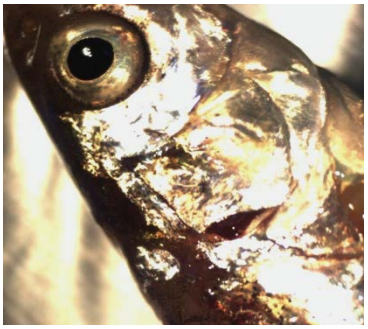
U.S. Fish & Wildlife Service

Rio Grande Silvery Minnow Health Study



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Cover photography (beginning top left): 1) "Examination of fish health"; 2) "Rio Grande silvery minnow with shortened opercula"; 3) "Deployment of water quality monitoring equipment in the Rio Grande"; and 4) "Examination of fish for parasites" by U. S. Fish and Wildlife Service/J. Lusk (photographs 1, 3, and 4) and A. Davis (photograph 2).

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Example appendix or chapter citation: Papoulias, D. M., M. L. Annis, A. P. Davis, J. D. Lusk, E. Zabotkina, E. Nazarova, and D. E. Tillitt. 2009. Health and Histopathological Evaluation of Rio Grande Silvery Minnow from the Rio Grande, New Mexico. U.S. Geological Survey, Columbia Environmental Research Center, Columbia, Missouri.

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PROJECT SUMMARY

It is widely known that the health of fish can be adversely affected by exposure to water pollution. In 2005, the Middle Rio Grande Endangered Species Act Collaborative Program (Collaborative Program) requested proposals for monitoring the health conditions of Rio Grande silvery minnow (*Hybognathus amarus*; RGSM) in the mainstem of the Middle Rio Grande. In response, the U.S. Fish and Wildlife Service (Service) designed the RGSM Health Study to monitor and characterize the health conditions of RGSM in the MRG to provide baseline data on diseases and parasites, and document external and internal anomalies and pathologies in fish collected throughout the year. The Collaborative Program also requested that monitoring of RGSM health conditions be conducted in conjunction with a water-quality monitoring program to better link results to possible causes. In particular, the New Mexico Environment Department (2009) implemented (and reported separately) a companion water quality study that overlapped some of the same sites sampled during this RGSM Health Study.

Various agencies participated in the RGSM Health Study including the New Mexico Environment Department (NMED); the University of New Mexico, Museum of Southwestern Biology (Museum); the U.S. Geological Survey (USGS), New Mexico Ecological Services Field Office, and Dexter Fish Health Unit. We conducted the RGSM Health Study from July 2006 to July 2008 (Year 1 and Year 2). Rio Grande silvery minnows (*Hybognathus amarus*) were collected nearly every three months from six sites in the Rio Grande from Bernalillo to San Antonio, New Mexico. We used a suite of standard methods of observation of fish health conditions during necropsy to determine total length, mass, organ condition, the number and type of lesions and anomalies, the number and type of parasites and pathogens, and we analyzed their tissues for a variety of chemicals and genetic conditions. We also measured surface water quality parameters (i.e., dissolved oxygen (DO), oxygen saturation, temperature, pH, specific conductivity).

There are three chapters of this RGSM Health Study Report with three additional reports included as appendices, which describe the different activities undertaken during this RGSM Health Study. Chapter 1 includes introductory materials, selection of study sites, methods of fish and water quality data collection, results, and discussion including the use of oxygen measures to evaluate gross primary productivity (GPP) and ecosystem respiration (ER). Chapter 2 includes the Fish Health Assessment methods, necropsy results, and discussion. Chapter 3 includes the analytical methods, results, and discussion of chemicals in RGSM tissues. Literature cited and data appendices (Appendices 1A-1C, 2A-2K, and 3A-3F) are provided with the first three chapters. Some materials were made available online at http://www.fws.gov/southwest/es/newmexico/EC_reports.cfm. Appendix 4 includes the Fish Histology and Histopathology Report by Papoulias et al. (2009). Appendix 5 includes the Fish Pathogen Assessment Report that included methods of RGSM pathogen data collection, results, and discussion by Woodland (2012). Appendix 6

includes the Genetic Evaluation Report by Osborne and Turner (2009). Each chapter or appendix report also refers to their own figures, tables, and data appendices.
We measured individual fish and dissected them to observe their external and internal conditions (Chapter 2). We measured total length and weight and found fish collected in Year 1 were significantly larger than those collected in Year 2 were. We found a significant relationship between fish length and weight with average annual water temperature. We observed shortened opercula, gill pathologies, liver anomalies, and degenerated kidney tubules as anomalies with the greatest frequency and these anomalies were observed at a greater frequency in Year 2 than in Year 1. Changes in water temperature, nutrition, or discharge appeared to be the most likely factors affecting skeletal development and therefore shortened opercula in silvery minnows. Mean Feeding Index was significantly lower in Year 1 than in Year 2, suggesting that food was more readily available during Year 1. The percent of fish with anomalies or pathological lesions tended to increase from upstream to downstream. Generally, the health of the Rio Grande silvery minnows was moderately affected, indicative of chronic stress, deteriorated from Year 1 to Year 2, from upstream to downstream, and during the summer (as measured during July) (Chapter 2, Appendix 4, and Appendix 5).

Additionally, we collected samples of spleen, gonad, and other tissues for histopathological examination and hematology (Appendix 4). All silvery minnows observed were experiencing chronic stress likely associated with poor water or sediment quality as evidenced by a suite of health indicators. Those silvery minnows collected in July tended to be the least healthy, having more macrophage aggregates, granulomas, lower liver glycogen and increased spleen size with fewer red blood cells. Gill abnormalities (e.g., discoloration, fraying, clubbing, fused lamellae, embolism, and parasites) were commonly observed in over 96 percent of the silvery minnow gills and could be related to bacterial or parasite infestations, or elevated copper.

Female gonads were observed in 58 percent of samples, but percentage of females was not significantly different by sample year, site, or month. However, there was a significant trend of more females in samples collected sites downstream than upstream. We observed some atretic oocytes in females (i.e., ectopic oocytes in liver and kidney tissues); however, no oocytes were positively detected in the testes of males evaluated.

The majorities of livers were low to moderate in glycogen content, and were lowest in silvery minnows collected in July. While gross liver abnormalities were high (e.g., discolored or mushy liver), liver pathologies (e.g., edema, high fat content, pigments and macrophages, granulomas, parasites, etc.) were low. Granulomas and macrophage aggregates were more prevalent in July 2008 samples and hemosiderin and melanin pigments were more common in livers collected in Year 2.

After necropsy, the silvery minnow carcasses were chemically analyzed for a variety of metals, hydrocarbons, organochlorine compounds, stable isotopes, and fire

retardant residues (Chapter 3). Generally, organic chemical concentrations in silvery minnows were low. However, semivolatile hydrocarbons, especially naphthalene and other PAHs, were moderately elevated in silvery minnows. Several organochlorine pesticides, PCBs, and BDEs (flame-retardants), were also elevated in silvery minnows collected from Site 3 and Site 4. The bioaccumulation of chlorinated and brominated organic chemicals at these sites may have been related to silvery minnow size, lipid content, or potential site or dietary differences. Concentrations of inorganic contaminants in Rio Grande silvery minnow were similar to those in other fish species collected from the Rio Grande and were generally low, with the exception of aluminum, copper, nickel, vanadium, and zinc. Additional research was recommended for those contaminants in fish that were elevated.

We measured water quality parameters (e.g., dissolved oxygen, temperature, pH, and conductance) as well as used rates of oxygen change to estimate gross primary productivity and ecosystem respiration (Chapter 1). Average gross primary productivity rates ranged widely, but were generally higher at upstream sites and during summer. Low oxygen conditions were identified and were most likely associated with oxygen demanding substances in sediment. Increasing gross primary productivity and thereby increasing oxygen content will reduce oxygen demand and ecosystem respiration. Improvements in these conditions would likely result in increased populations of fish, increased species richness of benthic invertebrates, and decreased chronic stress reported in Rio Grande silvery minnows.

In a companion study, the New Mexico Environment Department (NMED 2009) evaluated the chemical quality of water, sediment and fish in the Middle Rio Grande. NMED evaluated chemical data in the water column to evaluate exceedances of New Mexico Water Quality Control Commission-approved water quality standards. The NMED (2009) concluded that very few water quality standards impairments were found. The NMED (2009) also evaluated water, sediment and fish tissue data to identify additional water quality issues of concern (i.e., issues that were not considered to exceed New Mexico water quality standards) and identified aluminum, copper, chromium, ammonia, low oxygen, elevated pH in water, zinc and DDT in fish tissues, and several semivolatile compounds (PAHs) in sediment as elevated. The Rio Grande Silvery Minnow Health Study findings were consistent with the NMED (2009) study.

In summary, the health of the Rio Grande silvery minnow was moderately impaired. The silvery minnows had skeletal deformities (i.e., shortened opercula) that suggested a causal association with factors of water temperature, nutrition, and/or discharge. Silvery minnows were generally healthier collected from upstream than from downstream sites. When water quality conditions deteriorated, including higher temperatures, lower oxygen content, and with increased pathogen burdens as were measured during July, the health of silvery minnows also generally declined. Several contaminants were prevalent in fish including hydrocarbons such as PAHs, and certain metals, including aluminum and zinc.