Whirling Disease Investigations

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Federal Aid in Fish and Wildlife Restoration

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Job No. 1: *Myxobolus cerebralis* in Colorado's Cutthroat Trout Populations

Project Objective: To determine, and then document through professional publication, the impacts of the myxosporean parasite *Myxobolus cerebralis (M. cerebralis)* on wild trout populations in selected stream ecosystems in Colorado with an overarching objective of developing risk assessment guidelines for the management of whirling disease.

Period Covered: July 1, 2007 through June 30, 2008

Principal Investigator: R. Barry Nehring

Job Objective: Determine the extent of occurrence and severity of impact of *Myxobolus cerebralis* on populations of greenback *Oncorhynchus clarki stomias,* Rio Grande *O. c. virginalis*, and Colorado River cutthroat trout *O. c. pleuriticus* throughout Colorado.

INTRODUCTION

"Whirling disease" (WD) is a debilitating malady of trout and salmon that was first observed in cultured rainbow trout in Germany in 1893 (Hofer 1903). The name comes from the abnormal swimming behavior of fry or fingerling salmonid fishes that can occur after becoming infected by the myxosporean parasite *Myxobolus cerebralis (M. cerebralis or Mc).* When frightened the fish appear to be chasing their tail. It was considered a serious problem for the aquaculture industry for much of the 20th century (Plehn 1905, 1924; Schäperclaus 1931; Uspenskaya 1957, 1982). The parasite life cycle remained an enigma until it was described in the early 1980s (Markiw and Wolf 1983; Wolf and Markiw 1984). The complex two-host life cycle alternates between a tubificid worm (*Tubifex tubifex*) and a salmonid fish. The parasite produces spores in each host that are infective to the alternate host. Myxospores produced in infected salmonids can be ingested by bottom-dwelling oligochaetes. Susceptible forms of *T. tubifex* that become infected produce a triactinomyxon (TAM) actinospore that is semi-bouyant, tumbles in moving water, and is infectious to susceptible salmonids.

Myxobolus cerebralis (Mc) was first detected in two public and two private trout rearing facilities in Colorado, late 1987 (Walker and Nehring 1995). Population level impacts among wild salmonid populations were unknown until the 1990s. However, severe losses of young rainbow trout first observed in major reaches of the upper Colorado, Cache la Poudre, Gunnison, Rio Grande, and South Platte rivers in Colorado in 1993 and 1994 were ultimately attributed to WD (Walker and Nehring 1995; Nehring and Walker 1996; Nehring et al. 1998; Nehring and Thompson 2001). The parasite became widely distributed in Colorado in the early 1990s through the stocking of millions of catchable size trout reared in waters enzootic for *M. cerebralis* (Schisler 2001). More than one million trout from *Mc*-infected hatcheries and rearing units were stocked into lakes, reservoirs and streams in the Cache la Poudre River drainage between 1990 and 2001. Moreover, this was not a highly unique scenario. Given such a management strategy, it is not surprising that *M. cerebralis* had been detected in feral salmonids at 118 different

locations in lakes, reservoirs and major stream segments in Colorado by October 1997 and at 208 sites by spring 2000. It is estimated that *Mc* infections have negatively impacted recruitment of wild rainbow and brook trout fry in 560 – 600 km (350-400 miles) of streams in Colorado (Nehring and Thompson 2001). A recent special technical report, **Colorado's Cold Water Fisheries: Whirling Disease Case Histories and Insights for Risk Management,** summarizes the effects of exposure to *M. cerebralis* upon Colorado's salmonid fisheries (Nehring 2006).

Debilitating effects of the parasite were documented on wild rainbow trout in major reaches of the Madison River in Montana in the 1990s (Vincent 1996a,b). Research efforts between 1994 and 2004 revealed the parasite was enzootic in many coldwater habitats in Colorado (Nehring and Thompson 2003) and western Montana (Baldwin et al. 1998). It has been detected at one or more locations in almost all states west of the 100th meridian in the continental U.S. (Bartholomew and Reno 2002). Detected in Yellowstone cutthroat trout (*O. clarki bouvieri*) in 1998, *M. cerebralis* infections have had serious impacts on spawning runs in the Yellowstone River immediately downstream of Yellowstone Lake and in Pelican Creek and Clear Creek, major spawning tributaries that drain into the northeastern corner of the lake (Koel et al. 2005, Koel et al. 2006). Recent studies suggest that *M. cerebralis* may be enzootic in one or more streams in south central Alaska near Anchorage (Arsan 2006).

Widely distributed in the mountainous regions of Colorado, *M. cerebralis* has been detected in feral salmonid populations in close proximity to areas designated as cutthroat trout recovery streams. In 2003, at the initiation of this study there were no known cases where the parasite had negatively impacted fry recruitment for any of Colorado's three sub-species of cutthroat trout. At that time, the parasite was enzootic among Colorado River cutthroat trout in Trappers Lake in western Colorado and in greenback cutthroat trout in Zimmerman Lake in north central Colorado. Both trout populations were managed for spawn-taking operations. Field exposure of young-of-the-year (YOY) of all three sub-species of Colorado's cutthroat trout to ambient levels of *M. cerebralis* in the Colorado River in the 1990s clearly demonstrated these fish are particularly vulnerable to developing a lethal infection after exposure (Thompson et al. (1999).

Once *M. cerebralis* becomes enzootic in an aquatic ecosystem, total year class failure can occur among susceptible species of salmonids under the proper suite of environmental conditions. And it is being shown in this study and others that the proper suite of environmental conditions is not very restrictive and does not necessarily involve environmental degradation.

The lack of a systematic effort to evaluate the distribution, establishment and spread of *M. cerebralis* into Colorado's aquatic ecosystems capable of supporting native cutthroat trout was the primary impetus for the initiation of this research project.

STUDY DESIGN

The primary study objective is to determine whether or not the parasite has spread into habitats capable of supporting cutthroat trout populations. A multi-faceted approach is being used to determine whether or not significant exposure and spread of *M. cerebralis* has already occurred. In the event that there has been only minimal establishment in most regions of the state, an effort is being made to determine whether introduction actually took place or not. In the event that introduction and exposure actually took place but the parasite was unable to establish itself, the objective will be to determine what factor(s) might have prevented establishment of the life cycle. In those areas where the parasite is not enzootic and there is no record of initial exposure, the protocol will be to collect aquatic oligochaetes and genetically test them to determine whether or not they are the lineage of *T. tubifex* that is highly susceptible to infection by the parasite. A statewide systematic sampling process should provide significant insight(s) into the mechanisms and factors that facilitate the spread of *M. cerebralis*. The study should also provide significant insights into the potential risk for spread of the parasite through the development of statewide distribution maps for all lineages of *T. tubifex.* This is most important for the lineage III strain that produces very high numbers of TAMs even from a low dose of *M. cerebralis* myxospores.

For the first level of assessment, in most cases trout population estimations are conducted on one or more segments of each study stream that are at least 91 meters (300 feet) long. When possible, two population estimates are conducted, one in the headwaters and another near the downstream end of the drainage. In general, the two-pass removal estimator is used to estimate population size and determine relative density, size and approximate age structure for all species of trout in the study reach (Seber and LeCren 1967). Study reaches are selected to include fry (YOY) and juvenile habitats in the population estimation process.

After the sampling process is completed, ten young-of-the-year (YOY) trout and ten age 1 juvenile trout are sacrificed to test for the *M. cerebralis* parasite. The YOY trout are tested by polymerase chain reaction (PCR) for genomic DNA unique to *M. cerebralis* (Cavender et al. 2004). Yearling juvenile trout are tested for myxospores of the *Mc* parasite using the pepsin-trypsin digest (PTD) method (Markiw and Wolf 1974). In most instances, cutthroat trout are euthanized for disease testing only when they occur in allopatry. When cutthroat trout are sympatric with other salmonids, these other fish are sacrificed for disease screening to avoid unnecessary depletion of the cutthroat trout population.

In the event that the study reveals there is little evidence of spread, there are several plausible explanations for such an eventuality. First, in many instances the particular habitat being studied may have never been exposed to the parasite. Second, the habitat in question may have been exposed, but the parasite never completed its life cycle. If the parasite did not become established there could be at least two plausible reasons. First, there may be very little stream habitat suitable for development of colonies of *T. tubifex* of sufficient density to sustain the life cycle in the aquatic oligochaete host. Second, aquatic oligochaetes may be present in the drainage but not the right species or proper lineage of *T. tubifex* that is susceptible to *M. cerebralis.* Recent studies have shown that among the four different lineages of *T. tubifex* (I, III, V and VI) currently known to exist in Colorado, lineages I,V and VI are not susceptible to

infection by *M. cerebralis* (Beauchamp et al. 2001, 2002; Baxa and Hedrick 2008). Kerans et al. 2004 found that other tubificid oligochaetes such as *Limnodrilus hoffmeisteri and Ilyodrilus templetoni* do not become infected when exposed to *M. cerebralis* myxospores in a laboratory setting. Research efforts in New Mexico demonstrated that only lineage III *T. tubifex* become infected when exposed to *M. cerebralis* (DuBey and Caldwell 2004; DuBey et al. 2005). In the New Mexico studies lineages I, III and VI *T. tubifex* were tested.

Substantial effort is being expended to collect substrate samples containing aquatic oligochaetes in as many habitats as possible to determine which possibility might be the most plausible explanation. The collections are made concurrent with the trout population estimation surveys. The samples are sorted to determine the relative abundance of "haired" and "non-haired" oligochaetes. The standard protocol is to separate and sort oligochaetes until two sub-samples of 50 "haired" worms per collection site have been identified and preserved in 70% reagent grade ethanol for quantitative PCR testing (hereafter qPCR) to determine which lineages of *T. tubifex* (if any) are present in the study reach. Recent advances in testing and development of DNA-based genetic markers specific to at least six different lineages of *T. tubifex* make this possible (Beauchamp et al. 2001, 2002). Each 50-worm composite sample is quantitatively screened for DNA markers specific to each lineage within the mitochondrial DNA 16S *T. tubifex* oligochaete group. A private laboratory (Pisces Molecular) developed a four-probe-multiplex qPCR test that allows the screening of a sample of up to 50 aquatic oligochaetes for the percentage of DNA for each of the four lineages of *T. tubifex* contained in the sample. The test can also provide a relative indication of the total amount of DNA from *T. tubifex* in the sample. This testing procedure will facilitate development of spatial distribution maps for the various lineages of *T. tubifex* by drainage basin and on a statewide basis.

In addition, each worm sample can be screened by qPCR using the Hsp70 (Heat shock protein gene 70) test (Cavender et al. 2004) to determine if DNA of *M. cerebralis* is present in the worm sample. The Hsp70 test targets a highly conserved region of the heat shock protein gene 70 that is found in a wide array of living organisms and also occurs in the genome for *M. cerebralis.*

METHODS

Trout Population Assessment - In most study streams, the objective was to estimate the salmonid species composition, density and size structure of the trout population at two or more sampling sites using the two-pass removal estimator as described by Seber and Le Cren (1967). Data collected during this effort were run through the Colorado Division of Wildlife's GOLDMEDL or JAKOMATIC computer software programs to develop the population estimates (N), 95% confidence limits, density (n/ha), biomass (kg/ha) and develop a relative estimate of year class abundance for the first three-year classes based primarily on length-frequency distribution. All sampling sites were identified by GPS to facilitate mapping the collection locations using the mapping software package ARC VIEW 9.

Parasite Screening in Fish – In streams where adequate numbers of salmonids were present, we collected ten YOY and ten juvenile (≥age 1) trout for screening for *M. cerebralis* infection. Juvenile trout were tested for *M. cerebralis* using the PTD methodology (Markiw and

Wolf 1974) while YOY trout were screened for parasite DNA using the Hsp 70 test (Cavender et al. 2004). In streams where cutthroat trout were sympatric with other salmonids, the nonnative salmonids were sacrificed for disease testing. Only when the population was allopatric were cutthroat trout taken for testing.

Aquatic Oligochaete Studies –Sediment-laden microhabitats from multiple locations within a study reach on each study stream were sampled for aquatic oligochaetes. All samples were thoroughly screened for aquatic oligochaetes. Oligochaetes were examined by stereo-zoom microscopy, separated into haired and non-haired forms and preserved in 70% reagent grade ethanol and distilled water for qPCR testing. Haired oligochaetes have a high probability of being *T. tubifex* (Kathman and Brinkhurst 1998). Our protocol was to preserve at least one sample of 50 haired oligochaetes from each collection to determine whether or not the sample contained DNA specific to any of the four lineages (I, III, V or VI) of *T. tubifex.* Each sample was prepped for total DNA extraction to preserve all of the genetic material in the sample. When large numbers of worms were encountered, two aliquots of 50 haired worms were preserved for qPCR testing. First, each sample was screened using a four-probe-multiplex qPCR technique to determine the relative percentage of DNA derived from four different lineages of *T. tubifex* in each 50-worm aliquot. Total DNA extraction preserves any *M. cerebralis* DNA in the sample, affording subsequent testing of the worms for *M. cerebralis* infection (Cavender et al. 2004).

Development and testing of the four-probe multiplex qPCR protocol that allowed simultaneous testing and quantification of mitochondrial DNA from four different lineages of *T. tubifex* in a single sample (Beauchamp et al. 2002) was completed in 2003 (John Wood, Pisces Molecular; personal communication). A large amount of testing was done to develop and refine the four-probe multiplex qPCR test for quantifying the relative amount of DNA for the various lineages (I, III, V and VI) of *T. tubifex* in aquatic oligochaetes. Large numbers of worms were tested individually as well as in pooled aliquots of 5, 10, 25, 50 and 100 worms. These tests afforded 1) the development of standards for calibration of the test for the four lineages of worms, and 2) demonstrated the optimum number of worms in an individual aliquot to produce the most reliable (accuracy and precision) and repeatable results. The 50 worm aliquot provided the best results across a broad range of worm sizes while concurrently minimizing the reagent costs at the laboratory. Results of those efforts are summarized in Tables 10a, 10b, 10c and 10d in Nehring 2006 and will not be reiterated here.

RESULTS and DISCUSSION

Historically, nine major river basins in Colorado have supported native cutthroat populations. These include the Arkansas, Colorado, Dolores, Gunnison, Rio Grande, San Juan, South Platte, White and Yampa river systems. Greenback cutthroat trout are native to the Arkansas and South Platte river basins. Rio Grande cutthroat trout are native to the Rio Grande basin. Colorado River cutthroat trout are native to the Colorado, Dolores, Gunnison, San Juan and White and Yampa river systems. No cutthroat trout were ever native to the North Platte drainage in Colorado. An overview of the number of streams and sites sampled each year for each of the three sub-species of native cutthroat trout is summarized below:

Year	Greenback		Rio Grande		Colorado River	
	streams	sites	Streams	Sites	streams	sites
2003	9	12	9	13	22	29
2004	3	5	18	26	24	36
2005	9	12	18	24	10	12
2006	---	---	10	18	49	73
2007	1	2	3	5	48	61
Total	**22**	**31**	**58**	**86**	**153**	**201**

When this project was initiated in 2003, there were 46 bodies of water (35 streams, eight lakes and three reservoirs) listed as habitats that were thought to support greenback cutthroat trout populations or could in the future. All occurred in the Arkansas River and South Platte River basins. Over the past five field seasons electrofishing operations have been conducted on 22 streams at 31 sites. For the most part, these streams were either classified as greenback cutthroat trout recovery streams or have direct connectivity to waters thought to support this sub-species. Detailed information regarding streams and sites sampled from 2003 through 2005 can be seen in Nehring 2004, 2005, 2006. No electrofishing operations were conducted on streams supporting greenback cutthroat trout during the 2006 field season.

In 2003 there were 82 bodies of water listed as habitats that either supported Rio Grande cutthroat trout populations or could in the future. The vast majority of them were either creeks or rivers; however, the list included one reservoir and ten lakes. All streams feed into the Rio Grande drainage that flows through the San Luis Valley in south central Colorado. Over the past five field seasons electrofishing operations have been conducted on 58 streams at 86 sites. These streams were either classified as Rio Grande cutthroat trout recovery streams or had direct connectivity to waters thought to support this sub-species. Details regarding streams and sites sampled from 2003 through 2006 can be seen in Nehring 2004, 2005, 2006, 2007.

Historically, Colorado River cutthroat trout occurred in the majority of coldwater streams west of the Continental Divide in Colorado. In 2003, the number streams, lakes and reservoirs listed as present or future Colorado River cutthroat trout recovery areas by major drainage basin were as follows:

Major Drainage Basin	**Streams**	**Lakes**	**Reservoirs**
Colorado River	74	7	1
Dolores River	5	0	0
Gunnison River	10	0	2
San Juan River	11	0	0
White River	6	1	0
Yampa River	17	1	0
Totals	**123**	**9**	**3**

Over the past five years, we have conducted fish population estimates and/or collected trout for disease testing for evidence of *M. cerebralis* infection from 153 streams and 201 sites in the greater Colorado River drainage in western Colorado. During the 2007 field season we collected fish and/or aquatic oligochaetes from 48 streams and lakes from 61 separate locations that were considered to have potential as cutthroat trout recovery streams, or were in proximity of or connected to streams containing Colorado River cutthroat trout. Sites sampled each year between June 2003 and May 2008 are shown on Map 1 in the Appendix. Test results for presence or absence of *M. cerebralis* in fish collected between 2003 and 2007 are shown on Map 2 in the Appendix. Most of the sampling effort for fish and worm collections during the 2007/2008 segment was concentrated in the Blue, Eagle, Gunnison, San Juan, White and Yampa River basins. In addition, an intensive effort was expended collecting additional worm samples at numerous sampling sites on the mainstem(s) of the Arkansas, Colorado, Dolores, Eagle, Gunnison, Rio Grande and White river basins. Maps 3 through 7 in the Appendix show the areas of the state where DNA specific for the various lineages of *T. tubifex* was detected in Colorado during the five-year study.

Trout Population Assessment and Parasite Screening -Trout population estimates and summaries of electrofishing surveys as well as PCR and PTD test results for evidence of *M. cerebralis* infection for all sampling sites for 2007 are organized by sub-species and presented in Tables 1 through 10. Odd-number tables contain bio-statistical estimates for population size, 95% confidence limits, density (N/ha) and biomass (kg/ha) for each stream sampled. Even-number tables summarize the PCR and PTD test results for fish collected and tested for evidence of *M. cerebralis* infection.

Greenback Cutthroat Trout – Tables 1 and 2 contain the data summaries for trout population bio-statistics and disease sampling for two sites on South Cottonwood Creek upstream of Cottonwood Lake west of Buena Vista, Colorado. These two sites were originally sampled in August 2000 (see Table 2.08 in Nehring 2003 for comparative data). The 2007 results largely mirror the results from the sampling effort in 2000, i.e., the trout at the upstream site (approximately 10 km above Cottonwood Lake) remain free of infection, while those collected at the downstream site nearer Cottonwood Lake were heavily infected by *M. cerebralis.* These data suggest that the upstream spread of the parasite through migration of infected fish, vectoring by avian or mammalian piscine predators, or as a "hitch-hiker" on angling equipment can be quite slow in some instances. The beaver ponds on South Cottonwood Creek upstream of the lake are the most probable factor that has slowed the upstream spread of the parasite by acting as a barrier impeding the upstream migration of trout.

Rio Grande Cutthroat Trout - Tables 3 and 4 contain the data summaries for trout population surveys and disease testing for three streams and five collection sites in the Rio Grande basin sampled during 2007. Trout collected at four of five collection sites tested negative for *M. cerebralis* infection. A single brook trout collected from Cumbres Creek tested PCR positive for infection. This was not surprising, given that both brook and brown trout collected from the Rio de los Pinos at two sites downstream of Trujillo Meadows Reservoir in 2004 near the summit of Cumbres Pass were heavily infected.

Colorado River Cutthroat Trout - Bio-statistical data for the fish population sampling and the corresponding PCR and PTD disease test results for the Colorado, Dolores, Eagle, Gunnison, San Juan, Uncompahgre, White and Yampa river basins are summarized in Tables 4 through 12. These rivers flow into the Colorado River basin that drains much of the southwestern U.S. Historically, all supported large populations of Colorado River native (CRN) cutthroat trout.

Trout population surveys were completed at 13 sites on nine streams tributary to the Dolores, Eagle and San Juan rivers in 2007 (Table 5). Colorado River cutthroat trout were encountered on four streams. Trout collected from 14 of 23 sampling sites tested positive for *M. cerebralis* infection by either PTD, PCR, or both (Table 6). Trout from 11 of the 23 sampling sites were provided by area biologists for disease testing and were not collected by Aquatic Research section personnel. In most instances the infection was detected in brown, brook or rainbow trout at sites that were not in the immediate proximity of streams supporting CRN trout. The only exception was Cross Creek, tributary to the Eagle River near Minturn, Colorado. Heavily infected brook and brown trout were collected near the lower terminus of Cross Creek. Cranial concentrations of *M. cerebralis* myxospores were very high among brown trout collected from numerous sites on the Eagle River during April 2007 by Area 8 personnel from the Glenwood Springs office. These data indicate that ambient levels of infection are very high in the reach of the Eagle River from at least the confluence with Turkey Creek near Red Cliff downstream to the confluence with Gore Creek west of Vail, Colorado.

Trout population surveys were completed at 12 sites on nine streams tributary to the Blue and Colorado rivers in 2007 (Table 7). Colorado River cutthroat trout were encountered on four streams. Allopatric populations of CRN trout were found on the North Fork of the Swan River, Clinton Creek upstream of Clinton Reservoir and the West Fork of Big Creek upstream of Bonham Reservoir. Brown and brook trout were sympatric with CRN trout on the Big Piney River approximately 4 km upstream of Piney Lake, north of Vail, Colorado. Trout collected from 19 of 21 sampling sites tested positive for *M. cerebralis* infection by either PTD, PCR, or both (Table 8). No CRN trout collected for disease screening tested positive for *M. cerebralis.* However, juvenile and adult brook trout sympatric with a single adult CRN trout in the Big Piney River 4 km upstream of Piney Lake tested positive for infection by PTD, while YOY brook and CRN trout collected from the same site tested negative for infection by the PCR test. These findings suggest that *M. cerebralis* is slowly spreading upstream in the Big Piney River via migration of trout infected with the parasite and that the area of the stream 4 km above Piney Lake was close to the upstream limit of spread in 2007.

Trout population surveys were completed at 26 sites on 24 streams tributary to the Gunnison and Uncompahgre river basins in 2007 (Table 9). Colorado River cutthroat trout were encountered on six streams. *Myxobolus cerebralis* infection was detected by PCR, PTD or both in trout collected at 15 sites from 13 of 24 streams (Table 10). Brook trout and CRN trout collected from East Dallas Creek SW of Ridgway, Colorado tested positive for infection. Two of 8 CRN trout tested positive by PCR and 1 of 10 brook trout tested positive by PTD. These data suggest the level of infection was low in 2007 and that exposure to the parasite in this reach of the stream is most probably a recent occurrence.

Trout population surveys were completed at 15 sites on eight streams tributary to the Yampa River in 2007 (Table 11). Colorado River cutthroat trout were encountered on seven of eight streams and were allopatric at all sampling sites. All CRN trout samples collected from streams tributary to the Yampa River tested negative for *M. cerebralis* infection (Table 12).

While no trout population surveys were conducted on any streams in the White River basin, fish collections to test for *M. cerebralis* infection were made at seven sites during 2007 (Table 12). Trout collected from all seven sites tested positive for infection. Prevalence of infection ranged between 50 and 100% among fish at all collection sites and severity of infection was high among fish testing positive. In Marvine Creek approximately 1 km downstream of Lower Marvine Lake, YOY brook trout with overt clinical signs of WD (blacktail, skeletal deformities and exophthalmia) were easily visible from the stream bank during mid-August. Exceptionally clear water in a shallow, narrow beaver pond approximately 0.5 km in length facilitated visual observation of the brook trout. There were virtually no trout under 30 cm in length, suggesting that the ambient level of infection was severe enough to result in acute mortality due to *M. cerebralis* infection before the end of the first year of life. The lack of fish in the population between 50-60 mm and 300 mm suggests that there has been very little recruitment beyond age 1 for at least two to three years. Brook trout collected by gill net from Upper and Lower Marvine lakes tested positive for infection by PTD, suggesting that *M. cerebralis* is probably enzootic throughout the drainage. Prevalence of infection was 100% (as determined by PCR testing) among YOY brook trout at all three sites tested in the vicinity of Marvine Lakes and in Marvine Creek downstream of the lakes. All of the foregoing indicates the intensity of *M. cerebralis* infection in the Marvine Creek basin is very high.

CRN trout collected in Cabin Creek and Fraser Creek (tributary to Trapper's Lake) tested positive for infection. The composite PCR scores for YOY CRN trout collected at the Fraser Creek inlet to Trapper's Lake was 47 of a possible maximum of 50, suggesting that ambient levels of infection are very high in the shallow bay at the south end of the lake (Table 12).

Up-ramping of *M. cerebralis* infection among brook trout and Colorado River cutthroat trout in Trappers Lake and its tributary streams, Cabin, Fraser and Heberton creeks is most troubling. This lake is in the Flattops Wilderness in the headwaters of the White River basin east of Meeker and has been the mainstay of spawning-taking operations for Colorado River cutthroat by the Colorado Division of Wildlife for a century. Overt clinical signs of WD were evident among many of the YOY and juvenile fish collected from these three streams as well as brook trout throughout the Marvine Creek basin. High levels of cranial myxospore concentrations among salmonids collected from several sites on the North Fork of the White River beginning at the USFS Himes Peak Campground just downstream from Trappers Lake suggests that Trappers Lake and/or microhabitats in the North Fork of the White River are point sources of infectivity (Nehring 2007). Prevalence of infection and cranial myxospore concentrations decrease among salmonids as the distance downstream from Trappers Lake increases. However, this is most likely an artifact of the changing species composition from cutthroat, brook and rainbow trout (at the higher elevations) to brown trout (at the lower elevations) and not necessarily evidence of increasing distance(s) from the point source(s) of infection. Brown trout are more resistant to infection and develop fewer cranial myxospores compared to levels observed in rainbow, brook and cutthroat trout exposed to the same levels of infection at the same point and time (Thompson

et al. 1999). The data collected during 2006 and 2007 demonstrate that there are most likely several point sources of *M. cerebralis* infectivity in the White River drainage in addition to Trappers Lake. Additional areas known to be locales of high *M. cerebralis* infection would include Upper and Lower Marvine Lakes, Slide Lake (Marvine Creek basin), Marvine Creek, North Elk Creek and Big Beaver Creek. There are likely others that have not been sampled as yet.

Tributary streams in the White River basin known to support viable populations of Colorado River cutthroat trout, include Big Beaver Creek upstream of Lake Avery, Fawn Creek, Lost Creek, the Middle Fork and East Fork of North Elk Creek, Snell Creek, Ute Creek, Fraser Creek, Cabin Creek and Heberton Creek. Only the Snell Creek population of Colorado River cutthroat trout is isolated upstream of a migration barrier. Migration barriers are of paramount importance to protect them from non-native salmonids carrying the *M. cerebralis* parasite that could migrate out of the White River, or the Big Beaver Creek inlet to Lake Avery. Although they do not produce high concentrations of cranial myxospores compared to similarly exposed rainbow trout, Colorado River cutthroat trout are highly vulnerable to the *M. cerebralis* infection and can suffer very high mortality when exposed to ambient levels of *M. cerebralis* actinospores as YOY fry in free-flowing streams (Thompson et al. 1999).

While there is currently no indication the mountain whitefish (*Prosopium williamsoni*) population in the White River drainage is significantly infected, laboratory exposure of 1 month-old whitefish alevins to *Mc* TAMs indicates there are very vulnerable to acute infection (George Schisler, unpublished data). Testing for *M. cerebralis* in the Roaring Fork River in Colorado in the 1990s revealed the prevalence and severity of infection among juvenile and adult whitefish was greater than that detected in concurrent collections of brown trout and rainbow trout (Nehring and Thompson 2003). Taken together, these data suggest that *M. cerebralis* infection could become problematic for the mountain whitefish population in the White River sub-basin of the Colorado River if the number of potential point sources of TAM production increase or ambient levels of infection increase over time.

Aquatic Oligochaete Sampling – Since 2001, substantial research efforts have been directed at developing a better understanding of the factors that affect the population dynamics and distribution of aquatic oligochaetes in the natural environment. In addition, much has been learned about the relative differences in susceptibility to *M. cerebralis* among the different lineages of *T. tubifex* (Beauchamp et al. 2001, 2002, 2005, 2006; DuBey and Caldwell 2004; DuBey et al. 2005; Kaesar and Sharpe 2006; Kerans et al. 2004: Winkelman and Nehring 2007). As more and more research investigations have been directed at the aquatic oligochaete side of the life cycle of *M. cerebralis* it has become increasing clear that the presence of the lineage III *T. tubifex* in an aquatic environment is often the primary determining factor governing whether or not *M. cerebralis* becomes established after the initial introduction occurs. In the San Juan River below Navajo Dam in New Mexico, DuBey and Caldwell (2004) found that only lineage III *T. tubifex* were infected with *M. cerebralis*, even though *T. tubifex* belonging to lineages I and VI were also present in the stream. Moreover, in a follow-up laboratory study where worms from lineages I, III and VI were exposed to myxospores of *M. cerebralis*, evidence of infection was only detected in lineage III worms (DuBey et al. 2005). Similar outcomes have emerged from laboratory tests where lineage I, III, IV, V and VI *T. tubifex* have been exposed to varying

concentrations of *M. cerebralis* myxospores in Colorado (Nehring, unpublished data), Oregon (Dr. Jerri Bartholomew, personal communication), California (Baxa and Hedrick 2008) and in states in the eastern U.S. (Dr. Vicki Blazer, personal communication). Although variations in sediment type or quality, (i.e., sand, mud or organically rich muck) can enhance the severity of infection among *T. tubifex* worms that are susceptible to *M. cerebralis*, parasite development and infectivity is not altered in lineages of worms that are refractory (V and VI) or highly resistant (I) to infection, regardless of sediment type (Baxa and Hedrick 2008). For these reasons, ascertaining the distribution and relative abundance of the various lineages of *T. tubifex* in Colorado's cutthroat trout streams is a critically important component in assessing the risk of establishment and spread of *M. cerebralis* in Colorado.

Over the past five years, the aquatic oligochaete field sampling effort has been a "learning experience". In 2003 and 2004, habitat(s) harboring oligochaetes were often difficult to find. Even at some sites where habitat conditions looked optimal (such as sediment-laden beaver ponds) considerable sampling was often required to find oligochaete colonies (Nehring 2004). Sample preservation ruined some samples. All of the samples from the upper Rio Grande basin in 2003 had no detectible DNA of *M. cerebralis* when tested by PCR. Apparently, chlorinated tap water was inadvertently used to dilute the pure ethanol to 70% for preservation of the samples. Minute amounts of chlorine will denature DNA, rendering PCR analysis ineffective.

Since 2004, greater effort has been expended to collect sediment samples and skills in recognizing probable habitats where oligochaetes are likely to occur have been improved. Aquatic oligochaetes in general more often than not have a very patchy distribution but live in very dense colonies. Visual detection of optimum areas to sample is a learned skill, that once acquired greatly enhances sampling efficiency. This has been the case on this study. Sample preservation and laboratory protocols have also improved. Taken together, these efforts have resulted in larger numbers of aquatic oligochaetes collected at most sampling sites since 2005. Results of the oligochaete sampling efforts for 2003, 2004, 2005 and 2006 are summarized in Appendix Tables 11 through 20 in the 2007 federal aid progress report (Nehring 2007). Summaries of the oligochaete samples collected and tested during the 2007-2008 study segment are shown in Tables 13 through 16.

Each sample location is referenced using global positioning technology (GPS). This allows all data to be plotted on maps that show the distribution of both aquatic oligochaetes and fish collected and tested for *M. cerebralis* infection. It also facilitates a visual representation of the distribution of the various lineages of *T. tubifex* by drainage basin for all of Colorado. Maps summarizing the distribution of those lineages can be seen in the Appendix. The locations of collections of *T. tubifex* identified as lineage I, III, V and VI are shown on Appendix maps 3, 4, 5 and 6, respectively. Examination of these maps indicate that *T. tubifex* belonging to lineage III are the most common and widely distributed throughout the Colorado, particularly within the Colorado River basin. The shaded dots on Map 7 show those sites where aquatic oligochaetes were collected, but there was no amplification of mitochondrial DNA for any of the four lineages of *T. tubifex*. Quality control checking has shown that tubificid DNA is present in the samples. Base pair sequence comparisons of DNA from some of the tubificid samples from the Rio Grande basin as well as other areas around the state with base pair sequences stored in GENBANK indicate the DNA contained in samples that did not amplify was usually from

Limnodrilus hoffmeisteri or *Ilyodrilus templetoni*. These two tubificid species are cosmopolitan and commonly found in both lake and stream habitats. Both occur in Windy Gap Reservoir and in the upper Colorado River basin (Zendt and Bergersen 2000). The open circles on Map 7 show the sites where sediment collections were made but no oligochaetes of any kind were detected.

It is unknown whether or not colder water temperatures or harsh environmental conditions found at high altitudes in the mountains of Colorado might prohibit the distribution of *T. tubifex,* particularly the lineage III worm that is the susceptible host for *M. cerebralis*. All sites where lineage III *T. tubifex* have been collected across Colorado from 2003 through 2007 were stratified into elevation zones to evaluate this possibility. The data summarized in the matrix format below do not support this hypothesis. Mitochondrial DNA specific to lineage III *T. tubifex* was detected in more worm samples at each 1,000 foot elevation increment between 6,000 and 11,000 feet above mean sea level than for lineages I, V and VI. It may be noteworthy that mitochrondial DNA for lineage III *T. tubifex* was detected in only one oligochaete sample collected at sites > 11,000 feet; however, oligochaetes have only been collected from eight sites at those high elevations. This is an inadequate sample size to determine whether or not harsh environmental conditions at elevations > 11,000 feet might be a factor limiting the distribution of *T. tubifex*. It is noteworthy that across all elevation zones in Colorado, mitochrondial DNA for lineage III *T. tubifex* was detected in 56% of the samples.

Elevation (ft.)	**Number of Sites where each Lineage of *Tubifex tubifex* was present**				
	Lineage I	**Lineage III**	**Lineage V**	**Lineage VI**	**No Lineage**
5,000 – 6,000	0	3	0	4	0
6,001 – 7,000	6	28	3	15	3
7,001 – 8,000	1	29	2	7	6
8,001 – 9,000	8	46	5	16	19
9,001- 10,000	3	33	2	7	25
10,001- 11,000	1	17	1	2	14
11,001 – 12,000	0	1	0	0	7
Total	19	157	13	51	74
Percent	7	56	5	18	26

Cutthroat Trout Population Status - More often than not, Colorado's self-sustaining populations of cutthroat trout persist at high elevations in stream reaches where the water temperatures are cold and the growing season can be quite short. In some cases these populations exist in habitats near the upper thermal limits of the species (Coleman and Fausch 2006). At lower elevations where the thermal regime may be more conducive to successful reproduction, growth and survival, cutthroat trout face extirpation due to competition from nonnative brown and brook trout (Peterson and Fausch 2003a; Peterson and Fausch 2003b; Peterson et al. 2004). Hybridization with nonnative rainbow trout dilutes the genetic purity. These factors, together with the high sensitivity of Colorado's cutthroat trout to infection by *M. cerebralis* (Thompson et al. 1999) present daunting obstacles for resource managers charged with recovery efforts.

One-time population surveys on short segments of stream (which were the norm in this study) are not adequate to not provide thorough insight or understanding of the degree of stability of the native salmonid populations. However, the large number of sites surveyed during this five-year study can provide some insight into the relative condition of Colorado's cutthroat trout sub-species. Tables 17, 18 and 19 in the Appendix summarize (for all streams and sites surveyed) the presence or absence of cutthroat trout, presence or absence of YOY cutthroat trout, an approximation of the number of cohorts of cutthroats observed within the study reach, and the presence or absence of nonnative salmonid competitors. Data for Rio Grande, greenback and Colorado River cutthroat trout are presented in Tables 17, 18 and 19, respectively. Table 20 contains a summary for each sub-species for each year of study (2003-2007) and the overall data set for each species. Given all that is known about the negative impacts of competition from nonnative salmonids existing in sympatry with Colorado's cutthroat trout sub-species (Peterson and Fausch 2003a; Peterson and Fausch 2003b; Peterson et al. 2004) it is not surprising that among allopatric cutthroat trout populations, three or more cohorts were found at 79 survey sites compared to only 36 sites when the cutthroat trout occurred in sympatry with one or more nonnative salmonid species (Tables 17, 18 and 19).

Among allopatric cutthroat trout populations we encountered 24 populations where the estimated number of cohorts present ranged between one and two. We found 33 sites where the estimated number of cohorts ranged between one and two among cutthroat trout populations sympatric with nonnative salmonids. These results may suggest (on a statewide basis) other abiotic factors may be a threat to the stability and persistence of numerous allopatric cutthroat trout populations.

CONCLUSIONS

Recent developments in the DNA typing and testing of the various lineages of *T. tubifex* for susceptibility or resistance to *M. cerebralis* infection are very encouraging. Results of studies conducted in Colorado, California , Oregon, New Mexico, and West Virginia have repeatedly shown that only lineage III *T. tubifex* are capable of producing TAMs of *M. cerebralis*. Oligochaetes belonging to lineages I, V and VI are either refractory or highly resistant to infection by the parasite and do not produce fish-infective TAMs. These results offer hope that lineage I, V and VI *T. tubifex* can act as "biofilters" to consume and deactivate *M. cerebralis* myxospores in habitats where the parasite is already enzootic, and dramatically reduce ambient levels of infection. Indeed, this appears to have been occurring at Windy Gap Reservoir in Colorado for the past five to six years (Winkelman and Nehring 2007).

Results of the aquatic oligochaete sampling and testing over the past five field seasons have demonstrated that the *M. cerebralis*-susceptible lineage III *T. tubifex* is far and away the most widely distributed of the four lineages *T. tubifex* known to occur in Colorado. Mitochondrial DNA specific to the lineage III oligochaete has been detected in far more worm samples at all elevation zones between 6,000 and 11,000 feet in Colorado than for worms belonging to lineages I, V and VI. Given the wide distribution and high abundance of the lineage III worms throughout Colorado, these findings indicate that the risk of establishment of *M. cerebralis* is quite high, once introduction into a previously unexposed aquatic ecosystem occurs.

The number of sites visited in 2006 (Nehring 2007) and 2007 where trout tested positive for *M. cerebralis* infection was considerably higher than in during the 2003, 2004 and 2005 field seasons. It was particularly troubling to see the high degree of prevalence and severity of infection evident in brook trout and cutthroat trout in Trappers Lake and Upper and Lower Marvine lakes in the Flattops Wilderness (Table 12). Unlike the case for Yellowstone Lake in Yellowstone National Park (Koel et al. 2006), the mechanism by which *M. cerebralis* was vectored into Trappers and Marvine lakes is probably not a mystery. All three lakes were probably exposed by upstream migrating trout infected with the *Mc* parasite sometime in the past ten years. *Myxobolus cerebralis* was detected in commercially-reared rainbow trout that had been stocked in a private pond on the North Fork of the White River in October 1988. The pond was fed by irrigation water diverted from the North Fork of the White River which then flowed back into the river. This pond was located approximately 10 km downstream of Marvine Creek where it flows into the North Fork of the White River. Trappers Lake is approximately 35 km upstream on the North Fork of the White River. *Myxobolus cerebralis* was first detected in brook trout and Colorado River cutthroat trout in Trappers Lake in 2003. However, age-specific disease testing indicated that fish older than the 1999 year class did not test positive for *Mc* myxospores (Kevin Rogers, personal communication). Clearly, there has been a significant up-ramping in the degree of infection among brook trout and cutthroat trout in these pristine lacustrine ecosystems in Colorado, similar to the case with Yellowstone Lake, where the parasite was first detected in 1998 (Koel et al. 2006).

After five field seasons, it is evident that *M. cerebralis* has become established in several aquatic habitats that support native cutthroat trout populations in Colorado. How serious a threat this poses for the recovery of Colorado's three sub-species of cutthroat trout is unknown. However, given the degree of spread of the parasite into high elevation habitats in the White and Yampa River basins that have direct connectivity to streams supporting excellent populations of Colorado River cutthroat trout, it is important that efforts be increased to construct barriers to isolate these populations and prevent invasion by non-native salmonids carrying the parasite from other areas where it is already enzootic.

Trappers Lake and Upper and Lower Marvine Lakes lie within the Flattops Wilderness Area in the White River National Forest. The headwaters of Piney Creek (above Piney Lake) north of Vail, Colorado lie within the Eagles Nest Wilderness Area. Like Yellowstone Lake in Yellowstone National Park, these aquatic habitats are in pristine areas, located at relatively high elevation, and have no habitat degradation problems. Yet the trout populations in all of these aquatic ecosystems are heavily infected with *M. cerebralis*. These cases belie the conventional "wisdom" that this parasite can only thrive in degraded or organically polluted environments. Rather, there are two important factors common to all four ecosystems. Those factors are 1) the presence of a highly susceptible salmonid host, and 2) the presence of lineage III *T. tubifex*. It has been a proven fact for a decade that brook trout and Colorado's three sub-species of native cutthroat trout are more prone to develop a lethal infection after exposure to *M. cerebralis* than either brown trout or rainbow trout exposed under identical conditions (Thompson et al. 1999). After five years of sampling and testing, we know that lineage III *T. tubifex* are highly abundant and the most widely distributed of the four lineages of *T. tubifex* known to occur in Colorado. Moreover, lineage III worms have been readily collected at all elevations in the state up to 3,354 meters (11,000 feet). The majority of core conservation populations of Colorado's native

cutthroat trout occur in lakes and streams at elevations between 2,439 and 3,354 meters (8,000 – 11,000 feet). The empirical evidence collected over the past five years reveal there is a **very high degree of congruence** between aquatic habitats that 1) either sustain or are capable of supporting core conservation populations of native cutthroat trout, and 2) support dense populations of lineage III *T. tubifex*. Given these realities, it would be foolhardy for fisheries resource managers to assume that threat or risk of exposure of Colorado's native cutthroat trout to *M. cerebralis* is minimal. On the contrary, the risk is high once introduction of the parasite occurs.

Testing for *M. cerebralis* in mountain whitefish on the White River was conducted in 2006 (Nehring 2007). Those results did not suggest ambient levels of infection were adequate to be causing population level impacts. However, it would be prudent to begin monthly YOY sampling efforts at selected sites in the White River basin in order to monitor changes in relative abundance of brown, brook, and rainbow trout fry as well as whitefish fry. There are plans to begin this sampling effort during the spring to fall period in 2008 in order to establish a baseline level of abundance for YOY fry of this species. This is especially important given that the mountain whitefish population in the White River in 2007 still appears to be very healthy, but could be negatively impacted if there is a significant increase in ambient levels of *M. cerebralis* infectivity in the basin in the future.

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APPENDIX A

TABLES

Table 1. Trout population biostatistics for trout ≥ 15 cm collected in streams and sampling sites within **Greenback** cutthroat trout (*Oncorhynchus clarki stomias*) recovery zones sampled during the summer of 2007.

Date	Brown Trout				Brook Trout				GBN Cutthroat Trout			
MMDDYY	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha
Cottonwood Creek, South (1 km upstream of Cottonwood Lake, lower station) GPS 13S 387634//4293200												
06/28/07	9[b]	--	48[b]	7[b]	1[b]	--	5[b]	0[b]	5[b]	--	82[b]	7[b]
Cottonwood Creek, South (10 km upstream of Cottonwood Lake, upper station) GPS 13S 379093//4290971												
06/28/07	--	--	--	--	4[e]	--	--	--	6[e]	--	--	--

[a]: Almost all trout captured were ≤150 mm. [b] Single electrofishing pass only; no population estimate. [c]: Rainbow trout, not brown, brook or cutthroat trout.
[d]: electrofishing for PTD and PCR samples only. [e] Disease collection only.

Table 2. Results of polymerase chain reaction (PCR) testing of young-of-the-year (YOY) salmonids and pepsin-trypsin digest (PTD) testing of salmonids ≥ age 1 for evidence of infection by *Myxobolus cerebralis* in drainages in the vicinity of streams designated as present or future areas for recovery of **Greenback** cutthroat (*Oncorhynchus clarki stomias*) trout in 2007. PCR score is the cumulative total for 10 fish (or standardized to 10 fish if "n" ≤ 9 or "n" ≥ 11) where a negative score= 1, weak positive (w+) =2, + = 3, ++ = 4, and +++ = 5. A cumulative score of 10 indicates all fish were negative and a score of 50 indicates all fish were rated 5 (+++). Fish from sites testing positive are highlighted in bold.

Stream Name	Approximate Collection Location	PCR (YOY)				PTD (≥ Age 1)			
		Species	N	N +	Score	N	n +	Mean (n+) myxospores	Range Myxospores (n+)
Greenback Cutthroat Trout *(Oncorhynchus clarki virginalis*) Recovery Areas and Nearby Tributary Streams and Reservoirs									
Cottonwood Creek,S.	**1 km above Cottonwood Lake**	**Brown**	**10**	**10**	**47**	**9**	**8**	**97,871**	**2,239 – 221,650**
Cottonwood Creek, S.	1 km above Cottonwood Lake	Brook	--	--	--	2	0	0	0
Cottonwood Creek, S.	10 km above Cottonwood Lake	GBN	6	0	10	5	0	0	0
Cottonwood Creek, S.	10 km above Cottonwood Lake	Brook	4	0	10	5	0	0	0

Table 3. Trout population biostatistics for trout ≥ 15 cm collected in streams and sampling sites within **Rio Grande** cutthroat trout (*Oncorhynchus clarki virginalis*) recovery zones sampled during the summer and fall of 2007.

Date	Brown Trout				Brook Trout				RGN Cutthroat Trout			
MMDDYY	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha
Cumbres Creek (At confluence with Rio de los Pinos) GPS 13S 374451//4096741												
07/25/07	17[a]	±3	931[a]	18	6	--	336	28	--	--	--	--
Rio Chama (Chama Basin at Forest Service corrals) GPS 13S 370138//4097705												
07/25/07	2[b]	--	54[b]	4[b]	--	--	--	--	--	--	--	--
Wolf Creek (At Cumbres Pass, below railroad bridge) GPS 13S 374452//4096742												
07/25/07	21	± 1	1114	196	--	--	--	--	--	--	--	--
Wolf Creek (At Cumbres Pass, above railroad bridge) GPS 13S 361946//40099853												
07/25/07	2[b]	--	211[b]	38[b]	--	--	--	--	2[b]	--	211[b]	14[b]
Wolf Creek (At Cumbres Pass above beaver dam and railroad bridge) GPS 13S 369221//4098333												
07/25/07	--	--	--	--	--	--	--	--	7	±106	448	18

[a]: Almost all trout captured were ≤150 mm. **[b]** Single electrofishing pass only; no population estimate. **[c]:** Rainbow trout, not brown, brook or cutthroat trout.
[d]: electrofishing for PTD and PCR samples only**.**

Table 4. Results of polymerase chain reaction (PCR) testing of young-of-the-year (YOY) salmonids and pepsin-trypsin digest (PTD) testing of salmonids ≥ age 1 for evidence of infection by *Myxobolus cerebralis* in drainages in the vicinity of streams designated as present or future areas for recovery of **Rio Grande** cutthroat (*Oncorhynchus clarki virginalis*) trout in 2007. PCR score is the cumulative total for 10 fish (or standardized to 10 fish if "n" ≤ 9 or "n" ≥ 11) where a negative score= 1, weak positive (w+) =2, + = 3, ++ = 4, and +++ = 5. A cumulative score of 10 indicates all fish were negative and a score of 50 indicates all fish were rated 5 (+++). Fish from sites testing positive are highlighted in bold.

Stream Name	Approximate Collection Location	PCR (YOY)				PTD (≥ Age 1)			
		Species	N	N +	Score	N	n +	Mean (n+) myxospores	Range Myxospores (n+)
Cumbres Creek	**1 km upstream Rio de los Pinos**	**Brook**	**10**	**1**	**14**	**10**	**0**	**0**	**0**
Rio Chama	At USFS campground/trailhead access	Brown	10	0	10	10	0	0	0
Wolf Creek	Below CO Hwy 17 road crossing	Brown	10	0	10	10	0	0	0
Wolf Creek	Cumbres Pass	Brown	10	0	10	2	0	0	0
Wolf Creek	Cumbres Pass	RGN	--	--	--	4	0	0	0

Table 5. Trout population biostatistics for trout ≥ 15 cm collected in streams and sampling sites within Colorado River cutthroat trout (*Oncorhynchus clarki pleuriticus*) recovery zones in the **San Juan, Dolores, and Eagle River** basins sampled during 2007.

Date	Brown Trout				Brook Trout				CRN Cutthroat Trout			
MMDDYY	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha
Cunningham Creek (above Silverton) GPS 13S 273029//4186895 (San Juan)												
10/23/07	--	--	--	--	--	--	--	--	20	± 1	451	46
Navajo River, Upper (on Banded Peak Ranch) GPS 13S 352040//4117731 (San Juan)												
07/24/07	--	--	--	--	--	--	--	--	13	±5	154	31
Navajo River, Lower (on Banded Peak Ranch) GPS 13S 349740//4104950 (San Juan)												
07/24/07	3[b]	--	4[b]	--	4[b]	--	5[b]	1[b]	--	--	--	--
Rio Blanco, Upper GPS 13S 344603//4124973 (San Juan)												
7/23/07	--	--	--	--	2[b]	--	48[b]	5[b]	--	--	--	--
Rio Blanco (Jim White's property) GPS 13S 340636//4119900 (San Juan)												
7/23/07	1[a]	--	5[a]	--	1	--	5	--	11[c]	± 21[c]	53[c]	20[c]
Rio Blanco, Lower (1 mile above 84 bridge) GPS 13S 340638//4119900 (San Juan)												
07/24/07	2[b]	--	11[b]	1[b]	--	--	--	--	3[bc]	--	16[bc]	1[bc]
Blue Creek, Upper (North of Nucla) GPS 12S 691684//4270378 (Dolores)												
09/13/07	--	--	--	--	--	--	--	--	18[c]	±0	461[bc]	55[bc]
Blue Creek, Lower (North of Nucla) GPS 12S 690658//4268832 (Dolores)												
09/13/07	--	--	--	--	--	--	--	--	62[ac]	±11[ac]	4521[ac]	40[ac]
Buckeye Creek (above Buckeye Reservoir) GPS 12S 670014//4257376 (Dolores)												
09/12/07	6	±0	203	24	--	--	--	--	19[c]	±0	643[c]	40[c]
Deep Creek (near Telluride) GPS 13S 242693 4205092 (Dolores)												
09/11/07	7[b]	--	68[b]	14[b]	--	--	--	--	--	--	--	--
Boothe Creek (near Vail) GPS 13S 386574//4389845 (Eagle)												
08/16/07	--	--	--	--	--	--	--	--	1[b]	--	32[b]	2[b]
Cross Creek GPS 13S 378371//4380662 (Eagle)												
08/17/07	9[d]	--	108[d]	15[d]	1[ad]	--	12[ad]	--	--	--	--	--
Pitkin Creek (approximately 2 km above trailhead) GPS 13S 388866//4389854 (Eagle)												
08/16/07	--	--	--	--	--	--	--	--	6[d]	--	46[d]	6[d]

[a]: Almost all trout captured were ≤150 mm. [b] Single electrofishing pass only; no population estimate. [c]: Rainbow trout, not brown, brook or cutthroat trout.
[d]: electrofishing for PTD and PCR samples only.

Table 6. Results of polymerase chain reaction (PCR) testing of young-of-the-year (YOY) salmonids and pepsin-trypsin digest (PTD) testing of salmonids ≥ age 1 for evidence of infection by *Myxobolus cerebralis* in drainages in the vicinity of streams designated as present or future areas for recovery of Colorado River cutthroat (*Oncorhynchus clarki pleuriticus*) trout in the **San Juan, Dolores, and Eagle River** basins in 2007. PCR score is the cumulative total for 10 fish (or standardized to 10 fish if "n" was ≤ 9 or ≥ 11) where a negative score= 1, weak positive (w+) =2, + = 3, ++ = 4, and +++ = 5. A cumulative score of 10 indicates all fish were negative and a score of 50 indicates all fish were rated 5 (+++). Fish from sites testing positive are highlighted in bold.

Stream Name	Approximate Collection Location	PCR (YOY)				PTD (≥ Age 1)			
		Species	N	n+	Score	N	N+	Mean (n+) myxospores	Range Myxospores (n+)
Blue Creek	Lower Station	Rainbow	10	0	10	--	--	--	--
Booth Creek	Tributary to Gore Creek	CRN	10	0	10	10	0	0	0
Buckeye Creek	**Above Buckeye Reservoir**	**Rainbow**	**12**	**10**	**32**	**11**	**11**	**243,182**	**30,556 – 1,033,333**
Buckeye Creek	Above Buckeye Reservoir	Brown	--	--	--	8	0	0	0
Cross Creek	**Near Minturn, CO**	**Brown**	**11**	**11**	**49**	**10**	**5**	**26,746**	**5,572 – 83,583**
Cross Creek	**Near Minturn, CO**	**Brook**	**1**	**1**	**50**	**--**	**--**	**--**	**--**
Cunningham Creek	Above Silverton	CRN	10	0	10	10	0	0	0
Deep Creek	**Near Telluride**	**Brown**	**10**	**0**	**10**	**10**	**3**	**25,648**	**6,667 – 46,667**
Dolores River	Bradfield Bridge	Rainbow	--	--	--	16	0	0	0
Dolores River	Bradfield Bridge	Brown	--	--	--	25	0	0	0
Dolores River	**Rock stockpile station**	**Brown**	**--**	**--**	**--**	**11**	**4**	**1,806**	**1,389 – 2,778**
Dolores River	Metaska station	Brown	--	--	--	10	0	0	0
Dolores River	**Dove Creek Pump Plant Stations**	**Brown**	**--**	**--**	**--**	**10**	**1**	**1,944**	**1,944**
Eagle River	**Site #1 at Redcliff, CO**	**Brown**	**--**	**--**	**--**	**10**	**8**	**79,293**	**2,794 – 287,828**
Eagle River	**Site #1.9 above Beldon**	**Brown**	**--**	**--**	**--**	**6**	**3**	**20,492**	**2,794 – 55,889**
Eagle River	**Site #2 at Beldon**	**Brown**	**--**	**--**	**--**	**5**	**4**	**11,178**	**5,589 – 16,767**
Eagle River	**Site #2.9 at Bishop Gulch**	**Brown**	**--**	**--**	**--**	**11**	**9**	**104,947**	**2,794 – 536,533**
Eagle River	**Site #3 at Elk Creek**	**Brown**	**--**	**--**	**--**	**10**	**6**	**53,560**	**5,589 – 111,778**
Eagle River	**Site #4 above Minturn**	**Brown**	**--**	**--**	**--**	**10**	**9**	**55,578**	**5,589 – 120,161**
Eagle River	**Site #5 below Minturn**	**Brown**	**--**	**--**	**--**	**10**	**8**	**84,881**	**2,794 – 438,728**
Eagle River	**Site #6 at Arrowhead**	**Brown**	**--**	**--**	**--**	**10**	**8**	**90,819**	**2,794 – 366,072**
Navajo River	Banded Peak Ranch	Brown	--	--	--	6	0	0	0
Navajo River	Banded Peak Ranch	Brook	10	0	10	4	0	0	0
Opal Lake	White Creek near Pagosa Springs	Brook	--	--	--	10	0	0	0
Pitkin Creek	Tributary to Gore Creek	CRN	1	0	10	1	0	0	0
Rio Blanco	1 mile upstream of US Hwy 84	Brown	--	--	--	5	0	0	0
Rio Blanco	1 mile upstream of US Hwy 84	Rainbow	1	0	10	5	0	0	0
Rio Blanco	Above private property	Brook	--	--	--	3	0	0	0
Rio Blanco	**Near Castle Creek Bridge**	**Rainbow**	**9**	**2**	**14**	**--**	**--**	**--**	**--**
Rio Blanco	Near Castle Creek Bridge	Brown	1	0	10	--	--	--	--

Table 7. Trout population biostatistics for trout ≥ 15 cm collected in streams and sampling sites within Colorado River cutthroat trout (*Oncorhynchus clarki pleuriticus*) recovery zones in the **Colorado River** basin sampled during 2007.

Date	Brown Trout				Brook Trout				Cutthroat Trout			
MMDDYY	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha
Big Creek, West Fork (at Grand Mesa) GPS 13S 248801//4330184												
08/26/07	--	--	--	--		--			2	±0	104	5
Big Creek, Middle Fork (above Bonham Reservoir at weir above road) GPS 13S 249096//4330986												
08/28/07	12	±8	529	45	2[c]	±0	92[c]	7[c]	8	±6	353	25
Clinton Creek (above reservoir) GPS 13S 399999//4362350												
08/15/07	--	--	--	--	--	--	--	--	2[b]	--	57[b]	8[b]
Piney River, Big (at Colorado River Confluence) GPS 13S 359516//4412934												
08/08/07	6[b]	--	16[b]	2[b]	--	--	--	--	1[bc]	--	3[bc]	--
Piney River, Upper Big (1.5 km below Piney Lake) GPS 13S 378385//4396389												
08/10/07	11	±1	151	12	4[b]	--	55[b]	4[b]	--	--	--	--
Piney River, Middle (At USGS gauge) GPS 13S 365248//4406054												
08/10/07	15	±1	193	38	--	--	--	--	8[c]	±0	103[c]	13[c]
Piney River, Upper (2.5 miles above Piney Lake) GPS 13S 382502//4398912												
09/10/07	3	±0	61	16	75	±8	1525	110	1	±0	20	3
Rock Creek (near McCoy) GPS 13S 353991//4426773												
08/08/07	36	±1	761	100	--	--	--	--	4[c]	±0	84[c]	11[c]
Rock Creek, Little (off Highway 134) GPS 13T 358978//4435720												
08/15/07	9	±3	544	56	4	±0	231	15				
Sheephorn Creek (near Radium) GPS 13S 369223//4098339												
08/08/07	11	±2	190	18	--	--	--	--	1[c]	±0	17[c]	0.2
Swan River, North Fork (Upper North Fork) GPS 13S 418635//4374124												
08/09/07	--	--	--	--	--	--	--	--	3	±0	34	2
Swan River, South Fork (Nuggetville) GPS 13S 322395//4530517												
08/09/07	--	--	--	--	5	±2	321	25	--	--	--	--

[a]: Almost all trout captured were ≤150 mm. **[b]** Single electrofishing pass only; no population estimate. **[c]:** Rainbow trout, not brown, brook or cutthroat trout. **[d]:** electrofishing for PTD and PCR samples only**.**

Table 8. Results of polymerase chain reaction (PCR) testing of young-of-the-year (YOY) salmonids and pepsin-trypsin digest (PTD) testing of salmonids ≥ age 1 for evidence of infection by *Myxobolus cerebralis* in drainages in the vicinity of streams designated as present or future areas for recovery of Colorado River cutthroat (*Oncorhynchus clarki pleuriticus*) trout in the **Colorado River** basin in 2007. PCR score is the cumulative total for 10 fish (or standardized to 10 fish if "n" was ≤ 9 or ≥ 11) where a negative score= 1, weak positive (w+) =2, + = 3, ++ = 4, and +++ = 5. A cumulative score of 10 indicates all fish were negative and a score of 50 indicates all fish were rated 5 (+++). Fish from sites testing positive are highlighted in bold. An "ns" abbreviation in any data column indicates no sample was collected.

Stream Name	Approximate Collection Location	PCR (YOY)				PTD (≥ Age 1)			
		Species	N	n+	Score	N	n+	Mean (n+) myxospores	Range Myxospores (n+)
Big Creek	**Upstream Bonham Reservoir**	**Brown**	**9**	**9**	**46**	**20**	**0**	**0**	**0**
Big Creek, W. Fk.	At Grand Mesa	CRN	10	0	10	10	0	0	0
Clinton Creek	Above Clinton Reservoir	CRN	10	0	10	8	0	0	0
Colorado River	Kemp/Breeze	Rainbow	2	2	50	--	--	--	--
Colorado River	Hitching Post	Rainbow	9	9	50	--	--	--	--
Colorado River	Lone Buck	Rainbow	11	10	46	--	--	--	--
Colorado River	Red Barn	Rainbow	7	7	49	--	--	--	--
Big Piney River	**6 miles above Colorado River**	**Rainbow**	**--**	**--**	**--**	**8**	**6**	**45,177**	**2,794 – 95,011**
Big Piney River	6 miles above Colorado River	Brown	--	--	--	2	0	0	0
Big Piney River	**At Colorado River confluence**	**Brown**	**--**	**--**	**--**	**7**	**5**	**98,923**	**19,561 – 290,622**
Big Piney River	**At Colorado River confluence**	**Rainbow**	**10**	**10**	**50**	**3**	**3**	**66,135**	**19,561 – 122,956**
Big Piney River	**0.5 miles below Piney Lake**	**Brown**				**12**	**6**	**39,006**	**5,572 – 66,867**
Piney River, M.	**At USGS Gauge**	**Rainbow**	**10**	**10**	**31**	**--**	**--**	**--**	**--**
Piney River	**Below Piney Lake**	**Brook**	**12**	**12**	**40**	**--**	**--**	**--**	**--**
Piney River	**Just above Piney Lake**	**Brook**	**--**	**--**	**--**	**15**	**6**	**38,148**	**556 – 97,222**
Piney River	**Above Piney Lake**	**Brook**	**10**	**10**	**46**	**10**	**0**	**0**	**0**
Piney River	**3 km upstream Big Piney Lake**	**Brook**	**11**	**0**	**10**	**14**	**8**	**65,306**	**1667 – 166,667**
Piney River	3 km upstream Big Piney Lake	CRN	12	0	10	--	--	--	--
Red Dirt Creek, W.	**North of Kremmling, CO**	**Brook**	**10**	**10**	**50**	**10**	**10**	**105,909**	**16,767 – 215,172**
Rock Creek, Little	**North of McCoy, CO**	**Brook**	**10**	**10**	**48**	**10**	**8**	**101,299**	**25,150 – 424,756**
Rock Creek	**North of McCoy, CO**	**Rainbow**	**10**	**9**	**33**	**4**	**2**	**356,623**	**139,306 – 573,939**
Rock Creek	**North of McCoy, CO**	**Brown**	**10**	**9**	**22**	**10**	**3**	**9,287**	**5,572 – 16,717**
Sheephorn Creek	**Near Radium, CO**	**Brown**	**10**	**8**	**25**	**10**	**5**	**42,349**	**5,572 – 89,156**
Sheephorn Creek	**Near Radium, CO**	**Rainbow**	**--**	**--**	**--**	**1**	**1**	**22,289**	**22,289**
Swan River, M. Fk.	**Near Missouri Gulch**	**Brook**	**10**	**5**	**24**	**10**	**0**	**0**	**0**
Swan River, S.	**Near Nuggetville, Co**	**Brook**	**10**	**1**	**12**	**10**	**0**	**0**	**0**
Swan River, N.	Upper North Fork	Brook	10	0	10	10	0	0	0

Table 9. Trout population biostatistics for trout ≥ 15 cm collected in streams and sampling sites within Colorado River cutthroat trout (*Oncorhynchus clarki pleuriticus*) recovery zones in the **Gunnison River** basin sampled during 2007.

Date	Brown Trout				Brook Trout				Cutthroat Trout			
MMDDYY	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha
Archuleta Creek (inlet to Upper Dome Lake) GPS 13S 348756//4228068												
06/27/07	--	--	--	--	3[b]	--	65	5	1[bc]	--	22[bc]	6[bc]
Anthracite Creek (Lake Irwin outlet) GPS 13S 316813//4305035												
07/12/07	--	--	--	--	9	±4	178	15	9[c]	±4[c]	178[c]	12[c]
Brush Creek (East River east of Crested Butte) GPS 13S 335167//4305751												
07/10/07	4	±0	71	6	1	±0	18	1	1[a]	±0	18[a]	--
Brush Creek, Middle (East River east of Crested Butte) GPS 13S 339257//4308918												
07/10/07	2[ab]	--	14[ab]	--	10[ab]	--	72[ab]	1[ab]	10[abc]	--	72[abc]	--
Cebolla Creek (at Mason Family State Wildlife Area) GPS 13S 321692//4219352												
10/25/07	5[b]	--	54[b]	2[2]	--	--	--	--	--	--	--	--
Cebolla Creek (above Spring Creek confluence) GPS 13S 321074//4216197												
10/25/07	3[b]	--	108[b]	5[b]	--	--	--	--	--	--	--	--
Cimarron Creek, West Fork Big (above Silverjack Reservoir) GPS 13S 278710//4224622												
07/19/07	--	--	--	--	--	--	--	--	9	±0	288	15
Cimarron Creek, Little (at Alpine trailhead bridge) GPS 13S 275987//4227753												
07/19/07	--	--	--	--	6[b]	--	120[b]	9[b]	--	--	--	--
Cimarron Creek, Middle Fork Big (at wilderness boundary above Silverjack Reservoir) GPS 13S 278710//4224621												
07/19/07	--	--	--	--	2[bf]	--	8[bf]	--	10[b]	--	39[b]	5[b]
Coal Creek (Kebler Pass) GPS 13S 318597//4302765												
07/12/07	--	--	--	--	11	±1	622	42	--	--	--	--
Cow Creek (Uncompahgre east of Ridgeway) GPS 13S 270179 4221679												
07/18/07	--	--	--	--	--	--	--	--	6[b]	--	54[b]	--
East Dallas Creek (At lower horse camp and at Blue lakes trailhead) GPS 13S 253497//4214930												
10/23/07	--	--	--	--	15[d]	--	34[d]	5[d]	8[d]	--	18[d]	2[d]
Dominguez Creek, Big (Gunnison River #1 west of Broughton) GPS 12S 712953//4291082												
11/14/07	--	--	--	--	--	--	--	--	32[c]	±2[c]	1,432[c]	103[c]
Escalante Creek (below forks on BLM) GPS 12S 731310//4282143												
11/12/07	--	--	--	--	--	--	--	--	3[bc]	--	14[bc]	3[bc]
Escalante Creek, East Fork (at State Wildlife Area) GPS 12S 726119//44273205												
11/12/07	--	--	--	--	5[bf]	--	53[bf]	--	10[bc]	--	107[bc]	15[bc]
Henson Creek (above falls) GPS 13S288938//4210732												
10/24/07	--	--	--	--	20[ad]	--	86[ad]	1[ad]	--	--	--	--

[a]: Almost all trout captured were ≤150 mm. **[b]** Single electrofishing pass only; no population estimate. **[c]:** Rainbow trout, not brown, brook, or cutthroat trout.
[d]: electrofishing for PTD and PCR samples only. **[e]:** estimates include young-of-the-year. **[f]:** Cutbow trout, not brook trout

Table 9 *continued*. Trout population biostatistics for trout ≥ 15 cm collected in streams and sampling sites within Colorado River cutthroat trout (*Oncorhynchus clarki pleuriticus*) recovery zones in the **Gunnison River** basin sampled during 2007.

Date	Brown Trout				Brook Trout				Cutthroat Trout			
MMDDYY	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha
Lake Fork of the Gunnison (above Lake San Cristobal) GPS 13S 296937//4201512												
10/24/07	1[d]	--	2	--	8[d]	--	14[d]	1[d]	--	--	--	--
Marshall Creek (Tomichi Creek at Sargents) GPS 13S 377301//4265331												
07/09/07	19	±5	547	49	--	--	--	--	--	--	--	--
Oh Be Joyful Creek (North of Crested Butte) GPS 13S 323458//4308762												
07/11/07	--	--	--	--	1[b]	--	51[b]	2[b]	--	--	--	--
Pass Creek tributary to Willow Creek (East of Taylor Reservoir) GPS 13S 374458//4300483												
06/28/07	3[b]	--	77[b]	9[b]	4[b]	--	103[b]	7[b]	--	--	--	--
Pass Creek (below Golden Lakes) tributary to Ohio Creek (Gunnison River Basin) GPS 13S 310656//429492												
09/30/07	--	--	--	--	--	--	--	--	11[bg]	--	120[bg]	--
Rock Creek (East River North of Crested Butte) GPS 13S 326198//4316694												
07/13/07	--	--	--	--	3[b]	--	144[b]	8[b]	--	--	--	--
Slate River (North of Crested Butte) GPS 13S 335157//4305749												
07/11/07	1	±0	20	12	6	±0	122	10	3	±7	61	26
Spring Creek (above Cebolla Creek confluence) GPS 13S 323178//4216553												
10/25/07	24	±1	786	102	11	±2	368	52	--	--	--	--
Tomichi Creek, Upper (above Marshall Creek confluence) GPS 13S 250255//4261168												
07/09/07	2[b]	--	15[b]	1[b]	--	--	--	--	--	--	--	--
Tomichi Creek, Lower (below Marshall Creek confluence) GPS 13S 374927//4250169												
07/09/07	22[ab]	--	95[ab]	1[ab]	--	--	--	--	--	--	--	--

[a]: Almost all trout captured were ≤150 mm. **[b]** Single electrofishing pass only; no population estimate. **[c]:** Rainbow trout, not brown, brook, or cutthroat trout.
[d]: electrofishing for PTD and PCR samples only. **[e]:** estimates include young-of-the-year. **[f]:** Cutbow trout, not brook trout.
[g]: Little Kern River Golden trout, not cutthroat trout.

Table 10. Results of polymerase chain reaction (PCR) testing of young-of-the-year (YOY) salmonids and pepsin-trypsin digest (PTD) testing of salmonids ≥ age 1 for evidence of infection by *Myxobolus cerebralis* in drainages in the vicinity of streams designated as present or future areas for recovery of Colorado River cutthroat (*Oncorhynchus clarki pleuriticus*) trout in the **Gunnison River** basin in 2007. PCR score is the cumulative total for 10 fish (or standardized to 10 fish if "n" was ≤ 9 or ≥ 11) where a negative score= 1, weak positive (w+) =2, + = 3, ++ = 4, and +++ = 5. A cumulative score of 10 indicates all fish were negative and a score of 50 indicates all fish were rated 5 (+++). Fish from sites testing positive are highlighted in bold. An "ns" abbreviation in any data column indicates no sample was collected.

Stream Name	Approximate Collection Location	PCR (YOY)				PTD (≥ Age 1)			
		Species	N	n+	Score	N	n+	Mean (n+) myxospores	Range Myxospores (n+)
Anthracite Creek	**Below Lake Irwin**	**Brook**	**10**	**0**	**10**	**10**	**6**	**80,227**	**17,911 – 268,667**
Archuleta Creek	Upstream of Upper Dome Lake	Rainbow	--	--	--	1	0	0	0
Archuleta Creek	**Upstream of Dome Lake**	**Brook**	**--**	**--**	**--**	**4**	**3**	**23,882**	**4,478 – 60,450**
Brush Creek	**Near Crested Butte, CO**	**Brook**	**--**	**--**	**--**	**5**	**1**	**20,150**	**20,150**
Brush Creek	**Near Crested Butte, CO**	**Brown**	**--**	**--**	**--**	**5**	**3**	**16,418**	**4,478 – 33,583**
Brush Creek, Middle	Near Crested Butte, CO	Brook	10	0	10	10	0	0	0
Cebolla Creek	**At Cebolla Creek SWA**	**Brown**	**10**	**10**	**29**	**10**	**5**	**3,555**	**1,111 – 5,556**
Cebolla Creek	**Above Spring Creek**	**Brown**	**10**	**10**	**29**	**--**	**--**	**--**	**--**
Cebolla Creek	**Above Spring Creek**	**Brook**	**--**	**--**	**--**	**10**	**3**	**2,500**	**1,389 – 2,778**
Cimarron Creek, Little	At Alpine Plateau TH	Brook	10	0	10	10	0	0	0
Cimarron River, M. F.	Tributary to Gunnison	CRN	--	--	--	10	0	0	0
Cimarron River, W. F.	Tributary to Gunnison	CRN	10	0	10	10	0	0	0
Coal Creek	**Near Crested Butte, CO**	**Brook**	**10**	**10**	**38**	**10**	**7**	**96,912**	**2,239 – 304,489**
Cow Creek	Near Ridgeway, CO	CRN	--	--	--	6	0	0	0
Dallas Creek, E.	**Uncompahgre River Basin**	**Brook**	**--**	**--**	**--**	**10**	**1**	**8,889**	**8,889**
Dallas Creek, E.	**Uncompahgre River Basin**	**CRN**	**8**	**2**	**15**	**--**	**--**	**--**	**--**
Dominguez Creek, Big		Rainbow	10	0	10	10	0	0	0
Gunnison River	**Upper Ute Park**	**Rainbow**	**20**	**20**	**50**	**--**	**--**	**--**	**--**
Gunnison River	**Lower Ute Park**	**Rainbow**	**11**	**11**	**50**	**--**	**--**	**--**	**--**
Henson Creek	**Above Treasure Falls**	**Brook**	**10**	**4**	**20**	**10**	**0**	**0**	**0**
Lake Fork Gunnison	**Above Lake San Cristobal**	**Brown**	**7**	**7**	**47**	**--**	**--**	**--**	**--**
Lake Fork Gunnison	**Above Lake San Cristobal**	**Brook**	**3**	**3**	**50**	**10**	**9**	**79,537**	**16,667 – 256,667**
Marshall Creek	**East of Sargents, CO**	**Brown**	**--**	**--**	**--**	**10**	**8**	**31,624**	**6,717 – 80,600**
Oh-be-joyful Creek	Near Crested Butte, CO	Brook	11	0	10	10	0	0	0
Pass Creek	**East of Taylor Reservoir**	**Brown**	**6**	**6**	**45**	**2**	**2**	**44,778**	**29,106 – 60,450**
Pass Creek	**East of Taylor Reservoir**	**Brook**	**4**	**4**	**50**	**10**	**9**	**71,005**	**8,956 – 210,456**
Rock Creek	**Near Crested Butte, CO**	**Brook**	**10**	**4**	**15**	**10**	**1**	**4,478**	**4,478**
Slate River	Near Crested Butte, CO	Brook	10	0	10	10	0	0	0
Spring Creek	Above Cebolla Creek	Brook	10	0	10	10	0	0	0
Tomichi Creek, L.	**West of Sargents, CO**	**Brown**	**10**	**10**	**44**	**10**	**7**	**66,207**	**15,672 – 167,917**
Tomichi Creek, U.	**North of Sargents, CO**	**Brown**	**10**	**10**	**43**	**11**	**8**	**56,937**	**2,239 – 141,050**

Table 11. Trout population biostatistics for trout ≥ 15 cm collected in streams and sampling sites within Colorado River cutthroat trout (*Oncorhynchus clarki pleuriticus*) recovery zones in the **Yampa River** basin sampled during 2007.

Date	Brown Trout				Brook Trout				Cutthroat Trout			
MMDDYY	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha	N	95% CI	N/Ha	Kg/Ha
Beaver Creek, tributary to S. Fork of the Williams Fork of the Yampa River (2 km from FS boundary) GPS 13T 297374//4456711												
08/22/07	--	--	--	--	--	--	--	--	6	±2	128	8
North Fork Elkhead Creek, Upper site (0.5 km above Trail 1146 crossing) GPS 13T 3077706//4510417												
08/24/07	--	--	--	--	--	--	--	--	3	±0	64	6
North Fork Elkhead Creek, Middle site(0.5 km above Trail 1146 crossing) GPS 13T 307458//4510120												
08/24/07	--	--	--	--	--	--	--	--	9	±0	43	3
North Fork Elkhead Creek, Lower site (North of Hayden) GPS 13T 307500//4509800												
08/24/07	--	--	--	--	--	--	--	--	1	±0	7	1
Indian Run Creek (tributary to Beaver Creek-S. Fork of the Williams Fork of the Yampa River) GPS 13T 293927//4458972												
08/22/07	--	--	--	--	--	--	--	--	7[b]	--	228[b]	22[b]
Johnson Creek-tributary to South Fork of the Little Snake River (old station #2, 0.5 miles upstream from confluence) GPS 13T 323050//4530159												
08/21/07	--	--	--	--	--	--	--	--	9	±0	219	15
South Fork of the Little Snake River, upper site, headwaters (Three Forks Ranch, above T in road – severe grazing impacts) GPS 13T 321437//4526649												
08/21/07	--	--	--	--	--	--	--	--	9	±0	242	23
South Fork of the Little Snake River, middle site (Three Forks Ranch, above Oliver Creek) GPS 13T 322189//4530280												
08/21/07	*No trout captured or observed in 1,200 feet of electroshocking – severe degradation from cattle grazing											
South Fork of the Little Snake River, lower site (below Oliver Creek confluence on Three Forks Ranch) GPS 13T 322302//4530388												
08/21/07	*No trout captured or observed in 300 feet of electroshocking – severe degradation from cattle grazing											
Oliver Creek – upper site (1500 feet above confluence w/ South Fork Little Snake River) GPS 13T 322374//4530187												
08/21/07	*No trout captured or observed in 300 feet of electroshocking – severe degradation from cattle grazing											
Smith Creek, Upper site(Tributary to the Elk River off USFS Road 56A SSW of Steamboat Lake) GPS 13T 328174//4509099												
08/23/07	--	--	--	--	--	--	--	--	9	±0	373	8
Smith Creek, Middle site(Tributary to the Elk River off USFS Road 56A SSW of Steamboat Lake) GPS 13T 328175//4509097												
08/23/07	--	--	--	--	--	--	--	--	2[b]	--	56[b]	3[b]
Smith Creek, Lower site(Tributary to the Elk River off USFS Road 56A SSW of Steamboat Lake) GPS 13T 327449//4507015												
08/23/07	--	--	--	--	--	--	--	--	1	±0	49	2
South Fork of the Williams Fork of the Yampa River – upper site (5.5 km from Seely Ranch, start of USFS Trail 1113) GPS 13T 292802//4449549												
08/22/07	--	--	--	--	--	--	--	--	3	±0	77	8
South Fork of the Williams Fork of the Yampa River (Lower station approx. 2 km upstream of Seely Ranch HQ) GPS 13T 292215//4450899												
08/22/07	--	--	--	--	--	--	--	--	6	±2	145	12

[a]: Almost all trout captured were ≤150 mm. **[b]** Single electrofishing pass only; no population estimate. **[c]:** Rainbow trout, not brown trout or cutthroat trout.
[d]: electrofishing for PTD and PCR samples only. **[e]:** young-of-the-year included in estimate.

Table 12. Results of polymerase chain reaction (PCR) testing of young-of-the-year (YOY) salmonids and pepsin-trypsin digest (PTD) testing of salmonids ≥ age 1 for evidence of infection by *Myxobolus cerebralis* in drainages in the vicinity of streams designated as present or future areas for recovery of Colorado River cutthroat (*Oncorhynchus clarki pleuriticus*) trout in the **Yampa and White River** basins in 2007. PCR score is the cumulative total for 10 fish (or standardized to 10 fish if "n" was ≤ 9 or ≥ 11) where a negative score= 1, weak positive (w+) =2, + = 3, ++ = 4, and +++ = 5. A cumulative score of 10 indicates all fish were negative and a score of 50 indicates all fish were rated 5 (+++). Fish from sites testing positive are highlighted in bold. An "ns" abbreviation in any data column indicates no sample was collected.

Water Name	Approximate Collection Location	PCR (YOY)				PTD (≥ Age 1)			
		Species	N	n+	Score	N	N+	Mean (n+) myxospores	Range Myxospores (n+)
Beaver Creek	S. Fork Williams Fork-Yampa R.	CRN	10	0	10	--	--	--	--
Bel-Aire SWA	White River -Hatchery Ponds	Rainbow	--	--	--	32	0	0	0
Bel-Aire SWA	**White River -Hatchery Ponds**	**Brook**	**--**	**--**	**--**	**10**	**8**	**61,444**	**3,072 – 156,683**
Bel-Aire SWA	White River - Hatchery Ponds	Brown	--	--	--	1	0	0	0
Cabin Creek	**Trappers Lake - fish trap weir**	**Brook**	**3**	**2**	**27**	**--**	**--**	**--**	**--**
Cabin Creek	**Trappers Lake - fish trap weir**	**CRN**	**15**	**6**	**25**	**--**	**--**	**--**	**--**
N. Elk Creek Ranch	**Wetland spring seep outlet**	**Rainbow**	**--**	**--**	**--**	**2**	**1**	**52,228**	**52,228**
N. Elk Creek Ranch	**Wetland spring seep outlet**	**Brown**	**--**	**--**	**--**	**4**	**4**	**197,390**	**18,433 – 466,978**
Elkhead Creek, N. Fork	Near Hayden	CRN	--	--	--	10	0	0	0
Fraser Creek	**Inlet to Trapper's Lake**	**CRN**	**9**	**9**	**47**	**--**	**--**	**--**	**--**
Marvine Creek	**Below Marvine Lake**	**Brook**	**10**	**10**	**46**	**11**	**7**	**42,715**	**2,794 – 103,394**
Marvine Creek	**At Marvine Lake outlet**	**Brook**	**10**	**10**	**44**	**--**	**--**	**--**	**--**
Marvine Lake, Upper	**Marvine Creek**	**Brook**	**--**	**--**	**--**	**20**	**11**	**58,255**	**5,572 – 172,739**
Marvine Lake, Lower	**Marvine Creek**	**Brook**	**10**	**10**	**50**	**11**	**3**	**70,983**	**33,533 – 168,239**
Marvine Lake, Lower	Marvine Creek	Rainbow	--	--	--	1	0	0	0
Smith Creek	North of Steamboat Springs	CRN	--	--	--	10	0	0	0
William's Fork, S. Fork	Near Seely Ranch	CRN	--	--	--	10	0	0	0

Table 13. Aquatic oligochaete collections within **Rio Grande** cutthroat trout (*Oncorhynchus clarki pleuriticus)* recovery zones in 2007. Numbers of oligochaetes represent the number of tubificid worms enumerated in qualitative kick screen samples taken from sedimented areas in the stream. Percentage of DNA by strain type represents the proportion of DNA for each of the four lineages of *Tubifex tubifex* detected by a 4 probe multi-plex qPCR test developed by Pisces Molecular LLC, Boulder, Colorado using genetic markers developed at the University of California-Davis (Beauchamp et al. 2002).

Stream Name	Sample Date Mm/dd/yy	GPS Coordinates	Number of Oligochaetes			Percent DNA by Strain Type			
			Total	Haired	Non-Haired	I	III	V	VI
Rio Chama	07/25/07	13S 361946//40999853	100	100	0	0	0	0	0
Rio Grande (Hanna Lane Bridge)	11/05/07	13S 371332//4172109	106	100	6	0	3	0	97
Rio Grande (Gerrard Bridge)	11/05/07	13S 359725//4171013	277	111	166	0	23	0	77
Rio Grande (Lower Coller SWA)	11/05/07	13S 348267//4176962	147	100	47	0	64	0	36
Rio Grande (Wagon Wheel Gap)	11/05/07	13S 338615//4182326	104	11	93	0	65	0	35
Rio Grande (Marshall Park Bridge)	11/05/07	13S 325639//4184478	124	100	24	11	60	0	29
Rio Grande (Fisherman Area)	11/05/07	13S 323410//4182808	103	100	3	7	35	0	58
Rio de los Pinos (Cumbres confluence)	07/25/07	13S0374451//4096741	112	100	12	0	100	0	0
Navajo River (Banded Peak Ranch)	07/24/07	13S0349740//4104950	166	100	66	0	65	0	35
Navajo River (Upper)	07/24/07	13S0352040//4117731	359	6	353	0	0	0	0
Wolf Creek (above Railroad)	07/26/07	13S0361946//4099853	189	100	88	0	100	0	0

nd[a] **:** Haired or non-haired traits were not determined. [a]**:** nw indicates no worms were visible in the sediment sample

Table 14. Aquatic oligochaete collections within **Colorado** cutthroat trout (*Oncorhynchus clarki plueriticus*) recovery zones in the **Colorado, Eagle, Dolores, Arkansas, and San Juan River** basins in 2007. Numbers of oligochaetes represent the number of tubificid worms enumerated in qualitative kick screen samples taken from sedimented areas in the stream. Percentage of DNA by strain type represents the proportion of DNA for each of the four lineages of *Tubifex tubifex* detected by a 4 probe multi-plex qPCR test developed by Pisces Molecular LLC, Boulder, Colorado using genetic markers developed at the University of California-Davis (Beauchamp et al. 2002).

Stream Name	Sample Date mmddyy	GPS Coordinates	Number of Oligochaetes			Percent DNA by Strain Type			
			Total	Haired	Non-Haired	I	III	V	VI
Big Creek (Above Bonham Reservoir)	08/28/07	13S 249096//4330986	103	100	3	0	100	0	0
Big Creek, W. Fk.	08/28/07	13S 248801//4330184	100	100	0	0	100	0	0
Buckeye Creek	09/12/07	12S 670014//4257376	102	100	2	0	100	0	0
Clinton Creek	08/15/07	13S 399901//4362409	121	100	21	0	50	50	0
Cross Creek	08/17/07	13S 378371//4380662	100	100	0	0	100	0	0
Cunningham Creek	10/23/07	13S 273029//4186895	115	100	15	0	0	50	0
Eagle River (At Wolcott CDOT)	08/17/07	13S 355406//4396114	100	100	0	80	14	6	0
Huerfano River, S. Fk.	11/15/07	13S 463676//4169567	103	100	3	0	100	0	0
Piney River (Above Piney Lake)	09/10/07	13S 382502//4398912	109	100	9	0	0	0	0
Big Piney River (Upper)	08/08/07	13S 378469//4397392	212	58	154	0	100	0	0
Big Piney River (Middle)	08/10/07	13S 365248//4406054	119	118	1	0	100	0	0
Big Piney River (Lower)	08/08/07	13S 359516//4412934	210	100	110	63	28	2	7
Rio Blanco (Upper)	07/23/07	13S 344603//4124973	53	0	53	0	0	100	0
Rio Blanco (White's Property)	07/23/07	13S 340636//4119900	85	10	75	0	100	0	0
Rio Blanco (Lower)	07/24/07	13S 340638//4119900	368	104	264	0	100	0	0
Rock Creek, Little (Above Rock Creek)	08/15/07	13T 358978//4435720	136	100	36	68	32	0	0
Sheephorn Creek	08/08/07	13S 369210//4422121	105	100	5	0	100	0	0

nd[a] **:** Haired or non-haired traits were not determined. [a]**:** nw indicates no worms were visible in the sediment sample

Table 15. Aquatic oligochaete collections within **Colorado** cutthroat trout (*Oncorhynchus clarki plueriticus*) recovery zones in the **Gunnison River** basins in 2007. Numbers of oligochaetes represent the number of tubificid worms enumerated in qualitative kick screen samples taken from sedimented areas in the stream. Percentage of DNA by strain type represents the proportion of DNA for each of the four lineages of *Tubifex tubifex* detected by a 4 probe multi-plex qPCR test developed by Pisces Molecular LLC, Boulder, Colorado using genetic markers developed at the University of California-Davis (Beauchamp et al. 2002).

Stream Name	Sample Date mmddyy	GPS Coordinates	Number of Oligochaetes			Percent DNA by Strain Type			
			Total	Haired	Non-Haired	I	III	V	VI
Anthracite Creek, Upper	07/25/07	13S 316813//4305035	154	3	151	0	100	0	0
Archuleta Creek	06/29/07	13S 348756//4228068	4	3	1	0	55	0	45
Brush, Middle	07/10/07	13S 339257//4308918	140	109	31	0	50	0	0
Cebolla Creek (@ Mason Family SWA)	10/25/07	13S 321692//4219352	106	100	6	0	100	0	0
Cebolla Creek (Above Spring Creek confluence)	10/24/07	13S 321074//4216197	100	100	0	0	100	0	0
Cimarron Creek, Little	07/19/07	13S 275987//4227753	188	100	88	0	0	0	0
Coal Creek	07/12/07	13S 318597//4302765	100	100	0	76	24	0	0
Cochetopa Creek (near Archuleta Creek confluence)	07/26/07	13S 347907//4231029	102	100	1	0	100	0	0
Cottonwood Creek, South (Above Cottonwood Lake)	06/28/07	13S 387634//4293200	0	0	0	--	--	--	--
Cow Creek	07/18/07	13S 270179//4221679	135	100	35	0	100	0	0
Dallas Creek, East	10/23/07	13S 253497//4214930	51	50	1	0	100	0	0
Dominguez Creek, Big	11/14/07	12S 712953//4291082	48	43	5	0	100	0	0
Escalante Creek (Above Escalante Forks)	11/13/07	12S 731310//4282143	204	100	104	0	100	0	0
Gunnison River (Upper Ute Park)	06/13/07	13S 251205//4282784	188	100	88	0	23	0	77
Gunnison River (Lower Ute Park)	06/13/07	13S 252382//4284855	370	83	287	0	0	0	100
Gunnison River (At island pool)	09/26/07	13S 252161//4283800	270	64	206	0	14	0	86
Lake Fork of the Gunnison	10/24/07	13S 296937//4201512	100	100	0	0	11	0	89
Marshall Creek	07/09/07	13S 377301//4265331	120	101	17	0	100	0	0
Oh-Be-Joyful Creek	07/11/07	13S 323458//4308762	113	0	113	--	--	--	--
Rock Creek	07/13/07	13S 326198//4316694	168	100	68	0	0	0	0
Slate Creek	07/11/07	13S 335157//4305749	102	100	2	0	100	0	0
Spring Creek (Above Cebolla Creek)	10/25/07	13S 323178//4216553	2	0	2	--	--	--	--
Tomichi Creek, Lower	07/09/07	13S 374927//4250169	100	100	0	0	100	0	0
Tomichi Creek, Upper	07/12/07	13S 250255//4261168	226	100	126	0	100	0	0
Uncompahgre River (Baldridge Park)	02/07/07	13S 248711//4250169	213	213	0	0	87	0	13
Willow Pass Creek	06/28/07	13S 374458//4300483	14	14	0	0	100	0	0

nd[a] **:** Haired or non-haired traits were not determined. [a]**:** nw indicates no worms were visible in the sediment sample

Table 16. Aquatic oligochaete collections within **Colorado** cutthroat trout (*Oncorhynchus clarki plueriticus*) recovery zones in the **White and Yampa River** basins in 2007. Numbers of oligochaetes represent the number of tubificid worms enumerated in qualitative kick screen sample taken from sedimented areas in the stream. Percentage of DNA by strain type represents the proportion of DNA for each of the four lineages of *Tubifex tubifex* detected by a 4-probe multi-plex q PCR test developed by Pisces Molecular LLC, Boulder, Colorado using genetic markers developed at the University of California-Davis (Beauchamp et al. 2002).

Stream Name	Sample Date mmddyy	GPS Coordinates	Number of Oligochaetes			Percent DNA by Strain Type			
			Total	Haired	Non-Haired	I	III	V	VI
Beaver Creek (At Indian Run SWA)	08/22/07	13T 297374//4456711	102	100	2	0	100	0	0
Cabin Creek (Trapper's Lake)	05/31/07	13S 310028//4429067	176	112	64	0	100	0	0
Cabin Creek (Trapper's Lake)	10/09/07	13S 310028//4429067	35	8	27	0	100	0	0
Fraser Creek (Trapper's Lake)	05/31/07	13S 309984//4428071	128	11	192	0	50	0	0
Fraser Creek (Trapper's Lake)	10/09/07	13S 309984//4428071	682	600	82	0	98	0	2
Heberton Creek (Trapper's Lake)	10/09/07	13S 309117//4428276	54	50	4	0	100	0	0
Heberton Creek (Trapper's Lake)	06/01/07	13S 309117//4428276	307	15	292	0	0	0	0
Indian Run (Above Beaver Creek)	08/22/07	13T 293927//4458972	100	100	0	0	100	0	0
Little Snake, S. Fk.	08/21/07	13T 321437//4526649	100	100	0	0	100	0	0
Poose Creek	09/06/07	13S 307939//4445249	249	211	38	0	100	0	0
Scott's Bay (Trapper's Lake)	05/31/07	13S 308957//4439058	203	11	192	0	20	30	0
Smith Creek	08/24/07	13T 327449//4507015	101	100	1	0	100	0	0
Slide Lake (Marvine Creek)	08/17/07	13S 294127//4428376	118	100	18	0	100	0	0
Trapper's Lake (Lake outlet)	05/31/07	13S 309555//4429809	103	0	103	0	0	0	0
William's Fork, S. Fk. (Yampa River)	08/22/07	13T 292215//4450899	116	100	16	0	100	0	0
White River (CR 54 Bridge)	09/05/07	13S 269449//4425614	317	257	60	11	6	0	83
White River (Durey's Resort)	04/18/07	13S 266524//4435370	103	100	3	0	87	0	13
White River (K-Slash-K Bridge)	04/18/07	13S 263278//4425371	108	100	8	0	23	0	77
White River (Highland Ditch)	04/18/07	13S 259849//4429145	104	32	72	0	73	0	27
White River (Highland Ditch Diversion)	04/18/07	13S 259853//4429093	103	100	3	5	52	0	43
White River (Seely Ranch)	04/18/07	13S 258680//4431486	104	100	4	0	51	0	49
White River (K-Bar-T Ranch)	04/18/07	13S 258346//4433019	100	100	0	0	54	0	46
White River (Coal Creek Confluence)	04/18/07	13S 257354//4435197	100	100	0	1	10	0	89
White River (Franklin Ranch Bridge)	04/18/07	13S 254003//4435805	106	100	6	0	51	0	49
White River, N. Fk. (At Trappers Lake Lodge)	12/04/07	13T 309387//4430494	194	100	94	--	--	--	--

nd[a] **:** Haired or non-haired traits were not determined. [a]**:** nw indicates no worms were visible in the sediment sample

Table 17. Stream-specific presence/absence and approximate numbers of cohorts of Rio Grande cutthroat trout (*Oncorhynchus clarki virginalis)* in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2003 Sampling Sites						
Bear Creek	0.1 km upstream of Rio Grande confluence	08/11	Yes	5	Yes	None
Little Squaw Creek	Just upstream of Rio Grande confluence	08/12	Yes	1	No	RBT; BKT;LOC
Lost Trail Creek	1 km upstream of West Lost Trail Creek confluence	08/10	Yes	5	Yes	BKT
Lost Trail Creek	@ USFS Lost Trail Creek Campground	08/10	Yes	4	Yes	BKT;LOC
Middle Ute Creek	10 km upstream of Rio Grande Reservoir	08/14	No	None	None	BKT
Pole Creek	0.1 km upstream of Stoney Pass Road Crossing	08/11	Yes	6	Yes	None
Rio Grande	In Brewster Park @ 10 km upstream of Lost Trail Creek	08/12	Yes	2	No	RBT,LOC
Rio Grande	2 km downstream of Quartzite Creek confluence	08/11	Yes	6	Yes	None
Sangre de Cristo Creek	0.5 km downstream of Placer Creek confluence	09/25	Yes	1	No	BKT
Squaw Creek	@ confluence with Rio Grande	08/12	Yes	1	No	RBT,LOC
Ute Creek	@ confluence with West Fork Ute Creek	08/13	Yes	3	Yes	BKT
Ute Creek	0.5 km upstream of Rio Grande Reservoir high waterline	08/15	No	none	none	RBT,BKT
Weminuche Creek	3 km upstream of Rio Grande Reservoir	08/09	No	none	none	BKT
Weminuche Creek	1 km north of Continental Divide	08/09	No	none	none	BKT
West Lost Trail Creek	@ east end of 1991 landslide	08/10	Yes	5	Yes	BKT
2004 Sampling Sites						
Carnero Creek, M. Fk.	Upstream site near headwaters	07/20	Yes	3	Yes	None
Carnero Creek, N. Fk.	Two sites, one upstream headwaters, 1 downstream	07/20	No	none	none	None
Carnero Creek, S. Fk.	2 km upstream from Middle Fork confluence	07/20	Yes	5	Yes	LOC
Cascade Creek	0.2 km upstream of Cumbres/Toltec RR Trestle	09/22	Yes	3	Yes	BKT,LOC
Conejos River	Below Platoro near Robinson Gulch	09/20	No	none	none	LOC
Conejos R., Lake Fork	above Big Lake; between Rock & Big lakes; ↓ Rock Lake	08/07	No	none	none	BKT,LOC
Conejos R., M. Fork	0.5 km upstream of North Fork Conejos River	09/20	No	None	None	BKT,LOC
Conejos R., N. Fork	0.5 km upstream of Middle Fork Conejos River	09/20	Yes	3	Yes	BKT,LOC
Conejos R., S. Fork	0.1 km upstream of Hansen Creek confluence	09/23	No	None	None	BKT
Ford Creek	Upstream of Tuttle Creek confluence	07/22	No	none	none	BKT
Hansen Creek	0.1 km upstream of confluence w/S. Fork of the Conejos	09/23	No	none	none	LOC
Little Squaw Creek	4 km upstream of Rio Grande confluence	08/09	Yes	5	Yes	None
Osier Creek	3 km upstream of Cumbres/Toltec RR Trestle Bridge	09/22	Yes	4	Yes	None

Table 17 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Rio Grande cutthroat trout (*Oncorhynchus clarki virginalis)* in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2004 Sampling Sites (continued)						
Rio de Los Pinos	Above barrier falls upstream of Trujillo Meadows Rsrvr.	9/21	Yes	5	Yes	None
Rio de Los Pinos	below barrier falls upstream of Trujillo Meadows Rsrvr.	9/21	No	none	None	BKT, LOC
Rio de Los Pinos	0.2 km below Trujillo Meadows Reservoir	09/21	No	none	None	BKT,LOC
Rio de Los Pinos	At Cascade Creek confluence	09/22	No	none	None	LOC
Rio Grande	Below Quartzite Creek confluence	08/10	Yes	5	Yes	None
Tuttle Creek	Above Ford Creek confluence	07/21	Yes	1	No	BKT
2005 Sampling Sites						
Alamasito Creek	0.4 km upstream of N-S Ranch Road	07/25	Yes	4	Yes	None
Alder Creek	Lower reach near South Fork, CO	08/02	No	none	None	LOC
West Alder Creek	Near confluence of East Fork of West Alder Creek	08/02	Yes	2	None	BKT
Cross Creek	Upper station on Peterson Ranch	08/01	Yes	5	Yes	None
Cuatas Creek	Upper site on Cielo Vista Ranch	07/25	Yes	3	Yes	None
Cuatas Creek	Lower site on Cielo Vista Ranch	07/25	Yes	4	Yes	None
Jacks Creek	On SLB Ranch property leased by Sutherland's	08/01	Yes	3	No	None
Jaroso Creek	Upper site on Cielo Vista Ranch	07/25	Yes	5	Yes	None
Jaroso Creek	Lower site on Cielo Vista Ranch	07/25	Yes	5	Yes	BKT
Medano Creek	@ road crossing below Frenchman's Cabin	08/02	Yes	6	Yes	None
Middle Creek	@ Rio Grande NF trailhead	08/03	No	none	none	RBT,BKT,LOC
East Middle Creek	Headwaters, Saguache Creek drainage	08/03	Yes	4	Yes	None
East Pass Creek	2 km below Rio Grande NF Buffalo Creek Campground	08/01	Yes	1	None	None
East Pass Creek	4 km above Rio Grande NF Buffalo Creek Campground	08/01	Yes	3	Yes	None
East Pass Creek	5 km above Rio Grande NF Buffalo Creek Campground	08/01	Yes	3	Yes	None
Placer Creek	Below failed barrier	07/27	Yes	2	No	BKT
Placer Creek	Above failed barrier	07/27	Yes	5	Yes	BKT
Sand Creek	Lower reaches near Liberty, CO @ decadent fishponds	08/03	Yes	1	No	RBT,BKT
San Francisco Creek	On Cielo Vista Ranch	07/26	No	none	None	LOC
San Francisco Creek	South of Del Norte, CO	07/28	Yes	4	Yes	none
Torcido Creek	Lower site on Cielo Vista Ranch	07/25	Yes	4	Yes	None
Torcido Creek	Upper site on Cielo Vista Ranch	07/26	Yes	5	Yes	None

Table 17 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Rio Grande cutthroat trout (*Oncorhynchus clarki virginalis)* in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2005 Sampling Sites (continued)						
North Vallejos Creek	Lower site on Cielo Vista Ranch	07/27	Yes	1	None	LOC
North Vallejos Creek	Upper site on Cielo Vista Ranch	07/27	Yes	4	Yes	LOC
Vallejos Creek	Upper site, Cielo Vista Ranch; 3.5 km ↑road crossing	07/26	Yes	4	Yes	LOC
Vallejos Creek	Lower site, Cielo Vista Ranch; 3.5 km ↑road crossing	07/26	No	none	none	LOC
Wagon Creek	Upstream of confluence with Sange de Cristo Creek	08/02	Yes	3	Yes	BKT
2006 Sampling Sites						
Alberta Creek	Below Alberta Park Reservoir	07/14	No	none	none	BKT
Big Spring Creek	North Clear Creek basin, above Mesa Creek confluence	08/14	No	none	none	BKT
Culebra Creek, N. Frk	Downstream site, near confluence w/S. Fk Culebra Creek	08/17	Yes	4	none	BKT,LOC
Culebra Creek, N. Frk	Upstream site, above confluence w/S. Fk Culebra Creek	08/17	No	none	none	BKT,LOC
West Indian Creek	Trinchera Ranch	08/16	Yes	6	Yes	BKT
Pass Creek	East of Big Meadows Reservoir	07/14	No	none	none	BKT
Trinchera Creek	On the Trinchera Ranch	08/15	Yes	1-2	none	BKT
N. Fk Trinchera Creek	On the Trinchera Ranch	08/16	Yes	1	none	BKT
S. Fk.Trinchera Creek	On the Trinchera Ranch	08/15	Yes	1-2	none	BKT
Placer Creek	Sangre de Cristo Creek to 1st road crossing	07/25	Yes	1-2	None	BKT
Placer Creek	1 km below Greyback Creek confluence	07/25	Yes	1-2	None	BKT
Placer Creek	0.8 km below Middle Fork confluence	07/24	No	None	None	BKT
Placer Creek	0.5 km above Middle Fork confluence	07/26	No	None	None	BKT
Placer Creek	3 km above Middle Fork confluence	07/27	Yes	1	None	BKT
Placer Creek	5 km above Middle Fork confluence	07/27	Yes	3	Yes	BKT
M. Fork Placer Creek	0.8 km below S. Fork Placer Creek confluence	07/25	No	None	None	BKT
M. Fork Placer Creek	0.5 km above S. Fork Placer Creek confluence	07/26	No	none	None	BKT
M. Fork Placer Creek	5 km above S. Fork Placer Creek confluence	07/27	Yes	3	Yes	BKT

Table 17 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Rio Grande cutthroat trout (*Oncorhynchus clarki virginalis)* in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2007 Sampling Sites						
Cumbres Creek	1 km upstream of Rio de los Pinos River confluence	7/25	No	None	None	BKT;LOC
Rio Chama	100 meters below Cumbres & Toltec RR Bridge	7/25	No	None	None	LOC
Wolf Creek	100 meters below Cumbres & Toltec RR Bridge	7/25	No	None	None	LOC
Wolf Creek	Beaver ponds at Cumbres & Toltec RR Bridge culvert	7/25	Yes	4	Yes	LOC
Wolf Creek	↑beaver dam upstream of Cumbres & Toltec RR Bridge	7/25	Yes	2-3	Yes	None

Table 18. Stream-specific presence/absence and approximate numbers of cohorts of Greenback (*Oncorhynchus clarki stomias)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2003 Sampling Sites						
Apache Creek, North	Upstream site @ Bartlett Trail crossing	07/29	Yes	1-2	none	None
Apache Creek, North	Downstream site 0.5 km↑ of South Apache Creek confluence	07/29	No	None	None	None
Apache Creek, South	Downstream site 0.5 km↑ of North Apache Creek confluence	07/29	No	none	none	None
Greenhorn Creek	1.6 km ↑ San Isabel NF boundary near Rye, CO	7/29	No	None	None	RBT; BKT;LOC
Hayden Creek	@USFS San Isabel NF Coadale Campground	7/25	No	none	none	BKT;LOC
Hayden Crk., S. Prong	Upstream site San Isabel NF	7/25	Yes	1 fish	none	None
Hayden Crk., S. Prong	Downstream site San Isabel NF	7/25	No	none	none	LOC
Huerfano River	@ CDOW Huerfano SWA, downstream site	7/30	No	none	none	BKT;LOC
Huerfano River	Upstream site, 3 km below NF Lily Lake trailhead	8/1	No	none	none	RBT; BKT;LOC
S. Fork Huerfano R.	@ High Mesa Ranch downstream of Cascade Creek	7/30	No	none	none	BKT;LOC
Newlin Creek	@ Florence Mountain Park, NF Service boundary	7/28	Yes	5	Yes	None
South Arkansas River	South Fork @ USFS Monarch Park Campground	7/22	No	none	none	BKT
South Arkansas River	Middle Fork	7/22	Yes	2	none	None
St. Charles River	0.8 km upstream of San Isabel Lake	7/29	No	none	none	RBT; BKT;LOC
St. Charles River	0.1 km downstream of San Isabel Lake	7/31	No	none	none	RBT; BKT;LOC
2004 Sampling Sites						
Dry Gulch	1 km ↑Clear S. Crk confluence, 2 km east of Eisenhower Tunnel	7/28	Yes	5	Yes	None
Middle Fk. S. Platte R	Upstream of Montgomery Reservoir	7/27	Yes	6	Yes	None
South Clear Creek	@ I-70 Exit 218 2 km east of Eisenhower Tunnel	7/28	Yes	4	none	None

Table 18 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Greenback (*Oncorhynchus clarki stomias)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2005 Sampling Sites						
Bennett Creek	Upstream of Little South Fork Cache la Poudre R. confluence	9/28	no	none	none	RBT
Black Hollow Creek	Upstream sampling site	9/28	no	none	None	None
Black Hollow Creek	Lower reaches near confluence w/Poudre River	9/28	no	none	none	RBT; BKT;LOC
Cornelius Creek	Upstream reaches above George Creek confluence	9/28	no	none	none	BKT
George Creek	Upstream reaches above George Creek confluence	9/28	yes	2	yes	BKT
Herman Gulch	2 km ↑Clear S. Crk confluence, 2 km east of Eisenhower Tunnel	9/26	yes	7	yes	None
Pennock Creek	Upstream of Little South Fork Cache la Poudre River	9/28	No	none	none	BKT;LOC
Sheep Creek	Upstream of Eaton reservoir, N. Fork Cache la Poudre River	9/30	No	none	none	BKT
Sheep Creek	Downstream of Eaton reservoir, N. Fork Cache la Poudre River	9/30	No	none	none	BKT;LOC
Sheep Creek	Near confluence with North Fork Cache la Poudre River	9/30	No	none	none	LOC
Sheep Creek, West Fk.	Above confluence with East Fork Cache la Poudre River	9/28	Yes	1	none	None
Sheep Creek, East Fk.	Above confluence with West Fork Cache la Poudre River	9/28	Yes	1	none	None
2007 Sampling Sites						
S. Cottonwood Creek	Approximately 2 km upstream of Cottonwood Lake	6/28	No	None	None	BKT;LOC
S. Cottonwood Creek	Approximately 10 km upstream of Cottonwood Lake	6/28	Yes	4	Yes	None

Table 19. Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2003 Sampling Sites - Fryingpan River Basin						
Chapman Gulch	Above Fry/Ark Project Hunter Creek Diversion Tunnel	10/3	Yes	1	None	BKT
Cunningham Creek	1 km above USGS gage station	10/6	No	none	None	BKT;LOC
Cunningham Creek	1 km above confluence w/Middle & S. Fk. Cunningham creeks	10/6	Yes	5	Yes	None
Deeds Creek	1 km above Foster Gulch junction	10/3	No	none	None	BKT
Fryingpan River	At Nash Bridge	6/25	No	none	None	LOC
Fryingpan River	Below Chapman Dam outflow	6/25	No	none	None	LOC
Fryingpan River	Above Marten Creek @ Fry/Ark Diversion Tunnel	10/8	No	none	None	BKT
Fryingpan River, N. Fork	@ Bigelow	6/25	No	none	None	LOC
Fryingpan River, N. Fork	@ USFS White River NF Elk Wallows Campground	6/25	No	none	None	BKT;LOC
Fryingpan River, N. Fork	1 km downstream of Mormon Creek confluence	10/7	No	none	None	BKT;LOC
Fryingpan River, S. Fork	10 meters upstream of Fry/Ark Diversion Tunnel	10/3	Yes	5	Yes	None
Ivanhoe Creek	2 km below Ivanhoe Lake outlet	10/8	No	none	None	BKT
Last Chance Creek	↑ North Fork of the Fryingpan River @ Holy Cross Wilderness	6/25	No	none	None	BKT
Lime Creek	1 km upstream of Woods Lake	10/9	No	none	None	BKT
Lime Creek	3 km downstream of Woods Lake	10/9	No	none	None	BKT;LOC
Little Lime Creek	3 km downstream of Crooked Creek Reservoir	10/9	No	none	None	BKT;LOC
Marten Creek	Above Fryingpan River confluence @ Fry/Ark Diversion Tunnel	10/8	No	none	None	BKT
Rocky Fork Creek	2 km upstream of Fryingpan River confluence @ Ruedi Dam	10/7	Yes	4	Yes	none

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2003 Sampling Sites - Upper Colorado River Basin – Grand County						
Bobtail Creek	Upstream of trail crossing (Williams Fork River basin)	9/3	Yes	5	Yes	BKT
Cabin Creek	In Devils Thumb Park east of Fraser, CO; upstream site	9/19	Yes	4	Yes	None
Cabin Creek	downstream site near USFS Road 128 crossing	9/10	Yes	3	Yes	None
Fraser River	@ USFS Robbers Roost Campground	9/10	Yes	1	No	BKT
Kinney Creek	Tributary to the Colorado River near Sheriff Ranch; upper site	9/10	Yes	2	No	None
Kinney Creek	Tributary to the Colorado River near Sheriff Ranch; lower site	9/10	No	none	none	BKT
Hamilton Creek, upstream	Above High Lonesome Trail crossing east of Tabernash, CO	9/18	Yes	5	Yes	None
Hamilton Crk, downstream	Above Denver Water Department weir	9/18	Yes	3	none	None
Jim Creek	Tributary to the Fraser River south of Fraser, CO	9/10	No	none	none	BKT
McQuery Creek	Above Denver Water Department Diversion	9/16	Yes	4	Yes	None
South Fork Ranch Creek	Above Denver Water Department Diversion	9/11	Yes	3	Yes	BKT
Steelman Creek	Upstream site above Denver Water Department Diversion	9/16	Yes	5	Yes	BKT
Steelman Creek	Middle station above Denver Water Department Diversion	9/16	Yes	5	Yes	BKT
St. Louis Creek	Downstream sampling site	9/17	No	none	none	BKT
St. Louis Creek	upstream sampling site	9/17	No	none	none	BKT
S. Fork Williams Fork R.	At road crossing	9/15	No	none	none	BKT
Williams Fork River	Near the Ute Creek confluence	9/15	No	none	none	RBT;BKT;LOC
2004 Sampling Sites - Colorado River Basin – Battlement Mesa and Grand Mesa areas						
Battlement Creek	Upstream site	10/7	Yes	5	Yes	None
Battlement Creek	Downstream site	10/5	Yes	5	Yes	None
Beaver Creek	Downstream reach on north side of Battlement Mesa	8/18	Yes	3-4	Yes	None
Big Creek	200 meters upstream of Bonham Reservoir	9/9	Yes	2	None	BKT;LOC
Big Creek	100 meters downstream of Big Creek Reservoir	10/4	Yes	3-4	None	None
Big Creek	100 meters upstream of Big Creek Reservoir	10/4	Yes	2	Yes	None
Big Creek, West Fork	Approximately 1 km upstream of Bonham Reservoir	9/9	Yes	2	Yes	None

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2004 Colorado River Basin Sampling Sites – Roan Plateau, Douglas and Piceance creeks, Eagle, Fryingpan and Gunnison rivers drainage basin areas						
South Beaver Creek	Upstream of confluence with Deer Beaver Creek	6/24	Yes	3	Yes	None
Big Fish Creek	6 km upstream of West of the Fork Dolores River	8/26	No	None	None	None
Black Gore Creek	Downstream of Black Lakes near top of Vail Pass	7/27	Yes	2	Yes	BKT
Black Sulphur Creek	Upper station	10/25	Yes	2	None	None
Black Sulphur Creek	Middle reach sampling station	10/25	Yes	1	None	None
Brush Creek	Upstream site on the Roan Plateau	8/19	No	none	None	RBT
Canyon Creek	400 meters upstream of Black Sulphur Creek confluence	10/25	Yes	2	Yes	None
Carr Creek	Upstream site	8/23	Yes	3-4	Yes	None
Carr Creek	Downstream site	8/23	Yes	1	None	BKT
Cochetopa Creek	1 km upstream of Canyon Diablo Creek, above barrier falls	6/28	Yes	7	Yes	None
Cochetopa Creek	2 km downstream of Canyon Diablo Crk, below barrier falls	6/28	Yes	2	None	BKT;LOC
Cochetopa Creek, Lake Fork	1 km upstream of Saguache Creek transmountain diversion	6/27	Yes	7	Yes	None
Crooked Creek	Fryingpan River basin 2 km upstream Crooked Creek Rsrvoir.	10/5	No	none	None	BKT
Deer Beaver Creek	Upstream of South Beaver Creek, Gunnison River basin	6/24	Yes	2-3	None	None
East Douglas Creek	Downstream site below Brush Creek confluence	9/14	Yes[a]	2	None	RBT
East Douglas Creek	Upstream site above Brush Creek confluence	9/14	Yes	2	None	None
Fall Creek	Big Blue Creek basin, ↑ barrier falls, Big Blue Wilderness	6/21	Yes	7	Yes	None
East Parachute Creek	2 km above barrier falls on Roan Plateau	9/16	Yes	3	Yes	BKT
East Parachute Creek	Below barrier falls on Roan Plateau	10/6	No	none	None	None
Left Fork Lake Creek	Above Cathedral Creek confluence, Douglas Creek basin	9/13	Yes	2	Yes	None
Northwater Creek	Above Trapper Creek confluence on Roan Plateau	9/15	Yes	5-6	Yes	None
East M. Fk. E. Parachute Creek	Above barrier falls on Roan Plateau	9/15	Yes	4-5	Yes	None
East M. Fk. E. Parachute Creek	Below barrier falls on Roan Plateau	10/6	Yes	3-4	None	None
Roan Creek	Upstream site on Roan Plateau	8/24	Yes	3	Yes	None
Roan Creek	Downstream site on Roan Plateau	8/24	Yes	2	none	BKT
Soldier Creek	Above Cathedral Creek confluence, Douglas Creek basin	9/13	Yes	5	none	None
Trapper Creek	Above Northwater Creek confluence on Roan Plateau	9/15	Yes	3	none	None

[a]**:** These fish were rainbow cutthroat hybrids and/or rainbow trout; not pure cutthroat trout.

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2005 Sampling Sites - Colorado River Basin						
Big Creek, East Fork	Above Big Creek confluence, below Atkinson Reservoir	8/26	Yes	4	Yes	None
Big Creek, West Fork	Approximately 1 km above Bonham Reservoir above falls	9/16	Yes	5	Yes	None
Buzzard Creek	On Grand Mesa	09/15	No	none	none	none
Cow Creek	Upstream of Overland Reservoir, east end Grand Mesa	9/15	No	none	none	RBT;BKT
Hubbard Creek	Near Overland Reservoir, east end of Grand Mesa	10/25	No	None	None	BKT
Middle Hubbard Creek	Near Overland Reservoir, east end of Grand Mesa	10/25	No	none	None	BKT
Plateau Creek	Upstream of Vega Reservoir	9/16	No	none	none	RBT
Plateau Creek	Downstream of Vega Reservoir	9/16	No	none	None	BKT;LOC
Second Creek	Upstream of barrier to fish migration	10/24	Yes	4	Yes	None
Second Creek	Downstream of barrier to fish migration	10/24	No	none	None	RBT
Smith Fork, Gunnison R.	Upstream of Second Creek confluence	10/24	No	none	none	RBT;BKT;LOC
Unnamed Tributary	Tributary to Bonham Reservoir, west arm	10/20	No	none	none	BKT
2006 Colorado River Basin Sampling Sites - Dolores, Gunnison and Uncompahgre River sub-basins						
Dolores River	@ USFS Barlow Creek Campground	8/10	No	none	none	RBT;LOC
Scotch Creek	Dolores River Basin, above confluence with Dolores River	8/10	No	none	none	BKT;LOC
West Fork Cebolla Creek	Headwaters of Gunnison River Basin	8/14	No	None	none	BKT
Beaver Dams Creek	Uncompahgre Plateau, WSW of Montrose, CO	6/13	Yes	3	Yes	none
East Fork Dry Creek	Uncompahgre Plateau, WSW of Montrose, CO	6/13	No	none	none	RBT;LOC

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2006 Colorado River Basin Sampling Sites - San Juan River Drainage Basin						
Castle Creek	Rio Blanco River drainage	9/19	No	none	none	none
Corral Creek	Headwaters of Hermosa Creek drainage	7/10	Yes	3	Yes	RBT;BKT
Fish Creek	@ Fish Creek Trailhead, Rio Blanco River basin	9/19	No	none	none	BKT
Florida River	100 meters below Lemon Reservoir outlet	7/10	No	none	none	RBT
Hermosa Creek	Near Hermosa, CO, upstream of Animas River confluence	6/26	No	none	none	RBT;BKT
East Fork Hermosa Creek	Near Purgatory Ski Area	7/10	Yes	4	Yes	None
Piedra River	2 km below confluence of East & Middle Forks Piedra River	9/20	No	None	none	RBT;LOC
Piedra River	2 km upstream of U.S. Hwy 160 Piedra River Bridge	9/21	No	None	none	RBT;LOC
East Fork Piedra River	Upstream of confluence with Middle Fork Piedra River	7/11	No	None	None	RBT;BKT;LOC
Middle Fork Piedra River	Upstream of confluence with East Fork Piedra River	7/11	No	None	None	BKT;LOC
Rito Blanco River	@ San Jaun NF Road 024 culvert crossing	9/20	Yes	5	Yes	None
East Fork San Juan River	Headwaters above barrier waterfall	7/13	Yes	3-4	none	RBT
East Fork San Juan River	Below Sand Creek confluence	7/13	No	none	none	RBT;LOC
West Fork San Juan River	Upstream of confluence with East Fork San Juan River	7/13	No	none	none	RBT;LOC
South Branch South Creek	Headwaters, Rio Blanco River basin	9/19	No	none	none	BKT
White Creek	Headwaters, Rio Blanco River basin	9/19	No	none	none	BKT
Weminuche Creek	Headwaters	7/12	No	none	none	RBT;BKT;LOC
East Fork Weminuche Crk.	Upstream of confluence with Weminuche Creek	7/12	No	none	none	BKT;LOC
Williams Creek	1 km upstream of Williams Creek Reservoir	7/12	No	none	none	BKT
Williams Creek	@ San Juan NF Bridge Campground below Williams Creek Rsvr.	9/20	No	none	none	LOC
2007 Colorado River Basin Sampling Sites - San Juan River Drainage Basin						
Cunningham Creek	Above confluence with Animas River upstream of Silverton, CO	10/23	Yes	5-6	Yes	None
Navajo River	Upstream site on Banded Peak Ranch	7/24	Yes	5	Yes	None
Navajo River	Downstream site on Banded Peak Ranch	7/24	No	none	none	BKT;LOC
Rio Blanco River	Downstream site 2 km above Hwy 84 Bridge	7/24	No	none	none	RBT;LOC
Rio Blanco River	Middle station near Castle Creek Bridge	7/23	No	none	none	RBT;BKT;LOC
Rio Blanco River	Upstream site	7/23	No	none	none	BKT

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2006 Colorado River Basin Sampling Sites – White River Drainage Basin						
Big Beaver Creek	Above Lake Avery below Allen Creek confluence	9/6	Yes	4	Yes	None
Big Beaver Creek	200 m above Lake Avery	9/6	No	none	none	RBT;BKT
Cabin Creek	Inlet to Trappers Lake	10/9	Yes	1	Yes	BKT
Cabin Creek	Outlet of Little Trappers Lake	10/9	Yes	2	Yes	BKT
Fawn Creek	Lower station at Moeller Creek confluence	9/7	Yes	7-8	Yes	None
Fawn Creek	Upper station below confluence of East & West Forks Fawn Creek	9/7	Yes	6	Yes	None
Fraser Creek	Approximately 200 meters upstream of Big Trappers Lake inlet	10/6	Yes	2	Yes	BKT
Heberton Creek	Approximately 200 meters upstream of Big Trappers Lake inlet	10/6	Yes	3	Yes	BKT
Little Trappers Lake	Collection from within Little Trappers Lake	10/6	Yes	6	Yes	BKT
Lost Creek	Lower station approximately 2 km ↑ White River confluence	9/7	Yes	5	none	RBT
Lost Creek	Upper Station	9/7	Yes	3	none	None
Marvine Creek	@ USFS Marvine Creek Trailhead into Flattops Wilderness	9/7	No	none	None	RBT;BKT
North Elk Creek	@ USFS boundary adjacent to private land in-holdings	10/11	Yes	6[b]	Yes[b]	Rbt;Bkt:Loc;RXN
Middle Fk North Elk Creek	Upstream of East Fork of North Elk Creek confluence	10/11	Yes[c]	4	Yes[c]	BKT
Snell Creek	Lower station – 100 meters above White River road culvert	9/6	Yes	3	None	BKT
Snell Creek	Upper station - 3 km upstream of White R. confluence	9/6	Yes	5	Yes	BKT
Ute Creek	Lower station approximately 1.5 km ↑ White River confluence	9/5	No	none	none	BKT
Ute Creek	Upper station approximately 5 km ↑ White River confluence	9/5	No	none	none	BKT
White River	Upstream of County Road 10 (South Fork Road) Bridge	10/19	No	none	none	RBT;LOC;MWF
White River	Downstream of County Road 54 (Sleep Cat SWA) Bridge	10/19	No	none	none	RBT;LOC;MWF
White River	Near Wakara Ranch Bridge east of Meeker, CO	11/9	No	none	none	LOC
White River	Near Franklin Ranch Bridge east of Meeker, CO	11/9	No	none	none	LOC
North Fork White River	@ USFS Himes Peak Campground	10/24	Yes	3	Yes	RBT;BKT
North Fork White River	Downstream of Snell Creek confluence	10/24	Yes	3	Yes	RBT
North Fork White River	Near USFS North Fork Campground	10/24	No	none	none	RBT;BKT
South Fork White River	@ USFS Flattops Wilderness Boundary Trailhead	10/24	Yes	4	Yes	RBT;BKT

[b]**:** At this site there were numerous YOY fry that appeared to be rainbow trout X CRN trout hybrids. Most appeared to be F1 crosses but some of the fish ≥ age 1 and older were F2 back-crosses of various combinations.

[c]**:** There was no **overt** evidence of rainbow trout introgression/reproduction at this site on this sampling date**.**

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2006 Colorado River Basin Sampling Sites – Yampa River Drainage Basin						
Bear River	Headwaters, station 1 above meadows; 3 km↑ Stillwater Reservoir	8/23	No	None	None	BKT
Bear River	station 2, ↓ meadow beaver ponds; 1.5 km↑ Stillwater Reservoir	8/23	No	None	None	BKT
Bear River	station 3, below high gradient to meadow; 1 km ↑ Stillwater Rsrvr	8/23	Yes	3	Yes	BKT
Bear River	Station 4 below Yamcolo Reservoir	8/23	No	None	None	RBT;BKT;LOC
Circle Creek	Near confluence with Elkhead Creek	10/4	Yes	2	None	BKT
East Coal Creek	(Bear River drainage) station 1	8/21	Yes	2	None	RBT;BKT
East Coal Creek	(Bear River drainage) station 2	8/22	No	None	None	BKT
East Coal Creek	(Bear River drainage) station 3	8/22	Yes	1	None	BKT
West Coal Creek	(Bear River drainage) station 1	8/21	Yes	2	Yes	None
West Coal Creek	(Bear River drainage) station 3	8/21	Yes	5-6	Yes	None
West Coal Creek	(Bear River drainage) station 4	8/22	Yes	6	Yes	None
Elkhead Creek	Upstream of Circle Creek confluence	10/4	Yes	4	None	BKT
Green Creek	On Green Creek Ranch	8/24	No	None	None	BKT
Green Creek	Lower end of upper meadow	8/24	No	None	None	BKT
Little Cottonwood Creek	0.5 km upstream of inlet to Freeman Reservoir	10/12	Yes	2	Yes	None
Poose Creek	0.5 km downstream of Vaughn Lake Dam	10/2	Yes	3	Yes	None
Sand Creek	1 km upstream of inlet to Sheriff Reservoir	10/3	No	None	None	BKT
Slater Creek	Little Snake River basin	10/4	No	None	None	BKT
Trout Creek	1 km upstream of Sheriff Reservoir	10/3	Yes	2	None	BKT
Trout Creek	0.5 km downstream of Sheriff Reservoir	10/3	No	None	None	BKT

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2007 Colorado River Basin Sampling Sites – Blue River, Plateau Creek and upper Colorado River sub-basins						
Big Creek	Middle Fork @ weir above Bonham Reservoir	8/28	Yes	2-3	None	RBT;BKT;LOC
West Fork Big Creek	Approximately 1 km upstream of Middle Fork confluence	8/28	Yes	4	Yes	None
Clinton Creek	Upstream of Clinton Reservoir	8/15	Yes	4	Yes	None
Piney River	4-5 km upstream of Piney Lake in Eagles Nest Wilderness	9/10	Yes	2	Yes	BKT;LOC
Piney River	1 km downstream of Piney Lake	8/10	No	None	None	BKT;LOC
Piney River	Middle station; Piney Valley Ranch @USGS gage)	8/10	No	None	None	RBT;LOC
Piney River	@ Colorado River confluence above State Bridge (CO Hwy 131)	8/8	No	None	None	RBT;LOC
Rock Creek	Tributary to the Colorado River NNE of McCoy, CO	8/8	No	None	None	RBT;LOC
Little Rock Creek	North of CO Hwy 134 (Toponas to Kremmling road)	8/15	No	None	None	BKT;LOC
Sheephorn Creek	2 km downstream of Trough Road Bridge crossing-Radium SWA	8/8	No	None	None	RBT;LOC
Swan River, North Fork	Approx. 5 km upstream of Swan River near Rexford, CO	8/9	Yes	2-3	None	None
Swan River, South Fork	Near Nuggetville, ↑confluence with Middle Fork Swan River	8/9	No	None	None	BKT
2007 Colorado River Basin Sampling Sites – Dolores River Drainage Basin						
Buckeye Creek	1 km upstream of Buckeye Reservoir @ USFS road crossing	9/12	No	None	None	RBT;LOC
Deep Creek	Tributary to San Miguel River near Telluride	9/11	No	None	None	LOC
Deep Creek, East Fork #1	Tributary to San Miguel River near Telluride	9/11	No	None	None	None
Deep Creek, East Fork #2	Tributary to San Miguel River near Telluride	9/11	No	None	None	None
Blue Creek	Upstream site – 2 km downstream Dark Canyon off 19.50 Road	9/13	No	None	None	RBT
Blue Creek	Downstream site - 4 km downstream Dark Canyon off 19.50 Road	9/13	No	None	None	RBT
2007 Colorado River Basin Sampling Sites – Eagle River Drainage Basin						
Boothe Creek	Tributary to Gore Creek near Vail, CO	8/16	Yes	3	Yes	None
Cross Creek	Tributary to Eagle River near Minturn and Gilman, CO	8/16	No	None	None	BKT; LOC
Pitkin Creek	Tributary to Gore Creek near Vail, CO; 2 km up trail	8/16	Yes	4	Yes	None

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2007 Colorado River Basin Sampling Sites – Gunnison River Drainage Basin						
Anthracite Creek	Below outlet of Lake Erwin	7/12	No	None	None	RBT;BKT
Big Cimarron R., M. Fork	Above confluence with West Fork Big Cimarron River	7/19	Yes	4	None	RXN hybrids
Big Cimarron R., West Fk.	↑Silverjack Reservoir, ↑confluence w/Middle Fk. Big Cimarron R	7/19	Yes	4	Yes	None
Little Cimarron River	At Alpine Plateau Trail Bridge crossing	7/19	No	None	None	BKT
Big Dominguez Creek	Headwaters area near campground on 24.40 Road	11/14	No	None	None	RBT
Brush Creek	East River drainage east of Crested Butte, CO	7/10	No	None	None	BKT;LOC
Middle Brush Creek	East River drainage east of Crested Butte, CO	7/10	No	None	None	BKT;LOC
Cebolla Creek	At Mason Family SWA	10/25	No	None	None	LOC
Cebolla Creek	Above Spring Creek confluence	10/25	No	None	None	LOC
Coal Creek	Near Kebler Pass	7/18	No	None	None	BKT
Cow Creek	Tributary to Uncompahgre River east of Ridgway, CO	7/12	Yes	2	Yes	None
East Dallas Creek	At Blue Lakes trailhead	10/23	Yes	6	Yes	BKT
East Dallas Creek	At lower pullout near horse camp	10/23	Yes	3	Yes	BKT
Escalante Creek	Below Escalante Forks on BLM land	11/12	No	None	None	RBT
Escalante Creek	@ Walker Cabin	11/12	No	None	None	None
East Fork Escalante Creek	@ Escalante Creek SWA	11/12	No	None	None	RBT;RXN
Henson Creek	Above Treasure Falls Dam	10/24	No	None	None	BKT
Lake Fork of Gunnison R.	Above Lake San Cristobal	10/24	No	None	None	BKT;LOC
Marshall Creek	Upstream of Tomichi Creek confluence near Sargents, CO	7/9	No	None	None	LOC
Oh-Be-Joyful Creek	Upstream of Slate River confluence	7/11	No	None	None	RBT;BKT
Pass Creek	Above Taylor Park Reservoir west of Cottonwood Pass	6/28	No	None	None	BKT;LOC
Pass Creek	300 m below North Golden Lake outlet – West Elk Wilderness	9/30	No	None	None	CA Golden trout
Rock Creek	East River drainage north of Crested Butte, CO	7/13	No	None	None	BKT
Slate Creek	North of Crested Butte, CO, East River drainage	7/11	No	None	None	RBT;BKT;LOC
Spring Creek	Upstream of Cebolla Creek confluence	10/25	No	None	None	BKT;LOC
Tomichi Creek	Downstream of Sargents, CO on Nature Conservancy Easement	7/9	No	None	None	LOC
Tomichi Creek	Upstream site above Agate Creek confluence	7/9	No	None	None	LOC

Table 19 (continued). Stream-specific presence/absence and approximate numbers of cohorts of Colorado River (*Oncorhynchus clarki pleuriticus)* cutthroat trout in streams samples between 2003 and 2007.

Stream Name	Site Description	Date mm/dd	Cutthroat			Other Salmonids (RBT; BKT;LOC)
			Present	No. Cohorts	YOY < 1 yr	
2007 Colorado River Basin Sampling Sites – Yampa River Drainage Basin						
Beaver Creek	Above confluence with Indian Run Creek @ Indian Run SWA	8/22	Yes	5	Yes	None
Indian Run Creek	Above confluence with Beaver Creek @ Indian Run SWA	8/22	Yes	4	Yes	None
Johnson Creek	4 km above confluence with Little South Fork Snake River	8/21	Yes	3	None	None
N. Fork Elkhead Creek	Lower station, approximately 5 km ↑Elkhead Creek confluence	8/24	Yes	2	Yes	None
N. Fork Elkhead Creek	Middle station approximately 6 km ↑Elkhead Creek confluence	8/24	Yes	3	Yes	None
N. Fork Elkhead Creek	Upper station approximately 6.2 km ↑Elkhead Creek confluence	8/24	Yes	3	Yes	None
Oliver Creek	Above confluence with Little South Fork Snake River	8/21	No	None	None	None
Smith Creek	Lower station	8/23	Yes	2	Yes	None
Smith Creek	Middle station	8/23	Yes	2	Yes	None
Smith Creek	Upper station	8/23	Yes	1	Yes	None
S. Fk. Little Snake River	Lower station, below Oliver Creek confluence @ 3 Forks Ranch	8/21	No	None	None	None
S. Fk. Little Snake River	Middle station, above Oliver Creek confluence @ 3 Forks Ranch	8/21	No	None	None	None
S. Fk. Little Snake River	Upper station (headwaters), upstream of "T" in road	8/21	Yes	2	Yes	None
S. Fk. Wms Fork Yampa R	Lower station, approx. 1 km upstream of Seely Ranch HQ	8/22	Yes	4-5	Yes	None
S. Fk. Wms Fork Yampa R	Upper station, 5.5 km above Seely Ranch HQ at USFS Trail 1113	8/22	Yes	2	Yes	None
2007 Colorado River Basin Sampling Sites – White River Drainage Basin						
Marvine Creek	Between upper and lower Marvine Lakes	8/14	No	None	None	BKT
Marvine Creek	0.5 km downstream of lower Marvine Creek outlet	8/15	No	None	None	BKT

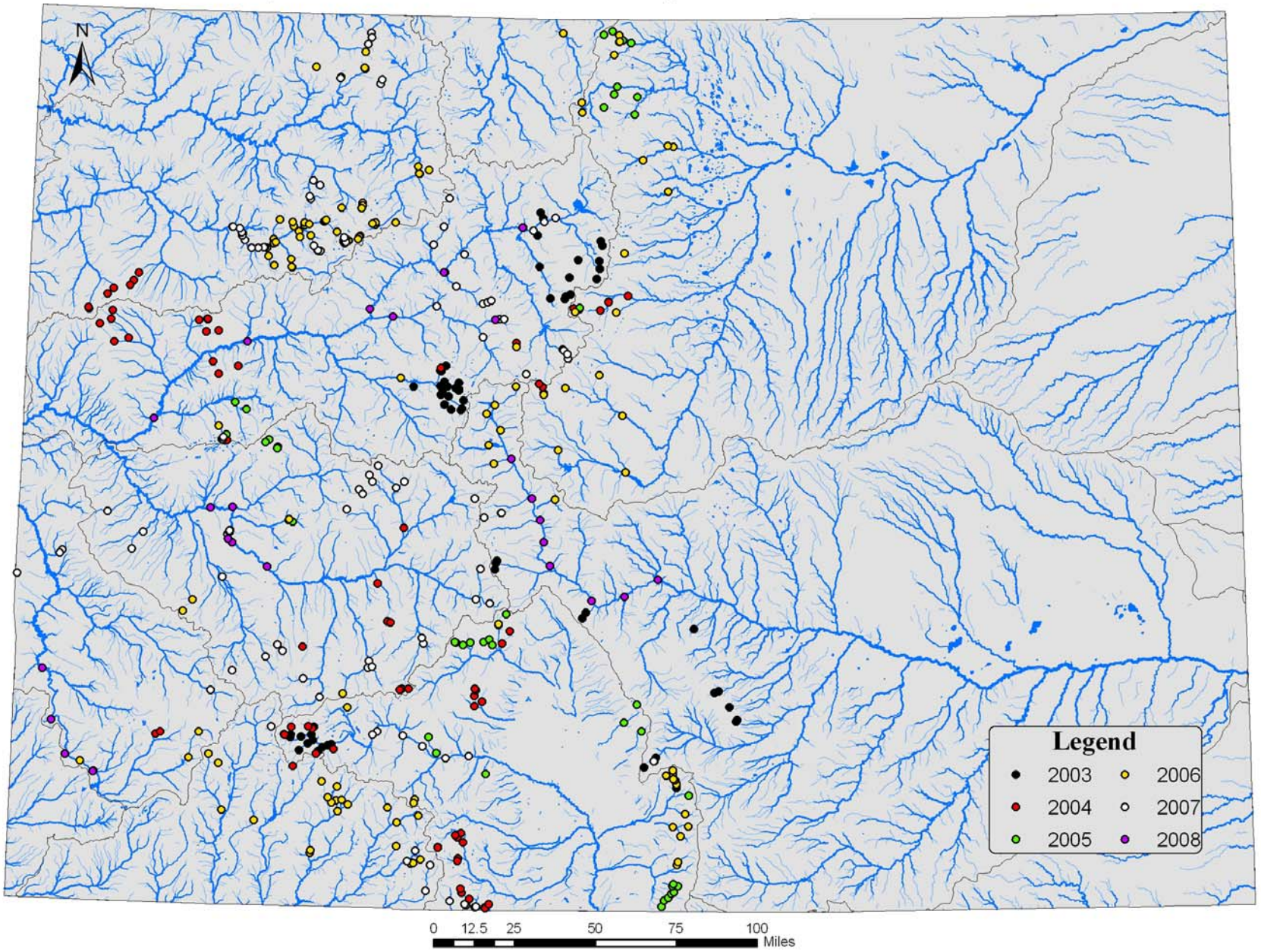
Table 20. Summary of survey results from 2003 through 2007 for presence or absence of cutthroat trout at survey sites, frequency of occurrence of young-of-the-year -YOY (≤ 12 months of age at time of survey) and the approximate numbers of cohorts (based on average size of the YOY cohort and length-frequency distributions of all cutthroat trout collected) present within the survey reach.

Year	Cutthroat Presence/Absence		YOY Presence/Absence		Approximate Numbers of Cohorts Present within Stream Reach Sampled					
	Absent	Present	Present	Absent	1	2	3	4	5	≥ 6
Rio Grande Cutthroat trout (*Oncorhynchus clarki virginalis)*										
2003	4	11	7	4	1	1	1	1	3	2
2004	10	9	8	1	1	0	3	1	4	0
2005	4	23	17	6	3	2	7	7	5	1
2006	8	10	3	7	6	0	2	1	0	1
2007	3	2	2	0	0	0	1	1	0	0
Totals	**29**	**55**	**37**	**18**	**11**	**3**	**14**	**11**	**12**	**4**
Greenback Cutthroat trout (*Oncorhynchus clarki stomias)*										
2003	11	4	1	3	2	1	0	0	1	0
2004	0	3	3	0	0	0	0	1	1	1
2005	8	4	2	2	2	1	0	0	0	1
2006	0	0	0	0	0	0	0	0	0	0
2007	1	1	1	0	0	0	0	1	0	0
Totals	**20**	**12**	**7**	**5**	**4**	**2**	**0**	**2**	**1**	**1**
Colorado River Cutthroat trout (*Oncorhynchus clarki pleuriticus)*										
2003	20	15	11	4	2	1	3	3	6	0
2004	4	30	17	13	2	12	8	1	4	3
2005	9	3	3	0	0	0	0	2	1	0
2006	38	33	24	9	2	7	10	5	4	5
2007	45	26	22	4	1	9	5	7	3	1
Totals	**116**	**107**	**77**	**30**	**7**	**29**	**26**	**18**	**18**	**9**
All Cutthroat Trout Sub-Species Combined										
GRAND TOTALS	**165**	**174**	**121**	**53**	**22**	**34**	**40**	**31**	**31**	**14**

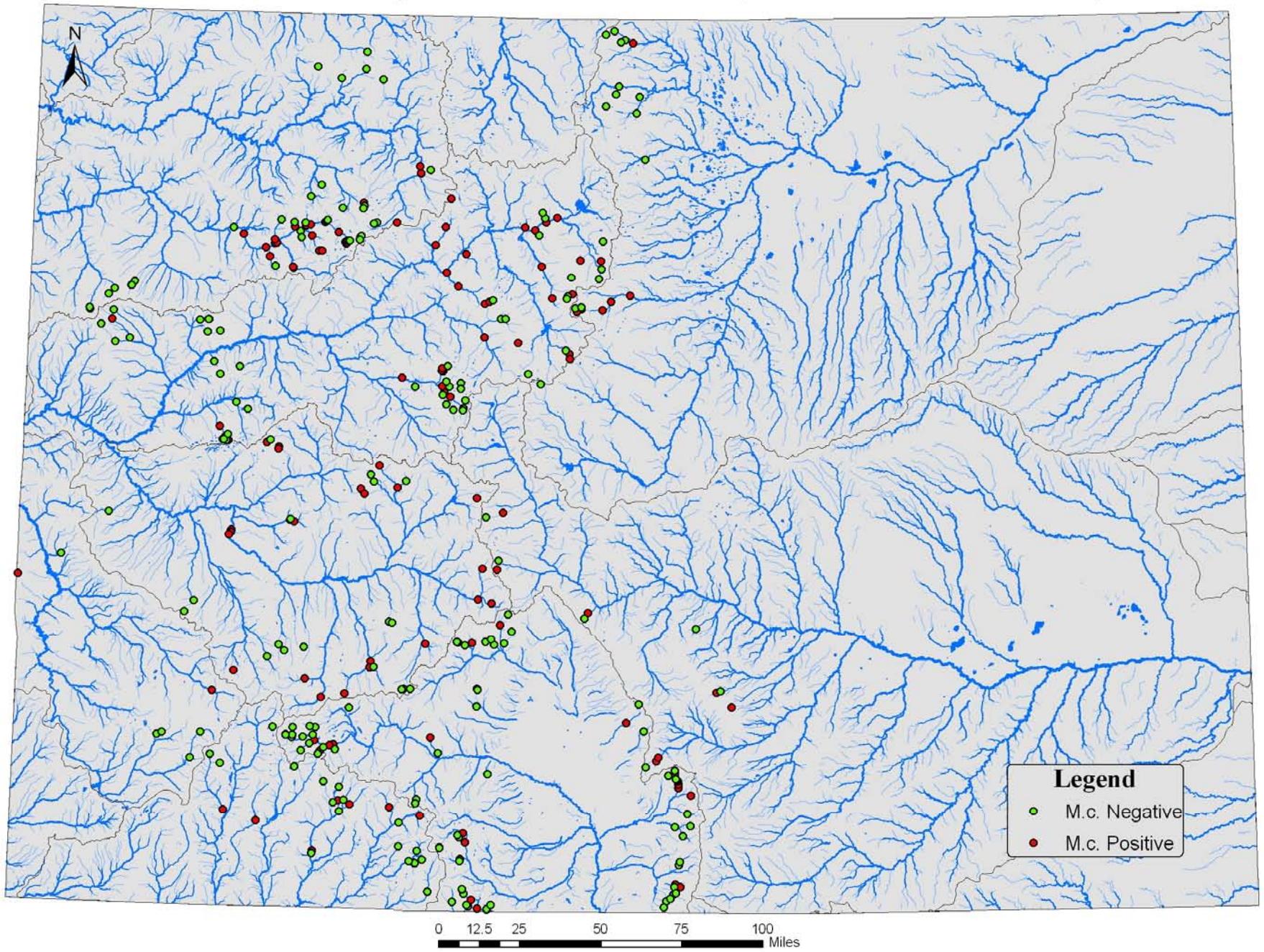
APPENDIX B

MAPS

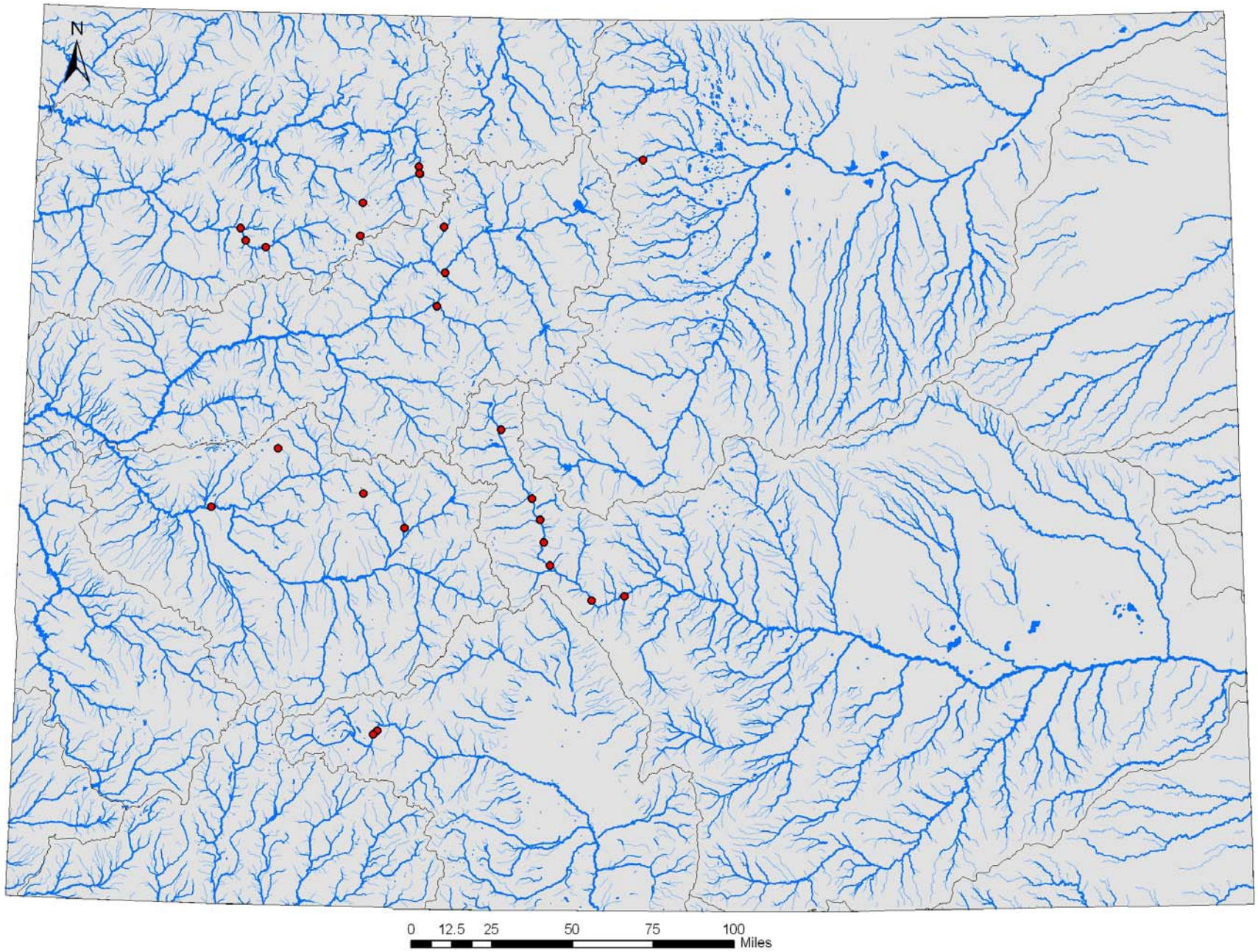
Map 1. Locations of sample sites for 2003-2008



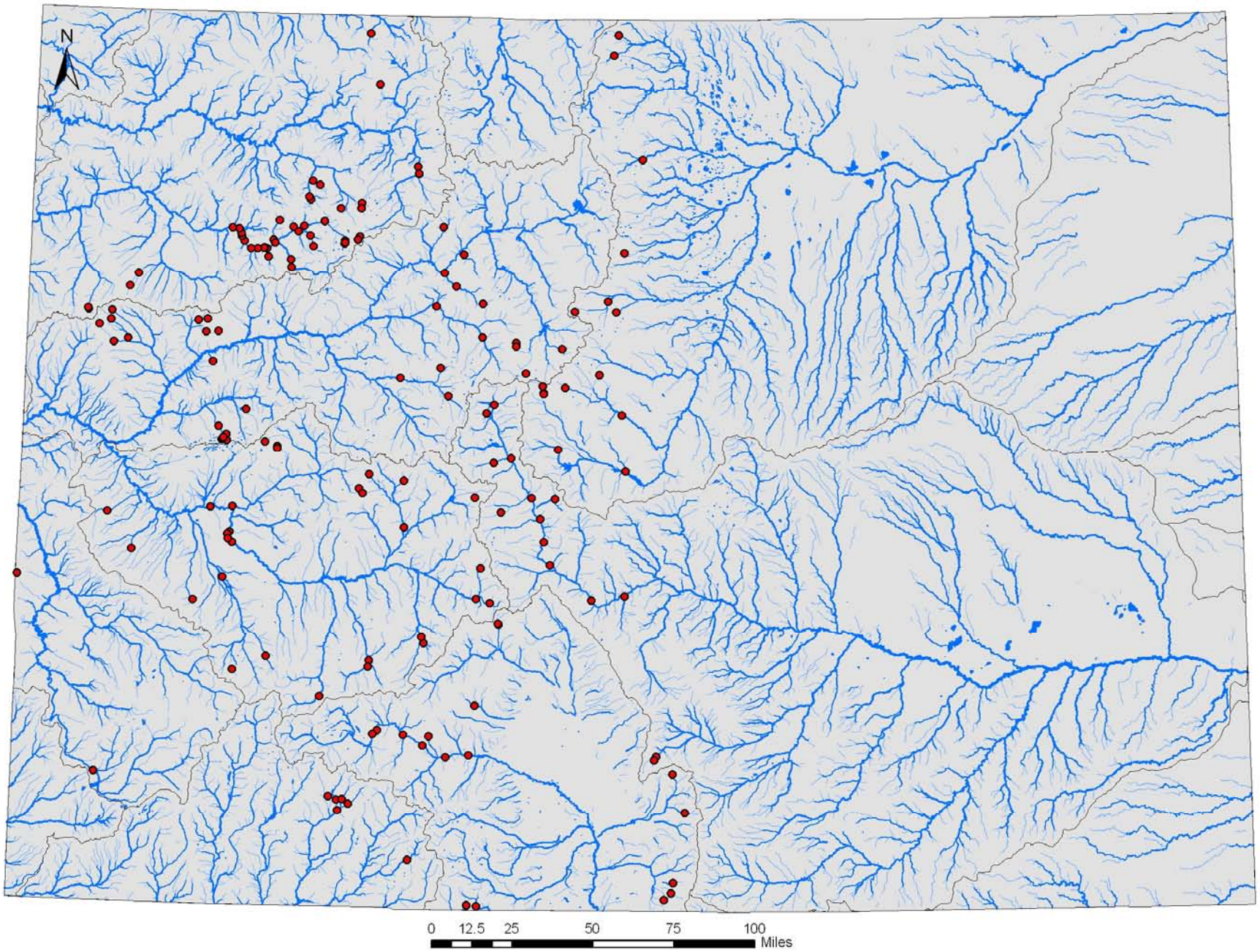
Map 2. Locations of sample sites tested for *Myxobolus cerebralis* (2003-2008)



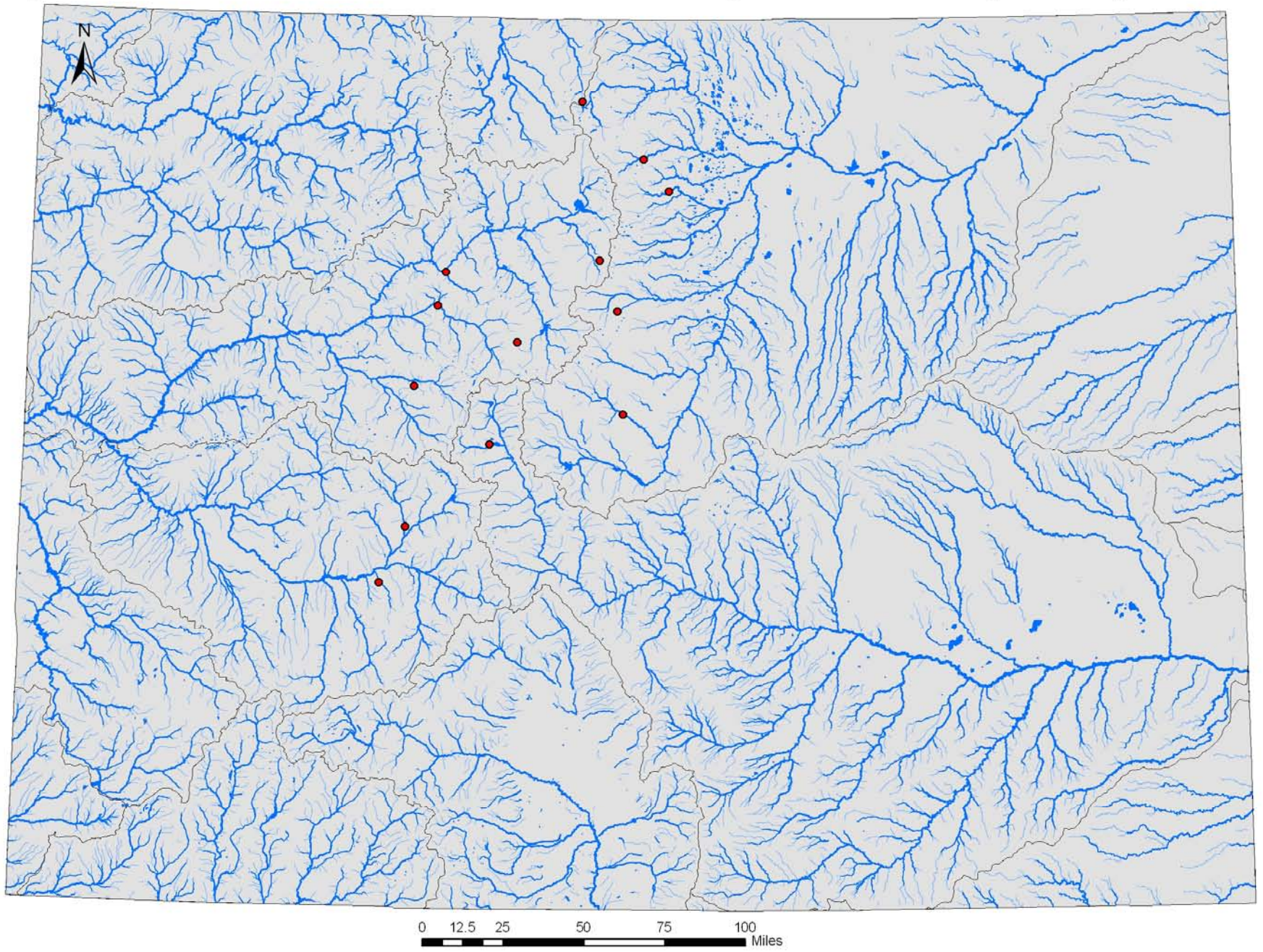
Map 3.*Tubifex tubifex* collection sites where lineage I worms were present (2003-2008)



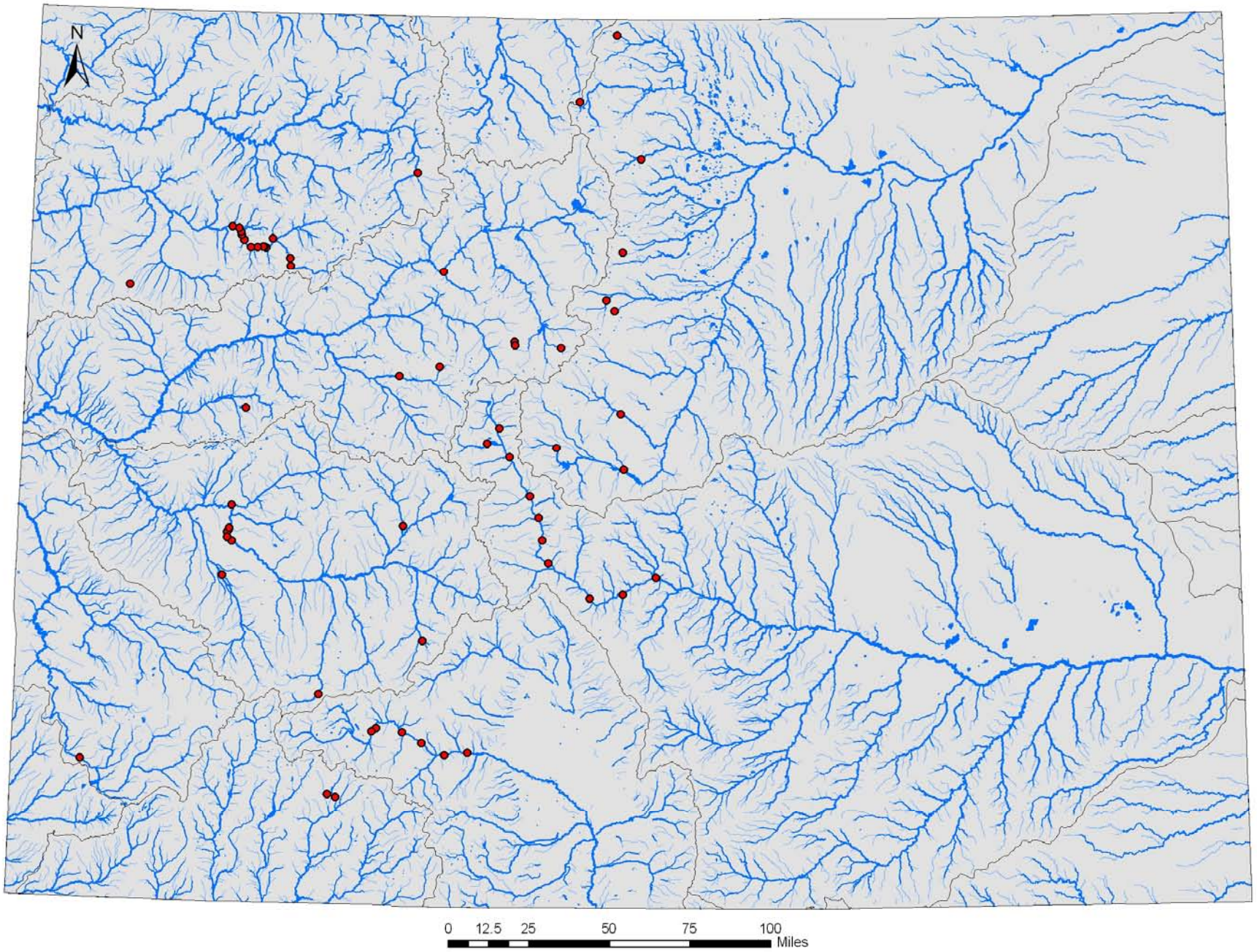
Map 4. *Tubifex tubifex* collection sites where lineage III worms were present (2003-2008)



Map 5.*Tubifex tubifex* collection sites where lineage V worms were present (2003-2008)



Map 6. *Tubifex tubifex* collection sites where lineage VI worms were present (2003-2008)



Map 7. Sample sites where *Tubifex tubifex* lineages were not found