

Project Number

J1730



Name(s)

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Project Title Equine White Line Disease

Abstract

Objectives/Goals I will collect and isolate samples from horses showing Equine White Line Disease symptoms then use DNA analysis identify Scedosporium ssp. as a causal microbe.

Methods/Materials

Nutrient agar, Petri dishes, inoculating loop, sample vials, hoof pick, cookie tray, box lid, lab coat, safety glasses, latex glove, hood, microscope slide, bunson burner, pipettes, water, cover slip, microscope and camera.

DNA Extraction:TE (Tris with EDTA),1.5ml tube,Microwave,Beaker,and Centrifuge. Polymerase Chain Reaction:Master Mix,Thermo-cycler,Freezer,Agarose gel,TE Running buffer,Electrophoresis gel box,Loading dye,UV lamp,Gel-documentation system,Thermo printer,Saccharomyces cerivisiae, and 1KB marker.

Results

I was able to collect samples from horses showing symptoms of Equine White Line Disease and a horse with no signs of Equine White Line Disease. Petri dish cultures that looked like they were growing fungus were samples taken from horses 1, 2, 3, and 4, the horses that had originally showed signs of Equine White Line Disease. The culture of horse 5 who did not show signs of Equine White Line Disease showed bacterial like growth. All the Petri dishes from horses 1, 2, and 3 showed signs of fungal growth in the micrographs. We chose samples from horses 1, 2, and 3 and extracted DNA from them using a microwave procedure and PCR. When we ran the 9 samples and 1 control sample of Brewers yeast electrophoresis. Four of the 9 gel lanes showed smeared bands of DNA. The Brewers yeast showed no results. Because our bands were smeared and indistinct I could not get DNA that we could sequence. A second analysis of the same samples using a different microwave technique showed smears in 9 of the 9 lanes and the Brewers yeast again showed no results. As in the first analysis the bands were smeared and indistinct so I was unable to get a clean sample of DNA to sequence.

Conclusions/Discussion

My results do not deny or support my hypothesis because I able to extract and find DNA in 9 of 10 lanes. But the results were indistinct and smeared so I didn't get a clean sample of DNA to sequence and identify a fungus. The smeared results could be a result of the microwave techniques or the PCR cycles. Since the Brewers Yeast control did not show any signs of DNA I believe the technique needs to be refined.

Summary Statement

DNA analysis will identify Scedosporium spp. as a causal agent of Equine White Line Disease

Help Received

Used lab equipment at University of California at Santa Cruz under the supervision of David Bernick.