GLEN CANYON ENVIRONMENTAL STUDIES

PHASE II

ANNUAL SUMMARY REPORT

Submitted By

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INTRODUCTION

This report covers ongoing research conducted in Phase II of the Glen Canyon Environmental Studies (GCES) and funded under Modification 002 of Cooperative Agreement 9-FC-40-07940 between the Bureau of Reclamation and the Arizona Game and Fish Department. The purpose of the report is to provide Reclamation with an annual summary of progress on Department research efforts directed at determining the effects of the operation of Glen Canyon Dam on aquatic resources of the Colorado River in Glen and Grand canyons.

Department research responsibilities under the Cooperative Agreement lie in three major areas of study: (1) ecosystem processes and lower trophic levels; (2) trout, and; (3) native fishes. Although major study areas are treated independently in this report, the Department is committed to integration of findings from these studies and those conducted by other research entities funded under the GCES.

The format for this report follows that of the Cooperative Agreement, i.e. by work items. For each work item, background information is provided to explain its importance in the context of the overall study. Scientific methods being used and progress in using those methods are then detailed. Finally, problems associated with fulfilling objectives of the work items are provided and, where applicable, suggested solutions to these problems are given.

The Department's research program is not at a stage where results from data compilation and analysis are appropriate. Therefore, this level of reporting is reserved for the EIS Draft and Final Technical Reports due October 30 and December 31, 1991, respectively.

ECOSYSTEM LEVEL PROCESSES AND LOWER TROPHIC LEVELS

Work Item 1.1 - Determine the effects of different flow regimes on primary production and organic matter and nutrient (nitrogen, phosphorus, and silica) loading rates and budgets for the Glen Canyon Dam tailwater to Lee's Ferry.

Background--Previous studies on the effects of Glen Canyon Dam operations on aquatic resources have not addressed ecosystem level processes such as primary production, nutrient cycling or organic matter processing. These processes are important determinants of fish production in the Lee's Ferry tailwater, and they may have important implications for the entire Grand Canyon ecosystem if downstream communities rely largely on nutrients and organic matter exported from the upstream reach.

Methods and Progress--Eleven input-output sampling efforts have been made to date. During each effort, water and organic matter are collected at Glen Canyon Dam and Lee's Ferry. Lake Powell is sampled quarterly. Sample components include biomass,

chlorophyll, and nutrient content of fine and coarse particulate organic matter, nutrient and organic matter concentration of the dissolved fraction, and taxonomic composition and density of zooplankton. Biomass, chlorophyll, and taxonomic determinations are made by Department personnel. Dissolved components are analyzed by the USGS laboratory in Arvata, Colorado. Particulate organic matter is being held frozen until a contract can be obtained with a laboratory having a CHN analyzer.

Problems--The only major problem with the organic matter/nutrient budget has been our inability to have the nutrient content of particulate organic matter analyzed in a timely manner. It has proven impossible for the USGS laboratory to analyze these constituents at a suitable minimum level of detection. We are in the process of negotiating a contract with a laboratory having the appropriate instrumentation for these analyses.

Only very limited primary productivity measurements have been made to date. Until recently we lacked the appropriate instrumentation and sufficient manpower to conduct these measurements. A goal of this portion of the ecosystem processes subproject is to develop a model of light limitation on primary productivity in the Colorado River as related to dam operations. In order to achieve this goal, we need to have meteorological inputs and turbidity measured in the Lee's Ferry reach. These measurements also would provide valuable data for a model predicting the degree of warming possible from the multiple inlet structure proposed as a structural element in the Glen Canyon Dam Environmental Impact alternatives. Requests for meteorological stations have been proffered to the GCES office, but as of this date no decision has been made.

weather station equip.?

Work Item 1.2 - Determine the life history, secondary production, and causes of mortality for the amphipod, *Gammarus lacustris*, in the Glen Canyon tailwater to Lee's Ferry.

Background--The amphipod, *Gammarus lacustris*, is an important component in the diet of rainbow trout in the tailwater. Effects of dam operations on *Gammarus* and availability of the amphipod to trout are not well known. Potential effects of dam operations are increased mortality due to stranding, drift and subsequent consumption by trout, transport out of favorable habitats, and limitations on the productivity of food resources important to the amphipod.

Methods and Progress--Several series of benthic samples have been collected using a mini-Ponar dredge to determine substrate-specific distribution of *Gammarus*. Three 24-hour trout diet sampling efforts have been completed to determine relative mortality of the amphipod through the diel discharge cycle. Trout were collected at 4-hour intervals over the diel period, and simultaneous drift samples were collected

using high-speed Miller tubes. Additional 24-hour studies are planned to investigate other sources of mortality.

Problems--Estimating reach-wide secondary production of *Gammarus* in the tailwater may be quite difficult due to the large area involved and major sampling problems associated with deep mainchannel habitats. Ponar sampling is not equally effective in all habitat and substrate types; thus, this gear is not suitable for making quantitative density estimates. Realistic estimates of secondary production can be made only if appropriate habitats can be sampled reliably. If *Gammarus* is widely (and deeply) distributed in the reach across a variety of substrates requiring the use of several sampling gears, reliable estimates of secondary production may not be obtainable. A problem with all aspects of the *Gammarus* studies under present flow regimes is that there are essentially no permanent habitats or algal food resources for the animals above the 5,000 cfs level.

Work Item 1.3 - Determine the effects of fluctuating flows on algal and invertebrate colonization rates, standing crops, and community composition.

Background--Effects of fluctuating flows on colonization rates of plants and animals in the tailwater are not well documented. Previous work has suggested that there are thresholds of tolerance to desiccation for certain taxa. We seek to expand our knowledge of this process, since it is important to understand ecosystem resistance to hydrologically mediated disturbance and rates of recovery from disturbance. Biotic communities (seral and equilibrium) that result from different levels of disturbance may not affect fishery and other downstream aquatic resources equally.

Methods and Progress--One colonization experiment has been completed utilizing sandstone substrates on two transects. An initial finding of interest is that there is more algal biomass associated with sand accumulating on the substrates than was previously suspected. Further studies will need to consider this substrate type, because it is so extensive in the tailwater and because *Cladophora* on coarser substrates is so susceptible to desiccation at low flows. Sand substrates may be a source of primary productivity (and dissolved organic carbon and nutrients) that is relatively less affected by dam operations than coarser substrates.

Problems--Lack of *Gammarus* habitat and algal food resources above 5,000 cfs also creates a problem for colonization studies. Natural cobble substrates above 5,000 cfs have not recolonized with algae since the beginning of the research flows. A technical problem with the use of sandstone substrates for colonization is that this substrate does not adequately sample some invertebrates, particularly *Gammarus*. Furthermore, in the zone of fluctuation, the amphipod probably will not colonize bare substrates. Thus, separate studies for invertebrate and algal colonization will be needed.

Work Item 1.4 - Determine the effects of different exposure and desiccation on the nutritive quality of exposed algae.

Background--The effects of fluctuating flows on algal quality and quantity are important for two reasons. First, it has been hypothesized that trout and other fishes derive a significant amount of energy by ingesting *Cladophora* that may or may not have associated epiphytic diatoms and bacteria. Desiccation of algae may cause partial processing which could increase nutritive quality of this resource. Secondly, dewatering and desiccation may increase the amount and quality of algae that enters the drift when algal-bearing substrates are reinundated and transported downstream.

Methods and Progress--Several small experiments have been completed in the sluiceways at Glen Canyon Dam to evaluate the effect of desiccation on algal appearance, biomass, and chlorophyll content. Cobbles were removed from the river and placed in the sluiceways. After an acclimation period, algal samples were taken. A subset of the cobbles was removed from the sluiceways, allowed to desiccate for different periods, and then sampled. Additional experiments of this type are planned.

Problems--We presently do not have the capability to assay algal samples for nutritive quality, and we are seeking the services of a contract laboratory to conduct these analyses. Manpower shortages also have precluded the initiation of larger scale desiccation experiments. } Recs.

TROUT

Work item 2.1 - Determine the potential loss of trout spawning, defined as areal loss of spawning bars and exposure of redds, at various flows in the reach of the Colorado River between Glen Canyon Dam and Lee's Ferry.

Background--GCES Phase I studies indicated that 27% of the adult rainbow trout collected in the Lee's Ferry reach during 1984-1986 were naturally produced within the system. Trout reproductive success can be affected by dam operations at egg to adult stages through daily water level fluctuations which cause stranding and desiccation or displacement out of preferred spawning and rearing habitats. Sustained releases of clear, nearly sediment-free reservoir water can also affect spawning through long-term armoring of spawning bars. This study will identify the extent of suitable spawning gravels in the Lee's Ferry reach at various water levels and the relative use of these substrates by spawning trout during different flow regimes.

Methods and Progress--Spawning bars at River Mile 4.0, 6.1, 8.9 and 14.0 were surveyed during 5,000 cfs discharges in October 1989, June 1990, and June 1991 for sediment particle size and degree of embeddedness. On each bar, three equidistant transects were run perpendicular to the river bank. Samples for evaluation

of spawning gravel size were taken from along each transect. Measurements of the wetted perimeter of each spawning bar at 3,000, 5,000, 8,000 and 11,000 cfs were made from stakes placed along the high water mark. Stakes and a subset of wetted perimeter locations were surveyed by the USGS and tied to known bench marks. These measurements are being used to draw contour maps of the bars and wetted perimeters at different flows.

Beginning in November 1990, the four spawning bars were checked weekly for redd development. Redd building activity did not begin until late December 1990. Spawning bars were surveyed at least once during each study flow to locate, mark, and map active redds. To date, 430 redds have been located at 4.0 mile bar and 178 at 8.9 mile bar. The study has concentrated on these two spawning bars, as there was very little observed spawning activity on 6.1 mile bar and none on 14.0 mile bar.

Problems--Other commitments by USGS has delayed generation of some discharge contour maps, thereby delaying our analyses of spawning gravel size, embeddedness and redd placement. In order to have the maps available in time for inclusion in EIS Technical Reports, we propose to have Department personnel generate these maps using computer hardware and software available in the GCES office in Flagstaff.

Compass coordinates are not accurate enough to uniquely identify redds in close proximity to each other. This problem could be alleviated by purchasing Lietz or Nikon surveying equipment and having a qualified surveyor available to pinpoint exact coordinates of the redds. Depressed levels and the delayed onset of trout spawning activities in 1991 will require continuation of research in this area into future years.

A possible future problem concerns getting low enough flows during daylight hours to count and map redds. Flows below 8000 cfs, and preferably below 5000 cfs are needed to accurately find and map redds. Typical winter operation of the dam brings the flow up before daylight. One day of low controlled flows every two weeks during the spawning season is necessary in order to complete this research.

Work item 2.2 - Determine the rate of stranding and mortality of naturally reproduced and stocked trout under different flow regimes in the Glen Canyon tailwater downstream of Lee's Ferry.

Background--It was observed during Phase I studies that some trout became stranded in backwaters and pools that had become isolated as water levels declined in the Lee's Ferry reach. Little was known about the degree of stranding and the rate of mortality for these fish. This study is intended to identify major stranding areas in the Lee's Ferry reach and determine mortality of adult fish relative to season and flow level.

Methods and Progress--Both daytime and nighttime stranding pool surveys were conducted between Glen Canyon Dam and Lee's Ferry several times at low flows to identify stranding sites and at what discharge levels these stranding pools became isolated. Ten major stranding areas at river miles -0.5R, 4.0R, 8.2L, 8.9R, 9.7R, 11.0R, 11.7R, 11.9L, 13.0R and 14.8R were identified. These areas were surveyed four times a month from March-August 1990 and at least twice during each study flow, or twice a month during periods of "normal flow" from September 1990-February 1991.

During each survey, pool dimensions, number of fish stranded, dead and alive, were recorded in addition to water temperature, dissolved oxygen, pH, and conductivity. Dead fish were collected, weighed and measured, and examined for fluorescent dye pigments. Viscera were collected for later analysis of stomach content and gonadal development. Heads were taken and frozen for later examination of vertebrae for presence or absence of tetracycline dye rings used to differentiate hatchery produced trout from those naturally reproduced in the Lee's Ferry reach.

Mortality surveys determined that only during summer research flows did changes in water quality parameters prove lethal to trout. Three times during the 1990 summer 5000 cfs flows trout were killed probably due to an increase in water temperature. Other water quality parameters ostensibly remained within acceptable limits. The only known causes of death during the spawning season were dewatering of the stranding pool and predation. Comparisons of food habits and gonadal development will be made between trout remaining in the main channel and those that get stranded.

Problems-- The level of detection of tetracycline dye marks in vertebrae has been too low to successfully discriminate between hatchery reared and naturally reproduced trout. This problem, which was previously undetected, has arisen as a result of changes in antibiotic levels administered to trout fingerlings prior to their stocking. However, total numbers stranded and mortality rates of those stranded can be calculated regardless of the origin of the fish. Most stranding occurred during the spawning season and thus very few data are available for stranding during the study flows, and thus comparisons can't be made among different flow regimes.

Addendum to Work Item 2.2. - Conduct a literature review of trout strains. This should include an assessment of the relationship between trout strains and their interaction with flows, growth, survivorship and movement.

Background--Over the years, several strains of rainbow trout were stocked at Lee's Ferry, but until recently poor records were kept on which strains were stocked. Trout which are capable of reproducing in the Lee's Ferry reach have come from an unknown genetic source. There is some speculation that another strain of trout might do better under the conditions encountered at Lee's Ferry. Of particular interest is

finding a strain which will reproduce during a narrower period of time, and therefore require controlled flows for a shorter period. Two studies were added to the Department's obligation to provide this information. First, a literature review of available trout strains and their characteristics is being undertaken. Second, trout which presently occur in the Colorado River and its tributaries, as well as hatchery stocks, are being analyzed by gel electrophoresis to determine their genetic composition and degree of relatedness. Both these studies are being conducted under subcontracts.

Methods and Progress--A request for proposal was developed for literature review of trout strains and proposals were accepted and reviewed. William Davis and Dr. George Ruffner were selected to conduct this review. The scope of the work will include compiling information on the performance characteristics of commercially available strains, identification of tailwaters which are similar to Lee's Ferry and their associated trout strains, and evaluation of available strains against conditions resulting from current operating regimes. This subcontract was let on April 30, 1991. A report is due on October 30, 1991, therefore it may not be available for the Draft EIS Technical Report. Results will be included in the Final EIS Technical Report.

Descriptive genetic studies were also undertaken in April 1991. Samples of adult and young-of-the-year trout were collected from 10 tributaries within the Grand Canyon and 9 mainstream sites (including 3 sites in the Lee's Ferry reach). Eyes, livers and muscle tissue were collected from adult trout and frozen in liquid nitrogen. Young-of-year fish were frozen whole. Samples were also collected from both groups of fish stocked at Lee's Ferry this year. These samples were sent to Dr. David Phillip and Ms. Julie Claussen at the Illinois Natural History Survey for analysis using gel electrophoresis and histochemical staining. Phillip and Claussen conducted a similar study during Phase I and have already identified enzymes and major loci which exhibit the greatest genetic differences. Samples will be analyzed at Illinois Natural History Survey starting in July. A draft report is due September 30, 1991 and a final report November 30, 1991. Therefore, results will be available for inclusion in the Final EIS Technical Report.

Work item 2.3 - Determine the effects of fluctuating flows on age and growth relationship of stocked trout in the Glen Canyon Dam tailwater downstream of Lee's Ferry.

Background--Past evaluation of age/growth relationships of trout stocked in the Lee's Ferry reach has been restricted largely to use of periodic length frequency distributions. Conventional methods for aging fish, such as scales and otoliths, are precluded because annuli are not laid down in the perennially cold tailwater. Since 1989, stocked trout have been marked with an external fluorescent pigment. This marker is considered adequate to identify cohorts for periods of 2-3 years, but there

remains a need to institute a method of marking which will suffice to follow trout until they reach trophy size. Interpretations of how growth has changed over time at the Ferry can be made, however, establishing the influence of fluctuating flows on trout growth is more difficult.

Methods and Progress--Trout were collected quarterly using an electrofishing boat with a Coffelt VVP delivering a modified pulse system developed to minimize damage to trout. Each time fish were collected from 4-8 randomly selected sites throughout the 16 mile reach from the Dam to Lee's Ferry. Fish collected were weighed, measured and examined for fluorescent pigments or other external markers. Representative samples were sacrificed for comparison of food habits, fecundity, and origin with stranded trout. Additional trout were collected from backwaters, side channels, and mainstream areas using seines and trammel nets. All fish collected were weighed, measured and tagged with numbered floy tags. Growth rates were calculated from data obtained from the recapture of tagged fish.

A parallel "laboratory" experiment to evaluate growth of fingerling trout under fluctuating and constant flows is to be conducted in the sluiceway below Glen Canyon Dam. Efforts to institute this study have been stymied until a way can be found to eliminate or dampen the surge which results from pumping seepage water from the dam.

Pigment retention characteristics were analyzed by holding a number of rainbow trout from the 1989 and 1990 stockings in the sluiceway below the dam. As of December 1990, 77% of the 1989 fish and 58% of the 1990 fish still carried the fluorescent mark. In the future, fluorescent pigment marks will be supplemented with a second, more permanent, marking technique. Different tagging techniques currently are being explored and, of those investigated, binary coded wire tags appear most promising. A satisfactory method will be chosen and implemented for the 1992 stocking.

Problems--A high priority for next year will be finding a reliable way to mark all hatchery trout so that they will be identifiable for a long period of time. This is important since many study questions are based on being able to differentiate naturally produced trout from hatchery stocks.

Several impediments plague attempts to design an experiment to mimic operational cycles in the sluiceway. These include a periodic surge in the waste water coming from the dam which either needs to be modified or dampened, and determining a means to control the amounts of food supplies which are present or delivered to each side. A possible future problem for studying age and growth relationships concerns interim flows and how they will impact the mark/recapture program. The 5000 cfs constant flow weekends were used extensively to net, mark, and release trout for the

growth studies. Higher water levels will make the task more difficult, and it is uncertain to what degree this may impact the study.

Work item 2.4 - Determine the behavioral responses of trout in the Glen Canyon Dam tailwater to different steady and fluctuating flow regimes.

Background--Changes in river stage, current velocity, and wetted area that accompany fluctuating flows have the potential to affect the behavior patterns of adult trout during reproductive and feeding activities. This may result in additional stress and energy expenditure in the trout.

Methods and Progress--Radio telemetry was used to evaluate the behavioral responses of adult trout to constant and fluctuating flow regimes. Ten adult trout (40-50 cm TL) collected and released around 13.5 mile bar and 6.1 mile bar were implanted with radio transmitters in November-December 1990. After implantation was complete, trout were given a numbered floy tag on the right side to differentiate them from other tagged trout.

Trout were located using a Model 2000 ATS programmable receiver with a mast antenna mounted on a boat, and portable loop antennas were used for directional triangulation. Triangulation was accomplished from stakes that had been surveyed to bench marks to facilitate mapping of locations. Triangulations on all tagged trout were completed before habitat measurements were taken. Measurements of depth, substrate size, cover, velocity, and distance to shore were made for each site.

Triangulations and habitat measurements were made on all located trout three times per day for two days during each study flow. Sample days during constant and fluctuating flow periods were sequential to avoid seasonal variation. Observations were timed to catch the low, peak, and declining stages during fluctuating flows. The same time periods were used during constant flows.

Individual fish also were followed for 24 hour periods during normal winter and spring flows, and during the 5,000 and 15,000 cfs steady flows. Hourly triangulations were made, but habitat measurements were not taken during this part of the study.

Problems--Areas where fish locations are being measured have been surveyed by the USGS, and these maps are needed before radiotelemetry data can be completely analyzed. There also is a limitation in accurately pinpointing trout locations using loop antennas and magnetic compasses. Thus, small-scale movements, on the order of several meters, and microhabitat use may not be measurable by this method. Augmentation of the existing methods with observations made by divers, as has been done in the past at Lee's Ferry, may alleviate this problem. Use of divers would require that certain areas of the tailwater be cordoned off from access by fishermen

and boats. This restriction would have to be imposed by the National Park Service at Glen Canyon National Recreation Area.

NATIVE FISHES

Several of the work items in this portion of our research effort are being conducted by project biologists responsible for studies in either the Little Colorado River or the Colorado River and other tributaries. Where appropriate, methods, progress, and problems with the research are divided according to these responsibilities.

Work Item 3.1. Continue the AGF monitoring and research program for native fishes of the Colorado River and its tributaries in Grand Canyon.

Background--The Department's native fish monitoring program was initiated in 1987 following the cessation of Phase I field studies. The project has been an annual effort timed to coincide with the reproductive period of humpback chub in the LCR and with the presence of early life stages of other native fishes in tributary and mainstream rearing habitats. Research personnel historically entered the LCR via Salt Trail Canyon and occupied the tributary for approximately one month a year, typically May, since 1987. This effort was augmented through additional personnel who traveled down the mainstream by river boat and entered the LCR near the middle of the sampling period. Other collections and measurements included plankton, benthos, hydrolab variables, and general habitat morphology. Coordination of this effort with other research entities is occurring through the Aquatic Coordination Team.

Methods and Progress (LCR)--USBR helicopters were utilized for shuttles of equipment and personnel due to the closure of the Salt Trail to GCES researchers. Standardized gear sets and marking procedures were continued. Thirteen hoop nets have been run nearly continuously in the lower 1200 m of the LCR since early May, and six additional sites between River Kilometers (RK) 9.1 and 11.6 since mid-May. Trammel nets were run sporadically during this period from both camps, but were discontinued due to mortalities of humpback chub. Considerable seining effort was expended in various habitats in the vicinity of both camps, and larval drift nets were run every other day from both camps during these periods. Extensive measurements of sampled habitats, including current velocities, substrates, depths, and cover features were also undertaken. Water temperatures and other physical-chemical variables (conductance, pH, dissolved oxygen) were monitored from the lower camp.

(Mainstream and Other Tributaries)--Mainstream activities associated with our annual monitoring largely will be subsumed under Work Items 3.4 and 3.9. There will not be a monitoring river trip *per se*, rather this trip will simply be one of several conducted annually.

Problems (LCR)--The decision to run hoop nets twice daily precluded setting nets at previously established locations at longer distances from the camps. Mortalities of adult humpback chub which occurred in as little as two hours in trammel net sets restricted our ability to maintain continuous sets of these gears. Certain net sets may not have been as exactly located as in earlier years since we did not have personnel experienced with those sites. An attempt to get C.O. Minckley for this purpose was unsuccessful due to a breakdown of helicopter support. This may restrict our ability to compare individual net sets with previous years, but should not impair temporal catch rate comparisons.

Work Item 3.2--Identify the temporal and spatial distribution patterns and movements of early life stages of fishes in the Little Colorado River and, if necessary, other tributaries.

Background--As indicated, the major emphasis for this work item is on the LCR. However, sampling of fishes in and above the confluence zone of other tributaries is being accomplished with a combination of larval drift nets and hoop nets. Using these gears, we hope to obtain measures of the amount of transport and active movement of fishes from tributary to mainstream habitats. These data will help us to further pursue an objective of GCES Phase I which was to determine the relative contributions of mainstream and tributary reproduction to fish populations in Grand Canyon. Native larval fishes collected with drift nets will have their otoliths analyzed for daily growth rings for comparison with individuals collected from mainstream rearing habitats (Work Items 3.4 and 3.7). Invertebrates and coarse particulate matter collected in these samples will also be analyzed to determine contributions of these materials from tributaries to the mainstream.

Methods and Progress (LCR)--Present investigations will substantially increase research activities directed at young-of-the-year fishes in the LCR. Information to determine the timing and duration of reproductive activity for different fish species will be provided by analysis of otoliths and length frequency distributions of early life stages. Measurement of contemporaneous environmental conditions to delineate cues that potentially serve to initiate spawning will be available from U.S. Geological Survey stations on the LCR at Cameron and above the mouth. Movements are to be evaluated through marking procedures and analyses of drift. Other behaviors of early life stage fishes will be evaluated both visually and with video, and may include laboratory experiments which have yet to be formalized.

Larval drift nets in the LCR thus far have been emplaced at two permanent locations--at the mouth of Salt Trail Canyon and RK 1.8 near our lower campsite. All nets had current velocities taken at their mouths to quantify the volume of water filtered. Since early May, between 1 and 3 nets at each site were run during crepuscular,

daylight and darkness hours every other day for periods that varied according to the amount of drift in the water column. Post-larval investigations have included sampling with fine-meshed seines and minnow traps, along with contemporaneous habitat measurements.

(Mainstream and Other Tributaries)--Larval drift samples in the mainstream and selected tributary mouths were taken during the March-April and May river trips. Larval and young-of-year fish collections were also made with seines and dip nets in Kanab Creek and mainstream backwaters. Preliminary sorting of all larval fish samples collected to date has been completed.

Problems (LCR)--The major impediment to further progress this reporting period was due to a delay in funding to Arizona State University. Department personnel fulfilled a commitment to conduct sampling in the upper reaches of the LCR study area that otherwise would have been done by University personnel. Sufficient manpower was not available for initiation of many intended activities regarding larval and post-larval fish investigations, including behavioral quantification and movements and marking. Other areas of investigation such as relating environmental cues to reproductive events, species segregation, temporal drift patterns, and food habits, are dependent on subsequent laboratory and statistical analyses.

Work Item 3.3--Provide for the propagation of native fishes of the Colorado River in Grand Canyon for use in laboratory or hatchery based studies necessary to satisfy the needs of the Section 7 Conservation Measures.

Background--This work item is directed at providing early life stages of native fishes for laboratory or hatchery-based studies on thermal shock, temperature dependent growth, reproductive behavior, and other areas of investigation. The Department's hatchery at Page Springs has been selected as the site for propagation. This facility presently is used for hatching and rearing of Colorado squawfish and razorback sucker destined for reintroduction in Arizona waters.

Methods and Progress--In late March of this year, adult humpback chub from the lower LCR and confluence area were collected by hoop and trammel nets and held in live cars. One ripe female was stripped of eggs and fertilized with sperm from several ripe males. These eggs, along with 10 adult humpback chub were transported by helicopter from the lower LCR to the head of the Salt Trail on April 1. From there they were transferred to a hatchery truck and taken to Bubbling Ponds Hatchery. An additional transfer of adult humpback chub is scheduled for this autumn. — problems?

Problems--The approximately 300 presumably fertilized eggs died shortly after arriving at the hatchery. Daily injections of carp pituitary extract into reproductive age females in the field failed to induce maturation and expression of eggs following

periods of up to 72 hours (3 injections maximum). Four adult chub at Bubbling Ponds died when they jumped out of a circular holding raceway.

Work Item 3.4--Determine changes in environmental conditions in mainstream and tributary confluence zone native fish rearing habitats under different flow regimes.

Background--Exact environmental conditions for successful rearing of native fishes in the Colorado River in Grand Canyon are unknown, but larval to young-of-the-year distributions suggest preferences for nearshore, low velocity habitats. The LCR provides important breeding and rearing habitats for humpback chub and other native fishes, although use by adults may be largely seasonal. Impoundment of tributary flows by high mainstream stages has been observed by Department researchers, who proposed that these impounded confluence regions might serve as important staging sites for adult native fishes and rearing sites for young-of-the-year. Under fluctuating flow regimes, however, confluence zone water temperature and current velocity vary through the course of each diel cycle in conjunction with changes in mainstream stage. Mainstream backwaters also are known to serve as important rearing habitats for native fishes. Fluctuating flows also affect these habitats. In cases of extreme fluctuations, backwaters may be converted to large eddies at high flows and subsequently be dewatered at low flows.

Methods and Progress (LCR)--Water depth, water temperature, pH, dissolved oxygen, and specific conductance will be measured by automated continuous or manual recording instruments in the LCR mouth and adjacent mainchannel sites under a variety of flow conditions. Supplementary interval measurements also will be made of air temperature, solar radiation, turbidity or light extinction at depth, and current velocity. All measurements will be made for a minimum of two flow cycles at consistent locations within each habitat. We anticipate that our measurements will be augmented by those made by the U.S. Geological Survey and by the Fish and Wildlife Service sampling program for habitat evaluation. Mapping will be tied to the Geographical Information System database wherever possible.

(Mainstream and Other Tributaries)--Sampling of rearing habitats has been divided into three levels of intensity. Level I involved sampling fishes, taking water temperatures, and making rudimentary measurements of habitat morphology. Level II added plankton, benthos, sediment characteristics, other hydrolab variables, current velocity, turbidity, and more refined measurements of habitat morphology. Level III measurements include all those of Level II, but involve taking them at six hour intervals to analyze changes associated with the 24 hour hydrograph. Hydrolab measurements, including change in water level, are also made at close intervals in time with automated Datasondes. Formal topographic surveys are made and fishes are sampled at fixed sites with minnow traps in order to equate their distributions with changes in local habitat conditions.

Some field methods described above have proven difficult to accomplish during extreme fluctuations and some sampling gears have not been productive. Therefore, these methods are being refined. Topographic surveys using professional Leitz equipment and an electronic datalogger have been completed on 12 backwater sites and data currently are being analyzed in order to create detailed site maps. Computer generated maps will be produced in the Flagstaff GCES office using available hardware and software. Beginning in July, topographic surveys will be supplemented by extensive plane table mapping. Although the new technique will produce lower precision maps, it will allow us to map an additional 20 sites per river trip. Mapping data will be linked with data concerning fish densities, algal and invertebrate standing crops, and physicochemical characteristics of the habitats in order to fully characterize patterns of habitat use for smaller fishes. Ultimately, many of these maps will be tied into the GIS being produced by Reclamation in Denver. We hope to be able to able to overlay habitat conditions and fish distributions onto the landforms generated from topographic surveys. With a refined flow-routing model from USGS, we should be able to make some credible predictions for habitat use by early life stages of native fishes during specific flow regimes.

Problems (LCR)--Delays in the purchase of automated recording equipment and additional commitments for ASU research have set back environmental measurements in the mouth of the LCR. Efforts on this work item will increase as ASU begins their research in July.

Problems (Mainstream and Other Tributaries)--This aspect of the study is well underway and sampling problems are being resolved. One eventual problem involves the complexity of proposed analyses. The envisioned spatial analysis and coupling with the GIS undoubtedly will require expertise presently not available within the Department. However, a new GIS Coordinator and Specialist have recently been hired, and they will be available for consultation on analyses of GCES data.

Work Item 3.5--Determine algal and invertebrate standing crops and their relative contributions to diets of young native fishes in tributary, backwater, and mainchannel habitats under different flow regimes.

Background--Very little information is available on the diets of early life stages of fishes in Grand Canyon, although some studies have been conducted on the Green and Colorado rivers in the Upper Basin. Densities of zooplankton necessary to maintain razorback sucker larvae have been suggested from laboratory studies, but other native fishes have not been investigated. Previous monitoring has shown that backwaters generally contain higher standing crops than mainchannel habitats and that tributary mouths are similarly limited when compared to reaches above the confluence zone.

Methods and Progress (LCR)--Perpendicular-to-flow transects will be established every 5 km starting from the mouth upstream to just below Blue Spring. These will be quantitatively sampled for water quality, algae and invertebrates on a quarterly basis using inflatible kayaks transported to Blue Spring by USBR helicopters. Samples of larval to juvenile native and introduced fishes will also be collected for analysis of digestive tract contents. The analysis will compare digestive tract contents with available food resources in these respective habitats for evidence of selectivity and as corollary information for determination of movements among habitats.

Work schedule

A full longitudinal collecting trip was undertaken between May 14-18 of this year. Water quality samples, including measurements of carbon dioxide and alkalinity, and algae and invertebrates were collected from transects at 20, 15, 10, 5, and 0.07 km above the mouth. Algal and invertebrate relative abundance and diversity appeared to decrease downstream, coincident with decreasing light penetration.

data?

(Mainstream and Other Tributaries)--Numerous coincident collections of early life stages of fishes and invertebrates have been made from backwaters and associated mainchannel habitats. Some invertebrate collections have been identified and enumerated, but the only analyses of early life stage stomach contents were completed under a subcontract in Phase I. Beginning in August, water samples will be analyzed for chlorophyll *a* and studies will be initiated to examine rates of zooplankton and particulate organic matter exchange between backwaters and the mainchannel. It is questionable whether this task or the one dealing with changes in zooplankton communities in backwaters during controlled flows will be completed in time for the EIS report deadline.

Problems (LCR)--An earlier trip scheduled in late March was cancelled due to inclement weather.

(Mainstream and Other Tributaries)--Some analyses requiring taxonomic identifications require expertise that is lacking in our mainstream crew. Identification of algae is particularly problematic. There is an individual on the LCR crew who has the necessary expertise and may be interested in working on these analyses. It's quite possible, however, that at least some of the taxonomy may have to be done through a subcontract.

Work Item 3.6--Determine behavioral responses of larval to juvenile native fishes to changing environmental conditions in rearing habitats during controlled flows.

Background--During October of 1986, Reclamation provided back-to-back periods of steady and fluctuating flows. Department research biologists conducted a mark-recapture experiment on juvenile fishes in backwaters during that time in order to compare changes in catch rates and body sizes and to track movements. Results

were not very definitive, but one conclusion drawn was that juvenile native fishes were seldom stranded on dewatered sediments by falling flows. Where then did they go? Were they able to find refuge in nearby eddies or were they displaced into the mainchannel and carried downstream? At what stage in the flow cycle did they vacate backwaters being dewatered? The native fish study plan proposes to answer these questions through a combination of direct observations (in sufficiently clear waters), recapture of marked fishes, and evaluation of stomach contents of fishes in backwaters and associated mainchannel habitats.

Methods and Progress (LCR)--Larval drift nets are being used to measure drift of early life stages and their potential passage into the mainstream. All native fishes of sufficient size captured in the confluence zone will receive a fin-clip unique to their original capture location. We also are investigating the use of immersion dyes as an alternative to fin-clip markings. Recaptured individuals will be preserved for analysis of otoliths and stomach contents. Otolith analyses, as described below, will be used to compare early life stages from tributary and backwater habitats in an attempt to determine the origins of these fishes (cold mainstem versus warmer tributaries), time of passage if spawned in tributaries and drifted into the mainstream, and their growth rates in these respective habitats.

Little progress has been made evaluating drift and behavior of early life stages in the confluence zone because of this year's additional monitoring responsibilities. Rather, most drift is being measured at the permanent camps approximately 1.8 km and 10.5 km above the mouth. More emphasis will be placed on the confluence zone when ASU enters the LCR in early July.

(Mainstream and Other Tributaries)--No larval and few juvenile fishes were present in rearing habitats during the March-April river trip. The same condition persisted during the May river trip until below Kanab Creek, at which time flows changed from 5,000 cfs constant to 15,000 cfs constant. Although relatively few larval fish have been encountered to date, a total of 27 humpback chub and approximately 2300 fish of other species have been collected and either released or preserved for further study. Techniques for marking smaller (<60 mm) fishes are being investigated and the implementation of small, binary coded wire tags appears promising. This summer's field activities will include refining the methods necessary to answer questions on movements and behavioral responses of larval to juvenile fishes. Thus, there will probably be limited information related to this work item for the EIS report.

Problems--A major problem for this work item facing both LCR and mainstream investigators is the development of a marking technique for larval fishes that will not induce high mortality or affect their behavior. This problem should be addressed using experimental animals from our propagation efforts.

Work Item 3.7--Determine age structure and growth rates of native fishes of the Colorado River in Grand Canyon. Relate these life history features to hydrologic and thermal conditions experienced by fishes during their growth to present size.

Background--Knowledge of age structure, growth rates, and survivorship to a given age is fundamental to our understanding of the life history of native fishes in Grand Canyon. Present emphasis on determination of the relationship between age and size is being placed on otoliths, because scale annuli have proven unreliable for humpback chub that have spent portions of their lives in the Little Colorado and Colorado rivers. Preliminary work on young-of-year humpback chub suggests that daily growth rings can be determined. Since growth rate is temperature dependent in fishes, we hope to be able to differentiate the histories of young humpback chub in relatively warm tributary waters from those that entered the cold mainstream at some point in their lives. If we can determine the date of their entry into the mainstream, then we can relate these movements to tributary and mainstream hydrologies.

Methods and Progress--Specimens of humpback chub for these determinations have been sent to Dr. Dean Hendrickson at the University of Texas at Austin. We anticipate that a series of 50 or more otoliths will be examined in time for incorporation into the Final EIS Technical Report. Hendrickson will be responsible for only daily age determinations on young-of-year humpback chub and annual determinations on older individuals; other native fishes will have to be done under separate contract, probably with Dr. Ed Brothers of Cornell University, who will do verifications on Hendrickson's initial analyses. Results from other native fishes likely will not be available for the report. Also, the otolith analyses will require verification of ages from experimental animals grown under differing thermal regimes, and it is very doubtful that these confirmatory analyses will be completed by that time. A surrogate species, bonytail chub, is presently being grown at three water temperatures at Dexter National Fish Hatchery to provide otoliths for the confirmatory analyses. Following successful propagation of humpback chub at the Department's Page Springs hatchery, these tests will be repeated on that species.

Problems--Initial humpback chub otolith analyses done on asterisci have proven to be unreliable for aging. Consultation with Dr. Brothers has confirmed that the lapillus is clearly the stone to use, since it displays clear daily increments in young fish and far less ambiguity in annual zones for older individuals. Field experiments described in this work item in which young humpback chub are held alternately in the Colorado and Little Colorado rivers have not been completed because of added monitoring responsibilities. Provided that permits for sacrifice of sufficient numbers of young humpback chub are authorized for 1991, we will attempt these experiments during summer months.

Work Item 3.8--Compare otolith edge chemistry of native fishes collected in tributary and mainstem habitats for use in growth and movement analysis.

Background--The major purpose of these studies is to determine the stream of residence for native fishes throughout their life until collection and to relate environmental conditions in the stream to rates of growth. Integration of this method with ageing techniques using otoliths provides a great potential for deciphering the relationships among age, growth, and environmental conditions in the streams of residence. The method involves microscale evaluation of elements or isotopes across the face of otoliths using a mass spectrometer or electron microprobe. It assumes a relationship between ion or isotope ratios and the chemistry of the aquatic milieu in which the fish was growing at the time otolith material was laid down. If this relationship exists, then native fishes should have different elemental ratios in their otoliths during periods of life spent in the Little Colorado and Colorado rivers.

Methods and Progress--Field collections for these analyses will be made by Department research personnel. Otolith analyses will be conducted by Drs. Hendrickson and Brothers. Chemical composition of otoliths from humpback chub of uncertain history caught in the Colorado and Little Colorado rivers and specimens of certain history held for known periods in each stream will be sampled along transects originating at the focus and extending to the edge. Sampling interval will be approximately 10 μm. Potential chemical sampling instruments include inductively coupled plasma atomic emission spectrometry, electron microprobe, laser ablation ICP mass spectrometry, or tunable laser dye mass spectrometry.

Progress on this work item largely has been restricted to design of field experiments and investigation of appropriate techniques for conducting the analyses. A single analysis was conducted at the University of California Davis on an astericus used for aging. Considerable variation occurred across the transect in Sr/Ca ratios used for evaluations in other published studies, but it appears that the lapillus should also be used in these analyses.

Problems--Analysis of otolith chemistry is expensive and requires the use of sophisticated laboratory equipment. Relating the stream of residence at a given time to elemental or isotopic composition requires as a precursor a definitive relationship between age and position on the otolith. Additional data on the comparative chemistries of the Colorado and Little Colorado rivers are also necessary to ascertain which elemental ratios can be expected to differ in otoliths based on the residence of fish in the respective streams. Fulfilling these requirements will be time consuming, and it is doubtful that these analyses will be available in time for EIS reports generated in 1991.

Work Item 3.9--Determine the extent to which limnological factors, with emphasis on water chemistry and aquatic productivity, potentially limit the distribution and abundance of native fishes in the Little Colorado River and other tributaries which might serve as streams for augmentation of humpback chub in Grand Canyon.

Background--Impetus for this work item arises from the observation that speckled dace is the only native species that exists in the upper 6 km of the perennial reach of the Little Colorado River and that, of all tributaries in Grand Canyon, humpback chub reproduce in only the LCR. The ostensible absence of humpback chub from the upper perennial reach of the LCR may be due to restrictions from large travertine dams and falls or from high levels of free carbon dioxide in spring inflows to that reach. The extent to which various structural, hydrological, and limnological attributes are important in determining relatively acceptability of other tributaries to humpback chub and other native fish species also is largely unknown.

Methods and Progress--Emphasis for this work item initially is on the LCR. Sampling of water chemistry, algae, and invertebrates in that tributary are being accomplished quarterly. The first successful sampling, conducted in June of 1991, was used to collect algae and invertebrates and to assess carbon dioxide, alkalinity, pH, water temperature, and dissolved solids at 5 km intervals from near Blue Springs to the mouth. In the future, quarterly assessments will be augmented by substrate colonization experiments similar to those being conducted in the Lee's Ferry reach of the Colorado River. As the Fish and Wildlife Service expands their habitat analysis to other tributaries next year, we should have the opportunity to work with them and augment their studies to include limnological aspects if deemed desirable. Methods to be applied will be the same as those being used in the LCR.

Preliminary evaluation of relative changes in CO_2 and alkalinity along the river continuum suggest that these factors do not solely account for the ostensible nonuse of the reach above Atomizer Falls (15 km above mouth) by humpback chub. Speckled dace, and apparently larvae of this species, were collected from this reach.

Problems--Some electronic water quality analysis gear did not function on this trip.