Project Summary Rocky Mountains Cooperative Ecosystem Studies Unit

Project Title: Evaluation of Water and Soil Samples from Rocky Mountain National Park for Chronic Wasting Disease Prions

Discipline: Natural Resources Type of Project: Research Funding Agency: National Park Service Other Partners/Cooperators: Colorado State University Effective Dates: September 15, 2009- December 31, 2012 Funding Amount: \$54,050

Investigators and Agency Representative:

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Project Abstract: Chronic wasting disease (CWD) is a transmissible spongiform encephalopathy of deer and elk. Research has indicated that CWD is transmitted horizontally, and that both blood and saliva can transmit disease. Urine and feces from rodents have also been shown to contain prions. Environmental exposure to pens where infected animals have been kept has resulted in disease transmission to deer. However, evaluation of environmental components such as soil and water that may act as transmission vehicles for prions has been hampered by sensitivity limitations of conventional western blotting and inoculation limitations of bioassays. In this study we propose to utilize protein misfolding cyclic amplification (PMCA) to detect CWD prions ($\mathtt{Pr}\mathtt{P}^{\mathtt{CWD}}$) in environmental samples such as water and soil from the various areas within Rocky Mountain National Park (ROMO). PMCA is an *in vitro* technique that enables the amplification of minute amounts of PrP^{CWD} by rapidly converting normal prion protein (PrP^{c}) to more PrP^{CWD} . The laboratory of Dr. Mark Zabel at Colorado State University has used PMCA to detect minute amounts of PrP^{CWD} in water samples from the Poudre River at a time of high snowmelt (manuscript reviewed and under revision). This proposed environmental research will complement a current study in the Zabel laboratory investigating CWD prion distribution in various tissues from CWD positive elk culled from ROMO through current management actions. Understanding environmental CWD prion contamination will lead to a greater understanding of CWD transmission in nature.

The Zabel laboratory has a scientific permit (ROMO-2008-SCI-0073) to collect environmental samples, including surface water and topsoil from areas frequented by CWD-infected elk, as well as from exclosures that have had no CWD-infected elk exposure for decades. We plan to collect and evaluate samples for PrP^{CWD} starting in the fall of 2009. ROMO is an ideal site to evaluate environmental samples because of its long history of clinical CWD in elk and deer herds within the park as well as having animal exclosures which likely limit CWD contamination. Additionally, our laboratory has been in involved in the evaluation of tissues and body fluids taken from euthanized CWD-positive elk from the park as part of a CWD surveillance study in collaboration with Drs. Margaret Wild and Jenny Powers with the National Park Service.

Water and soil samples will be collected from the Moraine Park area and various exclosures within ROMO. Surface water samples will also be taken in those areas from sources such as ephemeral ponds and free-flowing streams. Approximately 1L of water will be collected from each surface water site. Soil samples will be taken by collection of surface soil free from obvious fecal material. No holes or depressions will be left after collections due to the superficial nature of the soil desired for this study. We will perform PMCA on replicate test, positive control and negative control samples.

This study will help determine the source of environmental CWD contamination and will help inform CWD management decisions and remediation strategies, such as culling of CWD-positive animals, environmental carcass removal and potential exclosure construction.

Outcomes with Completion Dates:

List of Products:1) Study plan to evaluate water and soil samples from Rocky Mountain National Park for CWD prions, 2) Field collection of water and soil samples at Rocky Mountain National Park, 3) Laboratory analyses of water and soil samples, as well as archived mule deer fecal samples, for CWD prions, 4) Report evaluating detection methods and results of testing water and soil samples, and 5) Manuscript detailing findings submitted to a peer-reviewed journal for publication.

Keywords: Chronic wasting disease (CWD), transmission, ungulates, Rocky Mountain National Park, Colorado State University