

# **Report for 2004ND46B: Effects of West Nile Virus Infection, Immune Function, and Age on Female Yellow-headed Blackbird (*Xanthocephalus xanthocephalus*) Reproduction.**

## Publications

- Other Publications:
  - Newbrey, J.L., and W.L. Reed. 2005. Effects of nest contents and minimum daily temperature on female Yellow-headed Blackbird nest attentiveness. Presented in Wilson Ornithological Society and Association of Field Ornithologists Joint Meeting.
  - Newbrey, J.L., and W.L. Reed. 2004. West Nile virus antibodies in central North Dakota icterids. Presented in Cooper Ornithological Society 74 th Annual Meeting.
  - Newbrey, J.L. 2004. West Nile virus antibodies in central North Dakota icterids: Implications for ecology and management. Poster presentation in North Dakota Chapter of the Wildlife Society Annual Meeting.

## Report Follows

**EFFECTS OF WEST NILE VIRUS INFECTION, IMMUNE FUNCTION, AND  
AGE ON FEMALE YELLOW-HEADED BLACKBIRD  
(*XANTHOCEPHALUS XANTHOCEPHALUS*) REPRODUCTION**

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**DESCRIPTION OF THE CRITICAL WATER PROBLEM**

Recent high water levels and canalization of water resources (i.e., Garrison Diversion) in North Dakota have resulted in an increase of aquatic habitats for many wildlife species. Current water conditions correspond with increased numbers of wetland breeding birds and increased habitat for breeding mosquitoes. Because birds often serve as intermediate hosts for mosquito borne diseases, increased populations of birds and mosquitoes could impact the ecology, rate of emergence, and persistence of diseases in humans and wildlife. The recent spread of WNV into the state has produced a need for research to study the influence of the virus on wetland wildlife in North Dakota. The North Dakota Department of Health reported the first cases of WNV in the state in the summer of 2002. The first bird to test positive for WNV was an American crow (*Corvus brachyrhynchos*) found on July 14 th , and the first positive human cases were reported on August 28 th. There were 19 human cases of WNV and 2 deaths in the state in 2002. In 2003, the virus was much more widespread, with 422 human cases and 4 deaths. This year alone, 788 bird carcasses have been tested in the state and 189 were WNV positive.

Because stagnant water in wetlands is ideal breeding habitat for mosquitoes, wildlife associated with these habitats may suffer high rates of WNV infection. The recent arrival of WNV into the state necessitates a study of the prevalence and immunological impact of WNV on native North Dakota wetland species. Most research on the virus has focused on using carcasses of birds as a surveillance system for detecting the spread of WNV across North America. No published research has been conducted on a living population of free-ranging birds. Failure of biologists to adequately address disease emergence in free-ranging wildlife may lead to diminished geographic distributions and populations declines.

The Missouri Coteau of central North Dakota has many small prairie wetlands, which provide essential foraging and breeding habitat for many species of birds. Yellow-headed blackbirds are an ideal species to study WNV infection because they breed in high-density wetland colonies throughout the Coteau. Establishing rates of WNV infection in yellow-headed blackbirds is necessary to determine the vulnerability of this wetland dwelling species and the influence of WNV infection on reproduction. Information gathered on WNV for this study can also be used to model and predict potential impacts of the virus on other species of wetland birds.

**SCOPE AND OBJECTIVES**

The overall objective of this project is to determine the effects of female age and infection with West Nile virus on yellow-headed blackbird (*Xanthocephalus*

*xanthocephalus*) maternal investment into eggs. The specific objectives of this project are to identify the prevalence of WNV in a free-living population of yellow-headed blackbirds, to quantify variation in immune function of female blackbirds, and to measure the relationship between female immune function and age on carotenoid allocation to eggs. These objectives will allow us to evaluate potential relations between wetland bird WNV infection and increased aquatic habitat for breeding mosquitoes in North Dakota.

## METHODS

Female yellow-headed blackbirds were captured using nest traps to collect blood for WNV antibody detection and immunity quantification. Blood serum was tested for WNV antibodies using competitive enzyme-linked immunoabsorbent assay (ELISA) specifically designed to detect WNV antibodies in blood serum from avian species. To assess variation in immune function among females, I created blood smears and obtain ratios of immune system cells (heterophils to lymphocytes). Prior to release, each female was banded with a standard Fish and Wildlife Service aluminum band along with a unique color-band combination for individual field identification. I located and monitor nests of each banded female yellow-headed blackbird to determine nest success and nestling performance and how differences in carotenoid levels influences reproduction. Male yellow-headed blackbirds and other members of the Family Icteridae (red-winged blackbirds, common grackles, etc.) were also captured using live funnel traps for WNV antibody detection.

This study was conducted on several wetlands located within a five square mile area of the prairie coteau region of central North Dakota (Stutsman County). Central North Dakota has one of the highest concentrations of breeding yellow-headed blackbirds in North America. In addition, Stutsman County has one of the highest numbers of positive avian West Nile virus cases in central North Dakota.

## KEY RESULTS

- In 2003, third-laid eggs were collected from 51 female yellow-headed blackbirds on 7 different wetlands. Twenty female yellow-headed blackbirds and blood and feather samples were collected for WNV antibody detection and carotenoid analysis. Also collected were blood samples from 9 male yellow-headed blackbirds, 19 grackles, 2 red-winged blackbirds, and 1 western meadowlark to use for WNV detection. The blood samples were tested for WNV antibodies. Antibodies were detected in two individuals, one red-winged blackbird and one western meadowlark.
- In 2004, blood samples were collected from 91 yellow-headed blackbirds, 24 common grackles, three house sparrows, two brown-headed cowbirds, and one red-winged blackbird. All of the serum samples were tested for WNV antibodies and five positives were found for WNV antibodies, two female yellow-headed blackbirds, two house sparrows, and one common grackle. Also collected were 25,000 mosquitoes and a subset of the mosquitoes to the species level was

identified. *Culex tarsalis*, a species known to transmit the virus in North Dakota, were present in all of my collections, but at low levels (5%). The mosquitoes were tested for WNV RNA using polymerase chain reaction, but all samples were negative for the virus.

- In 2005, eggs, feathers, and blood samples were collected from 69 female yellow-headed blackbirds. Also monitored were nest success of all study nests. Daily growth rates of 133 chicks from 55 nests were measured. In the lab, white blood cell ratios for each female to quantify differences in female immune function were obtained.
- Research is being continued beyond the Fellowship funding period.

### **SIGNIFICANCE OF RESEARCH**

This study will provide essential information on the prevalence of WNV in a North American avian species. Infection with the virus can be lethal; however, the degree to which birds are adversely affected varies among species and even between individuals within a species. By testing for the presence of antibodies to WNV in yellow-headed blackbirds, I will be able to assess the vulnerability and degree of virus exposure in a free-living population of wetland dwelling birds. Many wildlife pathogens cause non-lethal physiological and reproductive effects that remain poorly understood. Because female birds allocate essential resources to eggs, exposure to pathogens can shift maternal resources away from reproduction. This seemingly small, non-lethal effect influences the survival of offspring and can therefore cause population level effects in the next generation.