

**AN IN VITRO CO-CULTURE SYSTEM AND RAPID DIFFERENTIAL RESPONSES TO *FUSARIUM OXYSPORUM* F.SP. *VASINFECTUM* RACE 4 IN DOMESTICATED COTTON**

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**Abstract**

*Fusarium oxysporum* f. sp. *vasinfectum* Race 4 (FOV4) is a devastating fungal pathogen that affects both domesticated cottons, *Gossypium hirsutum* (Upland) and *Gossypium barbadense* (Pima). Greenhouse and field-based resistance assays can be unreliable due to uneven inoculum load, the presence of endophytes, and uncontrollable environmental factors. An in vitro co-culture method that uses a modified Murashige and Skoog medium was designed to support uniform growth of both domesticated cotton species and FOV4 in a scalable format using an inert polyphenolic foam resin to enable host-pathogen investigation in a controlled environment. Roots grown in the aerated foam matrix were morphologically more similar to roots grown in the field when compared to conventional agar-based in vitro systems. Histological analysis demonstrated better differentiated xylem in the aerated foam matrix when compared to agar. Inoculation and subsequent co-cultivation of field-verified resistant and susceptible genotypes with FOV4 resulted in an average of 90% diseased or dead tissue within four weeks of inoculation in the susceptible variety, while the highly tolerant checks showed less than 20% symptomatic tissue. FOV4 was isolated from the vascular system of every genotype within 72 hours of infection. This in vitro format offers a scalable system to rapidly screen cotton germplasm for tolerance to fungal pathogens and to facilitate the study of biotic interactions in both domesticated cotton species.