirmed report of *Gymnosporangium clavipes* Cooke & Peck affecting *Crataegus mexicana* var. *Chapeado* and *C. gracilior* in Puebla Mexico.

**Keywords**: Aecia, Aeciospores, Peridium, Ornamentation, Fruits

Species of the genus, *Crataegus*, are commonly called “tejocote” in Mexico. This country is the largest producer of tejocote with 3,675 t in 2014 (SIAP 2015). *Gymnosporangium* species are heteroecious, producing the aecial phase on leaves and fruit of pome hosts and the telial phase on leaves, stems and branches of cypress (*Cupressus* spp.) and juniper (*Juniperus* spp.) (Sinclair and Lyon, 2005). In Mexico, there have been several species of *Gymnosporangium* reported, including *G. clavipes* in *Crataegus mexicana* and *Crataegus* sp. (León-Gallegos and Cummins, 1981). During May to September in 2012 and 2013, aecia fruits (Fig. 1A) were collected from *Crataegus mexicana* Moc. & Sessé var. *Chapeado* (SNICS TEJ-002-160709) and *C. gracilior* Phipps plantations in the Trans-Mexican Neovolcanic Belt in the State of Puebla. These samples were deposited as ENCB 121364 *Crataegus mexicana* var. *Chapeado* and *C. gracilior* as ENCB 121365 at the fungus collection of Escuela Nacional de Ciencias Biológicas at the Instituto Politécnico Nacional. Peridium and aeciospores measurements were taken using the software Images Plus 2.0 (Motic, Hong Kong, CH). The width and length of the aecia were measured using the Image Tool v 3.0 software (UTHSCSA, USA). Key (Kern, 1973) and the diagnostic protocol (EPPO, 2006) were used. These specimens were observed using a scanning electron microscope (SEM) XL 30 ESEM (Philips, A’dam, NED) with a range of 2.5-20 Kv. The ornamentation of the outer, side and inner walls of the peridial cells of aecia and the ornamentation of the aeciospores were observed based on the classification (Lee and Kakishima, 1999ab). Aecia were rosettoid and white in colour, measuring 7.59–15.25 × 0.54–0.79 μm, with little dehiscent peridium (Fig. 1B). On the peridial cell walls of aecia, outer (Fig. 1C) and side walls were smooth (Fig. 1D). However, the inner walls were tuberculate (Fig. 1D). Aeciospores were ellipsoid and orange in colour,
measuring 18.8–37.6 × 24.0–57.2 μm (Fig. 1E). The ornamentation of aeciospores presented small annulations (Fig. 1F). Based on morphological characteristics, the fungus was identified as Gymnosporangium clavipes (EPPO, 2006; Kern, 1973). For confirmation of morphology-based identification, the specimens (ENCB 121364 and 121365) were subjected to molecular analysis. Aecia samples amplified with the primers D1/D2 and Rust18S-R/NS1, resulted in the expected band of 700 bp and 1750 bp, respectively. The PCR products were purified and sequenced. The resulting sequence generating primer pair, D1/D2, was deposited to the GeneBank database as tejocote fruit aecia ENCB 121364 (GenBank accession No. KM382067) and analysed for similarity to known sequences in the database. The sequence showed high similarity to G. clavipes up to 99% (GenBank accession Nos. DQ354545 and HQ317528). In addition, the resulting sequence generating primer pair, Rust18S-R/NS1, the resulting sequence was deposited to the GeneBank database as tejocote fruit aecia ENCB 121365 (GenBank accession No. KM382070). The sequence also showed high similarity to G. clavipes up to 99% (GenBank accession No. DQ354546). In most reports of the species discussing about Gymnosporangium rust, the pathogenicity test was not carried out (Dervis et
al., 2010; Lee and Kakishima, 1999a, b; Yun et al., 2008; Yun et al., 2009). Although G. clavipes had been identified previously (León-Gallegos and Cummins, 1981) in C. mexicana and Crataegus sp., in this study, we have additionally employed SEM to identify infection on the specific tejocote species and varieties and molecular analysis. To our knowledge, this is the first confirmed report of Gymnosporangium clavipes affecting Crataegus mexicana var. Chapeado and C. gracilior in Puebla, Mexico.

Acknowledgments

The authors are grateful to Dr. Enrique Guizar Nolazco (Forestry and Ecology Division of Forestry Sciences, Universidad Autónoma Chapingo) for assisting in the identification of hosts and to Dr. Juan Manuel Tovar-Pedraza (Colegio de Postgraduados) for advising on the manuscript. This work was supported by Colegio de Postgraduados and SINAREFI-SNICS-SAGARPA (Secretaria de Agricultura, Ganaderia, Desarrollo Rural, Pesca y Alimentación).

References


SNICS. 2012. Catálogo nacional de variedades vegetales. SAGARPA, México.


