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Direct Detection of Coccidioides from Arizona Soils Using CocciENV PCR Assay.

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Introduction

Coccidioides immitis and *Coccidioides posadasii* are soil-dwelling fungi endemic to arid regions of the southwestern United States, particularly Arizona and California. These organisms are the causative agents of coccidioidomycosis, commonly known as Valley Fever, a respiratory disease that can range from mild flu-like symptoms to severe pulmonary and disseminated infections. Despite their public health significance, detecting *Coccidioides* in environmental samples has historically been challenging due to their sporadic distribution and the limitations of traditional culture-based methods [1, 2].

To address this gap, researchers developed the CocciENV real-time PCR assay, a highly sensitive and specific molecular tool designed to detect *Coccidioides* DNA directly from soil samples [3].

This assay represents a major advancement in environmental mycology and epidemiological surveillance, enabling large-scale screening of soil for fungal presence and improving our understanding of the ecology and distribution of *Coccidioides* [4, 5].

The CocciENV assay was adapted from CocciDx, a PCR assay originally validated for clinical diagnostics. Researchers at the Pathogen and Microbiome Institute at Northern Arizona University, led by Dr. Bridget Barker, modified the assay to work with complex environmental matrices such as soil. The team collected soil samples from known endemic areas in Arizona during fall 2013 and spring 2014, extracting DNA and applying the CocciENV assay to detect fungal presence [6, 7].

The assay targets specific genetic markers unique to *Coccidioides*, allowing for rapid and accurate identification. To validate the results, the team also employed next-generation amplicon sequencing targeting the ITS2 region, confirming the presence of *Coccidioides* DNA in multiple samples.The distribution of *Coccidioides* in soil is highly variable and cannot be explained by soil chemistry alone. Factors such as biotic interactions, climate, land use, and animal activity (especially rodent burrows) influence fungal presence. The CocciENV assay enables researchers to explore these associations by providing a scalable method for environmental surveillance [8, 9].

Understanding where *Coccidioides* thrives is critical for predicting outbreaks and guiding public health interventions. For example, construction and land development in endemic areas can disturb contaminated soil, releasing fungal spores into the air and increasing infection risk. The CocciENV assay can help identify high-risk zones and inform mitigation strategies [10].

Conclusion

Compared to traditional culture methods, which are slow and often yield false negatives, the CocciENV assay offers:Detects low levels of fungal DNA in complex soil samples. Targets unique genetic sequences of *Coccidioides*, reducing cross-reactivity.Provides results within hours, enabling rapid response.Suitable for large-scale environmental screening.These features make CocciENV a powerful tool for both research and

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public health surveillance.Valley Fever is underdiagnosed and often misidentified due to nonspecific symptoms. Environmental detection of *Coccidioides* can complement clinical diagnostics by identifying exposure risks and guiding awareness campaigns. For instance, the Arizona Department of Health Services has used environmental data to inform residents and healthcare providers about seasonal and geographic trends in Valley Fever incidence.Moreover, CocciENV can be used to monitor dust samples, construction sites, and agricultural lands, where fungal spores may become airborne. This proactive approach can help prevent outbreaks among vulnerable populations, including outdoor workers and immunocompromised individuals.Variability in soil composition can affect DNA extraction and assay performance.Fungal presence may fluctuate seasonally or due to environmental changes.Protocols must be harmonized across labs to ensure reproducibility.

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