

Las Vegas Wash Water Quality and Implications to Fish and Wildlife



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November 1, 2002

FFS Numbers: 1F27 and 1F31
DEC IDs: 199810009 and 200010001

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LAS VEGAS WASH WATER QUALITY AND IMPLICATIONS TO FISH AND WILDLIFE

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INTRODUCTION

Recent investigations have documented degraded water quality conditions in Las Vegas Wash and Las Vegas Bay of Lake Mead, Clark County, Nevada (Bureau of Reclamation 2001; Snyder *et al.* 1999; Covay and Beck 1998; Covay and Leiker 1998; Bevans *et al.* 1998, 1996). The degradation of water quality has adversely affected the quality of fish and wildlife habitat associated with these areas. Environmental degradation has affected Department of the Interior (DOI) trust resources, including National Park Service and Bureau of Reclamation managed-lands. Effects to DOI trust fish and wildlife resources, including three endangered species and a variety of migratory bird species, are still under investigation. However, the occurrence of a variety of environmental contaminants in water, sediment, and biological tissues from Las Vegas Wash and Las Vegas Bay indicate that environmental conditions are not conducive to healthy populations of fish and wildlife. Contaminant accumulation, tissue abnormalities, and evidence of reproductive effects (endocrine disruption) have been identified in fish from Las Vegas Wash and Bay (Goodbred *et al.* 1999; Bevans *et al.* 1998, 1996; Patiño *et al.* in review).

In 1998, the U.S. Geological Survey (USGS) Biological Resources Division and the U.S. Fish and Wildlife Service (Service) initiated a joint investigation to assess contaminants associated with Las Vegas Wash and effects to wildlife. The investigation was expanded in 2000 to address concerns specific to DOI trust resources. The objectives of the Service's components of the investigation included:

- 1) reviewing existing data to assess the existence and degree of threats to fish and wildlife,
- 2) assessing organochlorine (OC) compounds and trace elements in avian eggs,

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- 3) assessing razorback sucker (*Xyrauchen texanus*) exposure to endocrine disrupting compounds and other contaminants, and
- 4) assessing the potential for downstream impacts to endangered fishes and critical habitat.

Field data collection for this joint investigation was completed in 2001. A portion of the data have not been fully analyzed by USGS and are not ready for release. This report provides the findings of the Service components of the investigation. A comprehensive report of the combined USGS-Service investigation is expected in 2003.

The following report is divided into five sections. The first section provides an overview of the study area and DOI trust resources in the vicinity. The second section provides a review of existing data to identify contaminant concerns and assess implications of contaminants to fish and wildlife. The third section evaluates contaminant concentrations in avian eggs to evaluate contaminant availability and the potential for impacts to birds. The fourth section evaluates contaminant exposure, accumulation, and effects in endangered razorback suckers. The final section evaluates the downstream transport of contaminants associated with Las Vegas Wash and the potential for these contaminants to adversely affect fish and wildlife downstream of Hoover Dam.

STUDY AREA

Lake Mead was created in 1935 by the construction of Hoover Dam and is located on the main stem of the Colorado River, Arizona-Nevada (Figure 1). Tributaries to Lake Mead include the Las Vegas Wash and the Colorado, Virgin, and Muddy rivers. Las Vegas Wash extends over 15 miles from the Las Vegas Metropolitan Area to Las Vegas Bay in Boulder Basin of Lake Mead. The Wash provides the sole route of surface and sub-surface water drainage from the Las Vegas Valley, some 2,200 square miles (Bureau of Reclamation 1982), and therefore, receives all point and non-point source pollution. Prior to development in the Las Vegas Valley, flow in Las Vegas Wash was perennial (Kilroy *et al.* 1997). However, extensive groundwater withdrawal in the early 20th century resulted in diminished flow in the Wash causing stream flow

to become ephemeral, with flows resulting only from precipitation and runoff events. Flows in the Las Vegas Wash again became perennial following the increased discharge of treated wastewater, groundwater flows, urban runoff from landscape irrigation, and industrial process water (Covay and Leiker 1998). Flow volume in the Wash has steadily increased with rapid population growth in the Las Vegas Metropolitan Area, averaging over 160 million gallons per day at Northshore Road bridge in 2001 (Bureau of Reclamation 2001). Currently, the population in the Las Vegas Metropolitan Area exceeds 1.5 million permanent residents and some 35 million tourists annually, generating over 150 million gallons per day (> 90 percent of the total flow during dry weather) of wastewater effluent discharges into Las Vegas Wash (Montgomery Watson 1997). This volume is expected to increase to more than 250 million gallons per day by 2020.

Changes in land use and coverage have contributed to large-scale erosion in Las Vegas Wash. Glancy and Whitney (1986) estimated a loss of as much as 4 square miles of sediment from Las Vegas Wash between 1969 and 1984. Channel down-cutting and sediment deposition have contributed to the extensive loss of wetlands associated with the Wash. Several local, State, and Federal agencies have initiated efforts to reduce erosion and restore wetland habitat along Las Vegas Wash.

Lake Mead National Recreation Area (NRA), administered by the National Park Service, receives annual visitation in excess of 27 million people and is one of the most heavily used National Park Service facilities in the nation. Recreational activities include swimming, boating, fishing, and other forms of water contact recreation. The Nevada Division of Wildlife manages a recreational fishery in Lake Mead. Principle game fishes include striped bass (*Morone saxatilis*), largemouth bass (*Micropterus salmoides*), and channel catfish (*Ictalurus punctatus*). Lake Mead also serves as the primary source for drinking water to southern Nevada.

Department of the Interior trust responsibilities include lands administered by the National Park Service (Lake Mead NRA), land along Las Vegas Wash administered by the Bureau of Reclamation, and limited lands administered by the Bureau of Land Management. Additionally, six federally-listed species as well as a variety of migratory birds have been identified in areas associated with Las Vegas Wash, Las Vegas Bay, and Boulder Basin of Lake Mead. Endangered species include razorback sucker, southwestern willow flycatcher

(*Empidonax traillii extimus*), Yuma clapper rail (*Rallus longirostris yumanensis*), and bonytail chub (*Gila elegans*). Threatened species include bald eagle (*Haliaeetus leucocephalus*) and desert tortoise (*Gopherus agassizii*).

Razorback suckers were historically abundant throughout the Colorado River and its major tributaries. However, declining numbers and the extirpation of populations throughout its range prompted the listing of this species as endangered in 1991. The primary reasons for the decline were identified as changes to biological and physical features of the habitat. The effects of these changes are most clearly seen in the virtual lack of natural recruitment to any population in the historic range of the species. Although razorback sucker are producing viable young, few individuals appear to survive to adulthood. Predation of larval razorback sucker by non-native fish is an important factor contributing to the decline of this species. Recent studies also indicate that environmental contaminants may result in poor recruitment of this species in some areas (Hamilton and Buhl 1997). Critical habitat was designated in 1994 and includes, among other areas, portions of the Colorado River from Pierce Ferry to Hoover Dam, including Lake Mead to its full pool elevation, and from Hoover Dam to Davis Dam, including Lake Mohave to its full pool elevation.

Ongoing research sponsored by the Southern Nevada Water Authority (SNWA) has added to the knowledge of razorback distribution and habits within Lake Mead (Holden *et al.* 1999). Among the findings are that two populations of razorback suckers occur in Lake Mead, one at Echo Bay on the Overton Arm and another in Las Vegas Bay. In 1997, Holden *et al.* (1997) estimated that 300 to 400 adult razorback suckers occupied Las Vegas Bay and 50 to 60 occupied Echo Bay. Holden *et al.* (2001) reduced their estimates to #100 adults for each population in 2000, based on additional data. Trapping and telemetry studies indicate that razorback suckers occur throughout Las Vegas Bay and use the mouth of Las Vegas Wash. Razorback suckers were also found in lower Las Vegas Wash upstream of Lake Mead. Larval sampling indicates that razorback suckers spawn in Las Vegas Bay in the vicinity of Blackbird Point.

Like the razorback sucker, bonytail chub were historically abundant in the Colorado River and its major tributaries but population numbers have declined throughout its range. Bonytail chub were listed as endangered in 1980. The cause of the decline was identified as

changes to biological and physical features of the habitat. The effects of these changes have been most noticeable by the almost complete lack of natural recruitment to any population in the historic range of the species. Populations are generally small and composed of aging individuals. Designated critical habitat includes, among other areas, the Colorado River from Hoover Dam to Davis Dam, including Lake Mohave to its full pool elevation. Although bonytail chub historically occurred in Lake Mead, this species has not been recently documented in Lake Mead and is now presumed extirpated.

Southwestern willow flycatcher historically occurred in southern California, Arizona, New Mexico, western Texas, southwestern Colorado, southern Utah, northwestern Mexico, and extreme southern Nevada (Unitt 1987). Declines in numbers across its range prompted the listing of this species in 1995. The southwestern willow flycatcher is a riparian obligate, nesting in riparian thickets associated with rivers, streams, and other wetlands where dense tree or shrub growth is present. Since 1998, flycatcher surveys have been conducted in the Las Vegas Wash. Results of these surveys have detected flycatchers in most years, but individuals have not been identified to the subspecies level because breeding activity has not yet been observed (SWCA 1998, 1999, 2000). The flycatcher is insectivorous, and forages within and above dense riparian vegetation, taking insects on the wing or gleaning them from foliage (Wheelock 1912, Bent 1960).

Within the United States, Yuma clapper rail are most frequently observed in extreme southern California and southwestern Arizona. This species was identified in Las Vegas Wash during a survey conducted in 1998. Clapper rail occur in marsh habitats and forage on crustaceans, small fish, insects, and limited vegetation in shallow water or mud flats.

The bald eagle was previously listed as endangered due to population declines across much of its range. Like peregrine falcons (*Falco peregrinus*), contaminants contributed to the decline of this species. Its status was upgraded to threatened in the lower 48 States in 1995. Bald eagles have been reported to migrate through, and winter along, the lower Colorado River. Fish are the preferred food, although carrion, blacktailed jackrabbits, and wounded or injured waterfowl will also be consumed.

The Mohave population of desert tortoise was emergency listed as endangered in 1989 and as threatened under a normal listing procedure in 1990. The desert tortoise, a large

herbivorous reptile, is generally found in desert scrub habitats throughout its range, which includes a portion of the Mojave Desert and the Colorado Desert subdivision of the Sonoran Desert. The desert tortoise has the potential to occur in upland habitat north of Las Vegas Wash. On February 8, 1994, the Service designated approximately 6.4 million acres of critical habitat for the Mojave population of the desert tortoise (59 FR 45748), which became effective on March 10, 1994. Approximately 1.2 million acres were designated as critical habitat in Nevada.

Lake Mead NRA provides habitat for over 240 species of migratory birds. More than 200 bird species use the Las Vegas Wash and Bay for nesting, loafing, foraging, and resting during migration (SNWA 2002). Species in the area include the great blue heron (*Ardea herodias*), great egret (*Ardea alba*), snowy egret (*Egretta thula*), eared grebe (*Podiceps nigricollis*), Western grebe (*Aechmophorus occidentalis*), Clark's grebe (*Aechmophorus clarkii*), gadwall (*Anas strepera*), American wigeon (*Anas americana*), mallard (*Anas platyrhynchos*), blue-winged teal (*Anas discors*), cinnamon teal (*Anas cyanoptera*), northern shoveler (*Anas clypeata*), northern pintail (*Anas acuta*), American coot (*Fulica americana*), red-winged blackbird (*Agelaius phoeniceus*), brewer's blackbird (*Euphagus cyanocephalus*), yellow-headed blackbird (*Xanthocephalus xanthocephalus*), marsh wren (*Cistothorus palustris*), double-crested cormorant (*Phalacrocorax auritus*), American avocet (*Recurvirostra americana*), killdeer (*Charadrius vociferus*), and black-necked stilt (*Himantopus mexicanus*). Restoration efforts along Las Vegas Wash and the subsequent creation of suitable habitat will likely increase the diversity and abundance of migratory birds that use this area, including the Yuma clapper rail and southwestern willow flycatcher.

REVIEW OF EXISTING INFORMATION

A variety of environmental contaminants have been identified at levels of concern in Las Vegas Wash and Bay including nutrients, pathogens (e.g., bacteria), trace metals, pesticides, pharmaceuticals, and industrial compounds. Primary concerns are discussed below.

NUTRIENTS

Nutrients in aquatic systems include a variety of chemical constituents which are necessary for growth and survival of aquatic organisms. Certain nutrients, particularly nitrogen and phosphorus compounds, limit primary productivity and, therefore, control limnological characteristics of the water body. Eutrophication, or the occurrence of excessive nutrients in surface waters, may result in high primary productivity in the water body which, in turn, may cause a variety of potentially deleterious effects such as dissolved oxygen depletion in the water column and sediments, increased pH, and changes in the biogeochemical cycling of chemical elements. Eutrophication is commonly associated with structural simplification and reduced stability of aquatic communities (Wetzel 1983).

Concerns for elevated nutrients in Las Vegas Wash water and detrimental impacts to Las Vegas Bay were expressed in 1971 in a U.S. Environmental Protection Agency (USEPA) report on pollution affecting Las Vegas Wash, Lake Mead, and the Lower Colorado River (USEPA 1971). Due to population growth and increasing wastewater flows, elevated nutrient concentrations and loading continue to affect water quality in Las Vegas Wash and Lake Mead. Bevans *et al.* (1998) reported that nutrient concentrations at two Las Vegas Wash sites exceeded the 75th percentile of sites investigated under the USGS National Water Quality Assessment (NAWQA) program nation-wide. The lower Las Vegas Wash site, which included treated sewage effluent, exceeded the 99th percentile. The greatest concern was with un-ionized ammonia, which exceeded the USEPA aquatic life criterion in all water samples collected from April 1993 to April 1995. Following full implementation of tertiary treatment of sewage effluent discharged to Las Vegas Wash in the mid 1990's, un-ionized ammonia and phosphorous concentrations have declined (LaBounty and Horn 1997). However, total nitrogen concentrations entering Las Vegas Bay have remained relatively constant following

implementation of tertiary treatment and nitrogen loading continues to increase (University of Nevada, Las Vegas [UNLV] 2002).

The input of nutrients and organic material from Las Vegas Wash has contributed to the eutrophication and subsequent high primary production in Las Vegas Bay (LaBounty and Horn 1997). Boulder Basin is a phosphorus limited system, hence the availability of phosphorus drives primary productivity, which is measured by determining chlorophyll-*a* concentrations. From 1991 to 1996, concentrations of chlorophyll *a*, a measure of algae biomass, in Las Vegas Bay near the Las Vegas Wash confluence ranged from <3 to 253 ug/L with annual means ranging from 16 to 34 ug/L (Nevada Division of Environmental Protection [NDEP] 1998). LaBounty and Horne (1997) reported chlorophyll *a* concentrations between 300 and 400 ug/L during algae bloom events in the inner Las Vegas Bay during 1993 and 1996. The mean chlorophyll *a* concentration in eutrophic lakes was reported as 14.3 ug/L by Wetzel (1983). There are currently no State-imposed water quality standards for chlorophyll *a*, only recommended limits between 5 ug/L in Boulder Basin and 45 ug/L in Las Vegas Bay to “maintain existing higher quality.” Drought conditions and dropping lake levels contributed to a massive green algae bloom throughout Boulder Basin in 2001, which persisted for over six months and resulted in recreational losses. Although phosphorus concentrations in the Las Vegas Wash have declined since dischargers incorporated enhanced phosphorus removal in 1995 (average of 0.5 mg/L and maximum of 2.3 mg/L total phosphorus in 2000), due to the large and ever increasing volume of effluent flows, phosphorus and nitrogen loading of Boulder Basin continues to increase and primary productivity remains high. Eutrophication of freshwater lakes is undesirable, causing an imbalance in primary productivity that can lead to more toxic forms of algae, often resulting in fish kills.

High primary productivity and the occurrence of high concentrations of organic material may result in the diurnal and seasonal depletion of dissolved oxygen in the water column. In highly productive systems, photosynthetic production of oxygen may result in supersaturation of dissolved oxygen concentrations during periods of sunlight (Wetzel 1983). However, high chemical and biological oxygen demand associated with high productivity may result in the depression of dissolved oxygen concentrations during hours of darkness. Dissolved oxygen levels in the effluent dominated waters of lower Las Vegas Wash were consistently between 8 -

10 mg/L in 2000. Monitoring data from Las Vegas Bay indicates that dissolved oxygen exceeded the saturation point in the bulk of the near surface daytime observations (maximum observed value 19.3 mg/L) with areas greatly deficient in oxygen developing at night (minimum observed value less than 0.2 mg/L)(SNWA 2001).

LaBounty and Horn (1997) noted a pattern in the seasonal dynamics of dissolved oxygen in Las Vegas Bay and Lake Mead. These researchers consistently detected the lowest dissolved oxygen concentrations in Las Vegas Bay near the confluence of Las Vegas Wash. As thermal stratification of the Lake developed during the spring, conditions of low dissolved oxygen developed in the hypolimnion. Anoxic conditions expanded farther into the main body of Boulder Basin as stratification intensified through the summer. Anoxic conditions persisted below the thermocline from July into October (Figure 2). Depressed dissolved oxygen concentrations were observed as far away as Hoover Dam. These researchers attributed the spatial and seasonal dissolved oxygen patterns to the introduction of nutrients and organic material from Las Vegas Wash. Concerns with seasonal depletion of dissolved oxygen persisted at the time of completion of this report.

Conditions of low dissolved oxygen in water (less than 4 or 5 mg/L) have the potential to cause adverse effects to aquatic organisms and habitats. In addition to causing suffocation, the toxicity of many contaminants increases as dissolved oxygen concentrations decrease (Lloyd 1961). Increased toxicity may be attributed to the movement of greater water volumes and associated waterborne contaminants across the gill surface when dissolved oxygen is depressed. Conditions of depressed dissolved oxygen persist in the hypolimnion in a significant portion of Las Vegas Bay throughout the summer. Because razorback suckers are a benthic-dwelling species, anoxic conditions in the hypolimnion are of particular concern. Anoxic conditions do not appear to coincide with razorback sucker spawning in Las Vegas Bay. However, razorback sucker adults have been documented in Las Vegas Bay during seasons of depressed dissolved oxygen (Holden *et al.* 1997). It is uncertain if juvenile razorback suckers continue to reside in Las Vegas Bay during these periods. Seasonally anoxic conditions would presumably continue to have an effect on invertebrate communities, food availability, and general habitat quality throughout the year.

High concentrations of certain nutrients may be toxic to aquatic organisms. Ammonia levels are of particular concern in Las Vegas Wash and Bay due to the presence of razorback suckers. In natural waters, ammonia exists in an ionized and an un-ionized form. Un-ionized ammonia is considered the more toxic form and is acutely toxic to most fish species at elevated concentrations (Russo 1985). Chronic exposure to lower concentrations of un-ionized ammonia may cause a variety of sublethal effects to aquatic organisms, including decreased growth, reduced fecundity, reduced numbers of red blood cells, kidney and gill degeneration, and reduced resistance to disease (Russo 1985). Additionally, ammonia may increase the toxicity of other contaminants such as copper and zinc. The proportion of un-ionized ammonia is controlled by pH and, to a lesser extent, temperature. As such, the current aquatic life criteria recommended by the USEPA are based on pH. The pH of Las Vegas Bay generally ranges between 8 and 9, which corresponds to recommended acute ammonia nitrogen criteria (Criteria Maximum Concentrations; CMC) between 1.3 and 8.4 mg/L and chronic criteria (Criteria Continuous Concentration; CCC) between 0.25 and 1.27 mg/L. The CMC should not be exceeded more than once every three years. The CCC is averaged over thirty days, and again should not be exceeded more than once every three years. The State of Nevada acute and chronic water quality standards for ammonia in Las Vegas Bay and Lake Mead are 0.45 mg/L NH₃ (single value not to be exceeded more than once every three years) and 0.05 mg/L (average not to be exceeded more than once every three years), respectively. There is no ammonia standard for the Las Vegas Wash although the State restricts ammonia loading during the months of April through September.

As indicated previously, ammonia in all Las Vegas Wash samples collected from 1992 - 1996 by Bevens *et al* (1998) from areas downstream of the sewage discharges exceeded the aquatic life criterion recommended by USEPA. Review of 1996 data (following implementation of tertiary treatment of sewage) indicates that ammonia concentrations have declined, and in all but one sample were less than the CCC. It is important to remember, however, that EPA's criterion (CMC and CCC) are derived to be protective to the range of taxa tested (i.e., protect 95 percent of all fish and aquatic invertebrate taxa). This protection is considered adequate for protecting populations of organisms but implies that a small percentage of a population may be affected (i.e., loss of individuals). When interpreted within the context of the Endangered

Species Act, this small effect to a population is considered unacceptable for a listed species since any further decline in the population may lead to its local extirpation or cumulative extinction across its range. Average ammonia values in 2000 were 0.18 mg/L for Las Vegas Wash, which exceeds chronic effect levels for many sensitive species of fish (USEPA 1984), and 0.08 mg/L for Las Vegas Bay, which exceeds the State chronic standard. Therefore, additional data are needed on the sensitivity of razorback suckers to ammonia before it may be concluded that ammonia no longer represents a threat to this species.

PATHOGENS

Total and fecal coliforms are regularly monitored in Las Vegas Wash and Bay and bacteriological concerns have been identified. Although coliform bacteria are considered harmless to humans, elevated levels of these bacteria are considered an indicator of environmental conditions conducive to the occurrence and proliferation of other pathogens, including a variety of potentially harmful bacteria, viruses, or parasites. Possible sources of fecal contamination include septic tanks, illegal dumping into storm drains, domestic animals, wildlife, and treated wastewater. Fecal coliform densities in the Las Vegas Wash increased with the onset of effluent dechlorination in 1993 (UNLV 2002), a step that was taken to reduce chlorine toxicity in wastewater flows. The UNLV (2002) study also concluded that the source of fecal contamination in Las Vegas Wash cannot be attributed entirely to wastewater, with the majority of enterococci bacteria comprised of avian related species (81 percent) versus human (18 percent). The State of Nevada has adopted a human water contact standard of 200 colony forming units (CFU) per 100 ml of water and a propagation of wildlife standard of 1,000 CFU/ml. The State beneficial use standard for Lake Mead and Las Vegas Wash are that fecal coliforms “must not exceed a log mean of 200 most probable number (MPN)/100mL (for > five samples in 30 days), nor exceed 400 MPN/100mL for a single sample maximum (Nevada Administrative Code 445A.195).” Several states have adopted standards of 1,000 CFU/100 ml for total coliform bacteria. Fecal coliform counts during wet and dry weather flows have regularly exceeded applicable standards in Las Vegas Wash (Figure 3). In 1996, bacterial counts exceeded 400 CFU/100ml in at least half of the water samples at monitoring stations in Las Vegas Wash (NDEP 1998). Log mean concentrations at all sites also exceeded this standard.

Eight of 38 samples collected near the confluence of Lake Mead exceeded 1,000 CFU/ml. Contamination extended to Las Vegas Bay, where 17 percent of the samples collected from the inner Las Vegas Bay in 1996 exceeded the 200 CFU/100 ml standard (NDEP 1998). Approximately 4 percent of the samples collected near the razorback spawning area exceeded the 200 CFU/100 ml standard. Fecal coliform counts between 1992 and 1998 appear to be highest in the Las Vegas Wash during infrequent storm events, averaging 250,000 to 500,000 MPN/100 ml, with maximum concentrations ranging up to 5,000,000 MPN/100 ml. As a result of elevated bacterial counts, water contact recreation in a portion of Las Vegas Bay within the National Park Service Lake Mead NRA is not permitted.

Pathogens represent a threat to both terrestrial and aquatic species associated with Las Vegas Wash and Bay. The manifestation of disease in fish and wildlife populations are dependent on a number of factors. Herman (1990) identified pathogenic organisms, predisposing environmental conditions, and susceptible hosts as primary factors influencing the susceptibility of fish populations to disease outbreaks. Friend (1985) identified transmission, threshold levels, and susceptible hosts as key factors influencing the susceptibility of wildlife populations to diseases. Friend noted that contaminated water is an efficient vehicle for transmission of many diseases to wildlife. The occurrence of high concentrations of total and fecal coliforms in Las Vegas Wash and Las Vegas Bay near the confluence indicates that conditions are conducive to the survival and proliferation of pathogenic organisms. It is uncertain if threshold concentrations are exceeded. Stressful environmental conditions such as low dissolved oxygen and elevated salinity may increase the susceptibility of the host to infection by pathogenic organisms (Friend 1985). Similarly, a variety of chemical pollutants, such as un-ionized ammonia, several trace elements, pesticides, and industrial compounds have been shown to compromise immune response in fish and wildlife, thereby making affected individuals more susceptible to infection (Friend 1985).

Additional information is needed to adequately assess the implications of pathogens to fish and wildlife associated with Las Vegas Wash and Bay. The USGS is currently assessing the incidence of disease in common carp (*Cyprinus carpio*). Results of their investigation are expected to be released in 2003.

TRACE METALS

Concerns with a variety of metals and other trace elements have been identified in Las Vegas Wash. Few data are available for Las Vegas Bay; however, metal concentrations in Bay water should be comparatively lower than in Las Vegas Wash due to dilution. Water quality monitoring in Las Vegas Wash by the USGS (2001), NDEP (2000), and SNWA (2001) indicate regular exceedances of standards for iron, mercury, molybdenum, and selenium, with less frequent exceedances of standards for boron, lead, and cadmium. Numerical standards for some trace elements have not been revised since 1985 (or earlier) and are considered by many to be outdated and possibly inadequate for protection of sensitive species. USEPA issued a Notice of Intent in 1999 to revise aquatic life criteria for copper, silver, lead, cadmium, iron and selenium. The new criterion, when released, may be more stringent (e.g., lower values for chronic criteria) due to advances in the field of toxicology. At the time of preparation of this report the revision process was still ongoing, with the exception of cadmium, which was updated in 2001.

Aquatic life standards for cadmium, chromium, copper, lead, nickel, silver, and zinc are based on water hardness. Water hardness in Las Vegas Wash is high, ranging from 800 to over 1300 mg/L, which allows metal concentrations in water to be higher without exceeding water quality standards. For example, the CCC for cadmium is 1.07 ug/L at a hardness of 700 mg/L, but only 0.25 ug/L at a hardness of 100 mg/L. Regardless of water hardness, many trace metals are known to accumulate in sediments and biota and may adversely affect wildlife health in the long-term. Evaluation of water quality data compared to effect levels in fish and wildlife and standards for protection of aquatic life revealed concerns with several elements. In the following discussion, our interpretation of selenium and mercury data was hampered where sample concentrations were listed as non-detect but the detection limit substantially exceeded levels known to adversely affect fish and wildlife.

Arsenic, at a concentration of 0.04 mg/L in water, has been associated with adverse effects to amphibian embryos and larvae (USEPA 1985a) and low level toxicity to fish (Birge *et al.* 1980). Arsenic was detected in 60 percent (49 of 82) of the water samples from Las Vegas Wash. Sixteen percent of the samples exceeded the 0.04 mg/L effect level. The highest concentration was 0.12 mg/L.

Copper may be lethal or cause teratogenic effects to fish and amphibian eggs at concentrations in water between 0.005 and 0.01 mg/L (Eisler 1997). Copper concentrations met or exceeded 0.01 mg/L in more than half (66 of 127) of the Las Vegas Wash water samples reported by SNWA (Figure 4). The highest reported concentration was 0.270 mg/L.

Iron is considered an essential element for most organisms in small quantities but may be toxic at higher concentrations. The USEPA and State chronic standard for iron is 1.0 mg/L in water. Iron concentrations in Las Vegas Wash frequently exceed this standard, ranging from 1.0 to over 13.0 mg/L in 1999 and 2000 (NDEP 2000).

Lead may cause low level toxicity in fish at concentrations of 0.003 mg/L with substantial toxicity at a concentration of 0.11 mg/L (Birge *et al.* 1980). Reproduction of *Daphnia magna* was impaired 10 percent at a concentration of 0.001 mg/L and 50 percent at a concentration of 0.01 mg/L (USEPA 1985b). The USEPA chronic criterion for lead to protect aquatic life in Las Vegas Wash is 0.019 mg/L and the State chronic standard is 0.009 mg/L. The highest of four NDEP grab samples collected in 1999 and 2000 was 0.018 mg/L. More than 40 percent (42 of 102) of SNWA water samples exceeded 0.01 mg/L (Figure 4). Eight samples exceeded 0.11 mg/L. The highest reported lead concentration was 0.28 mg/L.

Mercury in water is toxic to aquatic invertebrates at concentrations as low as 0.002 mg/L for inorganic mercury or as low as 0.0009 mg/L for organic forms (USEPA 1980). Mercury has been shown to cause adverse effects to salmonids at concentrations below 0.0001 mg/L (Birge *et al.* 1980) and warm water fishes at a concentration as low as 0.0005 mg/L (Snarski and Olson 1982). Mercury also strongly accumulates in organisms and magnifies in food chains. Schwarzbach (1998) identified a concentration of 0.00000064 mg/L in water as a criterion for the protection of piscivorous birds. The USEPA and State chronic criterion for mercury to protect aquatic life is 0.000012 mg/L. Mercury concentrations in Las Vegas Wash exceed this standard and are routinely 0.0005 to 0.001 mg/L. The highest concentration was 0.006 mg/L.

Molybdenum may be acutely toxic to larval fish at concentrations as low as 0.028 mg/L (Birge *et al.* 1980). The State chronic standard for protection of aquatic life is 0.019 mg/L. Molybdenum concentrations in Las Vegas Wash ranged from 0.01 to 0.395 mg/L and averaged 0.195 between 1995 and 2000.

Nickel may cause mortality of sensitive fish species (salmonids) at concentrations as low as 0.011 mg/L (Birge *et al.* 1980). Eisler (1998) noted that toxic effects of nickel may be expected in salmonids at concentrations ranging from 0.03 to 0.05 mg/L, and recommended a safe level of less than 0.029 mg/L for salmonids. The USEPA chronic criterion for nickel is 0.27 mg/L. In Las Vegas Wash, nickel concentrations averaged 0.125 mg/L in water samples reported by SNWA from 1997 to 1999. The highest concentration was 0.25 mg/L.

Selenium is strongly bioaccumulated in aquatic organisms and relatively low concentrations in water can quickly become concentrated to potentially toxic levels in aquatic organisms and wetland birds (Lemly 1996). Toxic endpoints commonly include histopathology, physiological effects, reduced reproduction, and reduced survival. Selenium is recognized as a powerful teratogen and reproduction is generally considered the most sensitive and significant toxic endpoint. Waterborne concentrations in the low part per billion range can lead to reproductive failure in adult fish with little or no additional symptoms of selenium poisoning in the environment (Lemly 1996). Maier and Knight (1994) summarized seven recent studies that provided estimates of waterborne thresholds for food chain-mediated toxicity to fish and wildlife. All reports concluded that toxic thresholds were less than or equal to 0.003 mg/L in water. Selenium has been identified as a concern in the lower Colorado River and may affect reproductive success of native Colorado River fishes (Hamilton 1999). Anthropogenic sources, particularly agricultural drainage, were identified as the primary cause of elevated concentrations in the lower basin. Las Vegas Wash monitoring data indicate that the Wash is a selenium source to the Colorado River. The USEPA and State chronic standard for selenium is 0.005 mg/L. Average selenium concentrations in Las Vegas Wash exceed these standards, falling between 0.007 and 0.010 mg/L. The highest observed concentration in 1995 was 0.1 mg/L.

Zinc has been found to be acutely toxic to aquatic invertebrates at a concentration as low as 0.032 mg/L in water (USEPA 1987). Lethal and sublethal effects have been found in sensitive fish (salmonids) at concentrations of less than 0.01 mg/L (Eisler 1993). The most restrictive chronic criterion for zinc in Las Vegas Wash is 0.47 mg/L imposed by the State of Nevada. Over 80 percent of the samples reported by SNWA exceeded a 0.02 mg/L detection

limit (Figure 4). Average zinc concentrations in lower Las Vegas Wash ranged from 0.07 to 0.09 mg/L with a high of 0.13 mg/L (SNWA 2001).

The USGS computed an urban trace element index (UTEI) for seven sites in Las Vegas Wash and Bay (Bevans *et al.* 1998). Index values were determined by calculating the ratio of concentrations of cadmium, chromium, copper, lead, and zinc at each site to corresponding concentrations at a background site. Ratios were then summed to provide a single UTEI value. The UTEI values at sites in lower Las Vegas Wash and near the mouth of Las Vegas Wash were 1.5 to 2 times higher than the background site.

The toxicity of trace elements to birds may be modified in the presence of other environmental contaminants. As indicated previously, the toxicity of copper and zinc may be enhanced in the presence of ammonia (Russo 1985). Copper toxicity to aquatic organisms may also be enhanced in the presence of zinc. Finlayson and Ashuckian (1979) concluded that safe levels of copper and zinc were 0.011 and 0.083 mg/L, respectively. Mixtures of arsenic, boron, copper, selenium, and zinc in water have been reported to be toxic to razorback sucker larvae (Hamilton and Buhl 1997). Toxicities of the mixtures were found to be greater than the toxicity of any single element. Concentrations of arsenic, boron, copper, selenium, and zinc in Las Vegas Wash water exceeded or were commonly within the range of concentrations where risk to razorback sucker larvae was predicted by Hamilton and Buhl (1997).

PESTICIDES

The U.S. Geological Survey detected 24 pesticides in water samples collected in Las Vegas Wash downstream of Las Vegas (Bevans *et al.* 1998). The pesticides most commonly detected were simazine, prometon, diuron, dacthal, diazinon, and malathion. Diazinon and malathion exceeded aquatic life criteria in 47 and 25 percent of the samples, respectively. Carbaryl and chlorpyrifos concentrations exceeded levels recommended for protection of aquatic life by the National Academy of Sciences (1973). The highest pesticide concentrations were found in Las Vegas Wash upstream of the sewage treatment plant discharges. Concentrations at this site were greater than the 75th percentile of sites investigated by the USGS under the National Water Quality Program.

In 1996, the USGS used semipermeable membrane devices (SPMDs) to sample organic contaminants in water in Las Vegas Wash, Las Vegas Bay, and a background site (Callville Bay) in Lake Mead (Bevans *et al* 1996). Semipermeable membrane devices sequester lipid-soluble contaminants from the water column, thus allowing time-integrated sampling of a variety of organic contaminants. While this sampling technology allows for the detection of contaminants at low concentrations and provides an accurate representation of the biological availability of lipid-soluble compounds, it generally does not allow for the accurate assessment of exceedances of water quality standards. Bevans *et al.* (1996) identified a variety of OC and semivolatile industrial compounds in water, bottom sediments, and carp in Las Vegas Wash and Las Vegas Bay. The number and combined concentrations of OC compounds were highest in Las Vegas Wash, followed by Las Vegas Bay (Bevans *et al.* 1996). The compounds hexachlorobenzene, dacthal, trans-chlordane, cis-chlordane, trans-nonachlor, dieldrin, *p,p'*-DDE, *p,p'*-DDD, and total polychlorinated biphenyls (PCBs) were detected at all sample sites in Las Vegas Wash and Bay. The number of compounds and concentrations were lower at Calville Bay, the background site. Organochlorine compounds were also detected in sediment and carp from Las Vegas Wash and Bay. Organochlorine compounds were not detected in sediment from the background site. The number of OC compounds and concentrations of both total OC and total DDT were significantly higher in carp from Las Vegas Bay than in carp from the background site. Median concentrations of total DDT (DDT and its metabolites DDD and DDE) in cross-sectional carp tissue from Las Vegas Wash and Bay (72 µg/kg and 120 µg/kg, respectively) fell within levels of concern for at-risk groups based on human consumption guidelines (USEPA 2000). At present, the Nevada State Health Department and NDEP are considering whether a human health consumption advisory is warranted for OC compounds and other contaminants in game fish from Lake Mead.

PERCHLORATE

Perchlorate, a chemical used since the 1940s in solid rocket fuels, lubricating oils, and fireworks, has been detected in water from Las Vegas Wash, Las Vegas Bay, and the lower Colorado River. The source of perchlorate has been identified as an industrial site operated by Kerr-McGee Chemical Company located in Henderson, Nevada near Las Vegas Wash. The

perchlorate anion is highly soluble, extremely stable, and may persist in water for decades. Perchlorates may affect wildlife health by preventing assimilation of iodine and interfering with thyroid functioning. The potential biological and ecological effects of perchlorate toxicity in wildlife are still being investigated by the scientific community. Goleman *et al.* (2002) found levels as low as 0.059 mg/L ammonium perchlorate inhibited thyroid activity and altered sex ratios in the South African frog (*Xenopus laevis*), significantly reducing the percentage of males at metamorphosis. The mode of action of perchlorate in biological systems raises concerns for possible carcinogenic, neurodevelopmental, developmental, reproductive, and immunotoxic effects. Concern for perchlorate in water has prompted the State of California to establish a human health standard of 0.018 mg/L in drinking water. USEPA recently concluded that perchlorate may be harmful at lower levels than previously thought and is considering lowering the perchlorate level deemed safe in drinking water from 0.018 to 0.001 mg/L. Safe levels for fish and wildlife are uncertain. Perchlorate levels in Las Vegas Valley drinking water (i.e., tap water) are routinely 0.010 to 0.015 mg/L and reached a high of 0.030 mg/L in December of 2001, when limnological conditions were not favorable for mixing and Las Vegas Wash flows remained relatively intact within Boulder Basin. Sampling by the Bureau of Reclamation in 1998 revealed perchlorate contamination in Las Vegas Bay near the Las Vegas Wash confluence, with concentrations ranging from 0.65 to more than 1 mg/L. Recent concentrations in Las Vegas Bay ranged from 0.020 to 0.110 mg/L (SNWA 2001). Water quality data in 2002 ranged from 0.25 to 0.45 mg/L at the confluence of the Las Vegas Wash, indicating that perchlorate concentrations may be on the decline, possibly due to ongoing remediation efforts by NDEP and Kerr-McGee.

POLYCYCLIC AROMATIC HYDROCARBONS

Polycyclic aromatic hydrocarbons (PAHs) and phthalates were detected in water and sediment from Las Vegas Wash and Las Vegas and Callville Bays (Bevans *et al.* 1996). Combined concentrations of these compounds were an order of magnitude greater in sediment from Las Vegas Wash and Bay than from the background site (Bevans *et al.* 1996). The areal distribution of PAH compounds in Las Vegas Bay indicated that Las Vegas Wash was the source of at least some of these compounds in Lake Mead, but there were also more general sources in

Lake Mead. Covay and Leiker (1998) found highly elevated concentrations of a variety of PAH compounds in the upper Las Vegas Wash, which drains the Las Vegas Metropolitan Area, suggesting that municipal runoff is a source of PAH to Las Vegas Wash.

DIOXINS AND FURANS

Bevans *et al.* (1996) found that combined concentrations of tetrachlorodibenzo-*p*-dioxins (TCDD) in SPMDs and sediment from Las Vegas Wash and Bay were two orders of magnitude higher than the background site (i.e., Callville Bay). Again, the distribution of dioxins and furans in Las Vegas Bay indicate that Las Vegas Wash is the source of these compound in Las Vegas Bay. Bioassays using the tissues from carp confirmed the occurrence of dioxins and furans with low toxic equivalent factors relative to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin, thereby confirming biological exposure to these compounds.

ENDOCRINE DISRUPTING CHEMICALS

A variety of endocrine disrupting chemicals (EDCs) have been identified in Las Vegas Wash and Bay. An EDC, or endocrine disruptor, has been defined as “an exogenous chemical substance or mixture that alters the structure or function(s) of the endocrine system and causes adverse effects at the level of the organism, its progeny, populations, or subpopulations of organisms ...” (USEPA 1997). The mode of action of EDCs may include interference with the synthesis, secretion, transport, binding, or elimination of natural hormones in the body. In this manner, EDCs have the potential to compromise normal reproduction, development, growth, and homeostasis. Known EDCs include a variety of pesticides, industrial compounds, and pharmaceuticals and personal care products (PPCPs). An increasing number of studies have linked the occurrence of EDCs in the environment with biochemical and physiological changes and a variety of behavioral, reproductive, developmental, and immune system effects in invertebrates, fish, amphibians, reptiles, birds, and mammals (USEPA 1997). Effects are often subtle and may occur at extremely low concentrations (Daughton and Ternes 1999). Several of these studies linked the occurrence of effects with EDCs in effluents discharged from municipal wastewater treatment plants.

A variety of known EDCs, including a number of pesticides, industrial compounds, and PPCPs have been identified in water, sediment, fish blood plasma, and tissues collected from Las Vegas Wash and Bay. Bevens *et al.* (1996) found that water, sediment, and carp tissue from Las Vegas Wash and Bay contained elevated concentrations of a variety of known or suspected EDCs, including OC compounds, PAHs, phthalates, phenols, dioxins, and furans. Goodbred *et al.* (1999) reported ethynylestradiol, a synthetic estradiol typically found in oral contraceptives, in blood plasma of fish from Las Vegas Bay. Snyder *et al.* (1999) detected nonylphenol, octylphenol, 17 β -estradiol, and ethynylestradiol in water samples from Las Vegas Wash and Las Vegas Bay. Monitoring by several entities has documented elevated perchlorate concentrations in water from Las Vegas Wash and Bay. Perchlorate is known to affect thyroid function in humans and implications to endocrine system effects in wildlife are still under investigation.

Assessment of EDCs in biological samples from Lake Mead is continuing under the direction of Tom Lieker of the USGS National Water Quality Laboratory, Denver, Colorado. Several known or suspected EDCs have been identified in blood plasma and tissues of carp, largemouth bass, and razorback sucker from Lake Mead (Tom Lieker, USGS, pers. comm., 2002). The number of identified compounds and concentration of most compounds of concern is substantially higher in samples from Las Vegas Bay as compared to other sites in Lake Mead. Results of this investigation are expected to be released in 2003.

ASSESSMENT OF CONTAMINANTS IN AVIAN EGGS

INTRODUCTION

A diverse assemblage of migratory birds is associated with Las Vegas Wash and Bay. The number and diversity of birds using this area is expected to increase as a result of efforts to develop wetlands along Las Vegas Wash. However, several contaminants identified in this area have the potential to adversely affect avian reproduction and survival. This component of the study was initiated to evaluate the potential for OC compounds and trace elements and trace elements to adversely affect avian reproduction.

METHODS

Eggs of three and possibly four migratory bird species representing two feeding guilds (piscivorous and insectivorous) were collected in the spring of 1999 for OC, metal, and trace element and metals analyses. For piscivorous species, seven western grebe (*Aechmophorus occidentalis*) and/or Clark's grebe (*A. clarkii*) eggs were collected from Las Vegas Bay near the confluence of Las Vegas Wash. Eggs and nests of both species are similar in appearance and adult grebes were not on the nests at the time of egg collection. Therefore, it is uncertain whether the eggs were laid by western or Clark's grebes or both. A suitable background site for grebes was not located in the southern Nevada area. Therefore, no eggs from a background site were collected. For insectivorous birds, six cliff swallow (*Hirundo pyrrhonota*) eggs were collected from nests beneath the bridge over Las Vegas Wash on State Highway 147 and two barn swallow (*H. rustica*) eggs were collected from the Las Vegas Wash outflow structure on Lake Las Vegas Dam. Ten cliff swallow eggs were collected from nests beneath a bridge on State Highway 169 near Overton, Nevada to represent a background site.

Eggs were collected by hand, placed in a chemically cleaned jars, stored on ice in the field, and later opened using pre-cleaned stainless steel instruments in the laboratory. Embryos were inspected for gross abnormalities and egg contents were placed in 60 ml acid-washed glass containers and then frozen. Because of their small sizes, two swallow eggs from the same nest

were composited for each of the swallow egg samples. Each grebe egg was treated as a discrete sample.

Samples were submitted to designated laboratories under contract with the Service's Patuxent Analytical Control Facility (PACF) at Laurel, Maryland, for trace element and OC analyses. The OC scan was conducted using a gas-liquid chromatograph and included the following compounds: HCB, total PCBs, *alpha* BHC, *alpha* chlordane, *beta* BHC, dieldrin, endrin, *gamma* BHC, *gamma* chlordane, heptachlor epoxide, mirex, *o,p'*-DDD, *o,p'*-DDE, *o,p'*-DDT, oxychlordane, *p,p'*-DDD, *p,p'*-DDE, *p,p'*-DDT, toxaphene, and *trans*-nonachlor. The trace element scan included the following elements: aluminum, arsenic, boron, barium, beryllium, cadmium, chromium, copper, iron, mercury, magnesium, manganese, molybdenum, nickel, lead, selenium, strontium, vanadium, and zinc. Trace element analysis for arsenic and selenium was conducted using atomic absorption spectroscopy, mercury using cold vapor atomic absorption spectroscopy, and the remaining trace elements using inductively coupled plasma emission spectroscopy. Quality assurance and quality control of the chemical analysis were approved by PACF.

The toxicity and potential for OCs and trace elements to adversely affect migratory birds varies by compound and by species. Birds may respond differently to certain contaminants and doses. Species-specific differences among birds to contaminants are often reported in the scientific literature. Avian toxicity and effects on short and long term health are not well understood, particularly when wildlife receptors are exposed to complex mixtures of xenobiotics, as is the case for Las Vegas Wash. Thus, our review of avian contaminant effect studies should be viewed with caution. Contaminant related effects reported in other bird species may not necessarily be exhibited by western grebes, Clark's grebes, cliff swallows, or barn swallows.

RESULTS AND DISCUSSION

Several OCs were detected in grebe eggs collected from Las Vegas Bay and swallow eggs collected from Las Vegas Wash and Overton, Nevada. Principal compounds detected included DDT metabolites DDD and DDE, PCB compounds, chlordane compounds, and benzene hexachloride (BHC) (Figure 5). The number of detected OCs (15, 13, and 11 for Las

Vegas Bay grebes, Las Vegas Wash swallow, and Overton swallows, respectively) and total OC concentrations (Figure 6) were significantly higher in swallow eggs from Las Vegas Wash compared to the Overton reference site. DDT and its metabolites DDD and DDE (collectively termed total DDT) accounted for the majority of the total OC burden in all but one case in eggs from Las Vegas Wash and Bay. Principal compounds in Overton eggs included DDT metabolites and chlordane compounds.

The potential for DDT to cause the thinning of avian egg shells and, subsequently, a reduction in reproductive success in birds is well documented (Blus 1996). Eggshell thinning is primarily attributed to the metabolite, DDE. Sensitive species may experience significant reproductive declines with DDE egg residues as low as 3 ppm, wet weight (ww). Lindvall and Low (1980) did not find apparent effects to productivity in western grebes when concentrations in eggs averaged 6.6 ppm, ww. DDE concentrations in grebe eggs from Las Vegas Bay ranged from 1.3 to 13.9 ppm, ww with a mean of 7.1 ppm, ww (Figure 7). Relationships were not apparent between DDE or total DDT concentrations and egg shell thickness or Ratliffe index values for grebe eggs. DDE ranged from 2.1 to 8.6 ppm, ww (O = 5.2 ppm) in swallow eggs from Las Vegas Wash and from 0.2 to 2.1 ppm, ww (O = 0.8 ppm) in swallow eggs from Overton. Swallow egg shells were too small to provide accurate measurement of thickness with the instrumentation available.

Hoffman *et al.* (1996) identified an avian egg PCB concentration of 1 ppm, ww as a threshold for sensitive birds. Primary effects included reduced hatching. PCB concentrations in grebe eggs from Las Vegas Bay ranged from 1.5 to 6.6 ppm, ww (O = 3.4 ppm). Concentrations in swallow eggs from Las Vegas Wash ranged 0.9 to 1.2 ppm, ww (Figure 7). PCB concentrations in three of five samples from Overton were less than the 0.05 ppm detection limit. The highest concentration was 0.83 ppm, ww.

Hexachlorobenzene, dieldrin, and mirex concentrations were lower than known effect levels in avian eggs (Wiemeyer 1996, Peakall 1996). Information on egg residues of chlordane compounds and effects to reproduction are lacking (Wiemeyer 1996). Toxaphene was less than the lower analytical detection limit (0.05 ppm, ww) in all eggs sampled.

Due to the smaller size of swallow eggs and inadequate sample volume following OC analyses, trace element residue analyses for avian eggs could only be performed on grebe

samples. As stated previously, a suitable background (i.e., control) site for grebes was not located, therefore, interpretation of elemental residues in grebe eggs was limited to comparison with toxic thresholds in published literature.

Aluminum, arsenic, barium, beryllium, cadmium, lead, molybdenum, and vanadium were less than the analytical detection limits for all samples (Table 1). Boron (O = 0.321 ppm ww), chromium (O = 0.304), copper (O = 0.518), iron (O = 22.7), magnesium (O = 75.8), manganese (O = 0.304), mercury (O = 0.0249*), nickel³ (O = 0.044*), selenium (O = 0.39), strontium (O = 1.16), and zinc (O = 7.94) were detected but concentrations were less than known biological effect levels. Chromium residues in grebe eggs ranged from 0.246 to 0.417 ppm, ww which is within the range found in other avian species reported by Burger *et al.* (1999) and is not indicative of chromium contamination (i.e., not \geq 4.0 ppm) as described by Eisler (1986). Copper ranged from 0.383 to 0.678 ppm, ww and did not appear elevated compared to other bird studies (Hui *et al.* 1998; de Moreno *et al.* 1997; Morera *et al.* 1997). Likewise, selenium concentrations ranged from 0.259 to 0.495 ppm, ww and were lower than contaminant threshold concentrations reported by Skorupa and Ohlendorf (1991). Mercury concentrations ranged from 0.0219 to 0.0757 ppm, ww. Zillioux and others (1993) estimated that the significant toxic effects threshold concentration for mercury in most aquatic bird eggs was between 1.0 and 3.6 $\mu\text{g/g}$, ww. Observed mercury values were also well below safe levels reported by Eisler (1987) and Heinz (1979). Typically, iron, magnesium, manganese, and strontium are not ecological contaminants of concern and will not be discussed in this report.

CONCLUSIONS

The occurrence of OC compounds in avian eggs confirms that migratory birds utilizing the Las Vegas Bay are exposed to these contaminants. DDT metabolites and PCBs occur at levels that have the potential to affect embryo and hatchling survival. Although grebes are known to occupy Las Vegas Bay waters as year-round residents, there is inherently some degree

* indicates that more than half of the samples were below the lower detection limit. Values below the detection limit were halved to determine average concentrations (O).

of uncertainty when collecting avian samples as to whether contaminant concentrations reflect local conditions (i.e., whether or not exposure is from a local source). However, the occurrence of OCs in environmental media in Las Vegas Wash and Bay combined with higher concentrations in swallow eggs from Las Vegas Wash than from Overton suggests that Las Vegas Wash is a source of DDT metabolites and PCBs. In addition, the concentration of certain analytes in avian eggs (e.g., selenium) has been shown to strongly reflect local conditions due to the resident time needed for courtship, embryo development, and nesting activities (Joseph Skorupa, Service, pers. comm., 2002). Additional research is recommended to determine if OCs are affecting avian productivity in Las Vegas Wash and Bay.

Trace elements do not appear to be at levels of concern in grebe eggs. However, trace element concentrations in water, sediment, and biota from Las Vegas Bay are consistently lower than Las Vegas Wash due to dilution. Grebes prefer open water habitat and would presumably reflect conditions in Las Vegas Bay. Also, the number of samples analyzed for trace elements was small ($n = 7$) and should be verified through additional sampling to ensure concentrations are representative of the population as a whole.

A cooperative effort between the Service and SNWA to monitor trace element and organochlorine residues in bird eggs along Las Vegas Wash is planned for the spring of 2003. The monitoring will provide additional information needed to better assess risks to birds utilizing the Las Vegas Wash and its tributaries. Monitoring activities along Las Vegas Wash may be expanded in the future to include measures of avian reproductive success (e.g., nest success).

ASSESSMENT OF CONTAMINANT EXPOSURE AND EFFECTS IN LAKE MEAD RAZORBACK SUCKERS

INTRODUCTION

This portion of the study was initiated to determine exposure of adult razorback suckers to environmental contaminants in Las Vegas Wash and Lake Mead and to evaluate implications to survival and reproduction. Although active razorback sucker spawning, as evidenced by the presence of eggs and larvae, has been documented within each of two populations in Lake Mead, recruitment of juveniles is rare. The contribution of contaminants to declines in reproduction, recruitment, and adult survival is uncertain when compared with other factors such as habitat modification and non-native predation.

METHODS

Two frozen whole razorback suckers were opportunistically obtained due to mortalities from a telemetry study by a private consultant using implanted sonic tags in Lake Mead. One fish was a 588 mm (total length), 1,940 g male between 9 and 10 years of age collected at Blackbird Point in Las Vegas Bay. The other was a 381 mm, 575 g, 4 to 5 year old juvenile collected from Echo Bay. Both fish were submitted to PACF, Laurel, Maryland for determination of trace element and OC compounds in whole-body fish.

Hormone concentrations and ratios, contaminants in blood plasma, and sperm quality were assessed to evaluate endocrine system effects in razorback suckers. Trammel nets were used to collect a total of 44 male and 17 female razorback suckers from Lake Mead. Fish were collected between February and March of 2000 and 2001. Net success at each site over the two year study were 24 male and 8 female at Las Vegas Bay and 20 male and 9 female from Echo Bay. At the time of preparation of this report, evaluation of 2001 samples was in progress and will therefore be included in the joint USGS-Service investigation report to be released in 2003. The following discussion will include 2000 results only, consisting of 11 males and 1 female from Las Vegas Bay, and 10 male and 4 female from Echo Bay.

Echo Bay served as the reference site for the razorback sucker contaminant assessment portion of this study. Echo Bay receives naturally occurring runoff (infrequently) from an unpopulated region near the Overton Arm of Lake Mead and is therefore considered an ideal location for sampling background contaminant levels in razorback sucker. Razorback suckers were weighed, measured, and visually inspected for external lesions, parasites, and other abnormalities then released. All razorback suckers were implanted with passive integrated transponder (PIT) tags to prevent resampling of same fish and to assist in ongoing population studies sponsored by the SNWA. Approximately 3 cubic centimeters (cc) of whole blood was collected from the caudal peduncle of each fish and placed in lithium-heparinized vacutainer tubes. The blood samples were centrifuged at 10,000 RPM for 10 minutes, blood plasma was separated from packed cells with a pipet, and frozen on dry ice. Plasma samples were submitted to the Florida Caribbean Science Center for analysis of 17 β -estradiol, 11-ketotestosterone, ethynyl-estradiol, and vitellogenin. Analyses followed methods described in Goodbred *et al.* (1997).

An aliquot of whole-body razorback sucker homogenate and remaining razorback sucker blood plasma were also provided to Tom Leiker of the USGS National Water Quality Laboratory for assessment of non-conventional pollutants (e.g., pharmaceuticals). These analyses were in progress at the time of preparation of this report. Results will be included in the joint investigation report to be released in 2003.

To evaluate exposure to genotoxic compounds, two drops of whole blood from each razorback sucker captured in 2001 were frozen on liquid nitrogen and submitted to the University of California, Davis, Bodega Marine Laboratory for assessment of DNA adducts.

RESULTS AND DISCUSSION

Two populations of razorback suckers occur in Lake Mead, one at Las Vegas Bay and another at Echo Bay (Holden *et al.* 1999). Fish tagged at one location have not been captured at the other location, suggesting that there is little, if any, intermixing between the two populations. Water quality monitoring has demonstrated that a variety of contaminants occur in Las Vegas

Wash. Differences in temperature and total dissolved solids discourage mixing of Las Vegas Wash water with Lake Mead water (LaBounty and Horn 1997). As a result, Las Vegas Wash water intrudes into Lake Mead and maintains a distinct temperature, conductivity, and presumably chemical signature some distance into Lake Mead. Therefore, aquatic species residing in Las Vegas Bay and portions of Lake Mead may be exposed to chemical and biological (e.g., pathogens) contaminants at concentrations occurring in Las Vegas Wash. The depth of the intrusion and the distance that it extends into Lake Mead depend on seasonal conditions and are predictable seasonally. From February to late May, when razorback sucker adults, eggs, and larvae would be expected to be present near the spawning area in Las Vegas Bay, the intrusion occurs at a depth of 20 to 30 meters and intrudes well into the lake. In April, the intrusion was recorded at least 8 km into the lake. Blackbird Point, a documented spawning location for razorback suckers in Lake Mead, occurs approximately 4 km from the confluence of Las Vegas Wash and Bay. Depending on lake elevation, spawning in Las Vegas Bay may occur at depths between 10 and 25 meters.

As reported earlier, elevated concentrations of a variety of contaminants have been reported in Las Vegas Wash water. Concentrations were generally higher in the upper reaches of Las Vegas Wash but concentrations associated with both acute and chronic toxicity to aquatic organisms were identified in the lower reaches of the Wash. Concentrations of several inorganic contaminants were similar to, or exceeded concentrations found to adversely affect razorback sucker reproduction and recruitment (Hamilton and Buhl 1997). The intrusion of Las Vegas Wash water into Lake Mead (LaBounty and Horn 1997) increases the possibility that potentially toxic trace element concentrations occur at the area where razorback suckers spawn in Las Vegas Bay. Fish are more sensitive to the effects of certain contaminants during early developmental life stages.

With the exception of selenium, concentrations of potentially toxic metals and trace elements did not exceed concern levels in tissues of either of the two analyzed razorback suckers. Arsenic, boron, beryllium, cadmium, mercury, and molybdenum were less than the analytical detection limits. Chromium, copper, nickel, lead, and zinc were detected at one or both sites, but concentrations were less than known biological effect levels. Selenium concentrations were 6.2 $\mu\text{g/g}$, dry weight (dw) and less than 0.3 $\mu\text{g/g}$, dw in Echo Bay and Las

Vegas Bay razorback suckers, respectively. Selenium is strongly bioaccumulated by aquatic organisms (Lemly 1996). Skorupa *et al.* (1996) reported normal whole body selenium in fish as <1 - 4 µg/g, dw, and estimated a threshold range for reproductive impairment in sensitive fish species as between 4 and 6 µg/g, dw whole body concentration. Toxic endpoints commonly include histopathology, physiological effects, reduced reproduction, and reduced survival. Selenium is recognized as a powerful teratogen, and reproduction is generally considered the most sensitive and significant toxic endpoint. Selenium has been identified as a concern throughout the Colorado River basin (Hamilton 1999). Anthropogenic sources, particularly agricultural drainage, were identified as the primary cause of elevated concentrations in the lower basin.

Assessment of chemical concentrations in single razorback sucker samples collected from each of Las Vegas Bay and Echo Bay (reference) indicate that Lake Mead razorback suckers are exposed to and accumulate certain contaminants. Distinct differences were found in the samples collected from each area. Nine of 22 analyzed OC compounds were detected in the razorback sucker collected from Las Vegas Bay (Figure 8). The total OC concentration was 0.737 µg/g, ww. No OC compounds were detected in the razorback sucker from Echo Bay. DDT residues (the sum of DDT and the metabolites DDD and DDE) accounted for more than half of the total detected OC concentration (0.408 µg/g, ww) in the Las Vegas Bay razorback sucker. Total PCBs accounted for about a third of the total detected OC concentration (0.267 µg/g, ww). Bevans *et al.* (1996) found median total DDT concentrations of 0.072 and 0.120 µg/g, ww in cross sectional carp tissue samples collected from Las Vegas Wash and Bay, respectively. USGS analyzed only *p,p'*-DDT, *p,p'*-DDD, and *p,p'*-DDE. The total *p,p'*-DDT concentration in the Las Vegas razorback sucker sample was more than twice the median concentration for Las Vegas Bay carp (0.29 µg/g, ww). Bevans *et al.* (1996) found a median concentration of total PCBs of 0.13 and 0.14 µg/g, ww in carp tissue from Las Vegas Wash and Bay, respectively. The total PCB concentration in the Las Vegas Bay razorback sucker (0.267 µg/g, ww) was approximately twice these median concentrations.

DDT and PCBs have been associated with a variety of effects to fish. Tissue concentrations associated with adverse effects to fish have not been well established. According to the summary of Niimi (1996), tissue concentrations of PCBs in razorback suckers and carp

from Las Vegas Wash and Bay were below levels associated with effects to survival, growth, or reproduction in fish (> 50 mg/kg) and the range associated with biochemical changes and cellular-level effects (> 5 mg/kg).

The limited number of razorback suckers from Las Vegas Bay and Echo Bay examined during sampling did not exhibit overt external indications of adverse health effects from environmental contaminants. Size, weight, and Fulton's condition factor of male razorback suckers were not significantly different between sites (Table 3). No tumors or external lesions were noted on any of the razorback suckers examined. Histological analysis of common carp collected from Las Vegas Wash, Las Vegas Bay, and Calville Bay in Lake Mead indicated necrotic changes in hepatopancreas (e.g., liver) and kidney tissues (Bevans *et al.* 1996). The pattern and severity of necrosis were consistent with subchronic exposure to contaminants or combinations of contaminants. Because of their endangered status and limited numbers in Lake Mead, razorback suckers were not sacrificed for assessment of histopathology.

Dioxins, PAHs, certain metals, and other contaminants identified at elevated concentrations in Las Vegas Wash have the ability of cause DNA damage (genotoxic). Exposure to such compounds may result in carcinogenic or mutagenic effects in fish and wildlife. An initial step in chemical carcinogenesis is the modification of DNA by genotoxic compounds resulting in the formation of adducts (Beland and Kadlubar 1985, Poirier *et al.* 1991). The number of DNA adducts formed and the relative persistence of the adducts appear to be related to the carcinogenicity of a chemical (Reichert *et al.* 2001). As such, measurement of DNA adduct levels in fish may be used to assess relative risk from exposure to environmental carcinogens and mutagens in fish. The frequency of DNA adducts in razorback sucker red blood cells was not significantly different ($p = 0.59$) between Las Vegas Bay and Overton Bay (Table 3).

A variety of known EDCs have been identified in water, sediment, fish blood plasma, and tissues collected from Las Vegas Wash and Bay. Consistent with this, research has identified indications of endocrine disruption in fish from Lake Mead. Bevans *et al.* (1996) found significantly lower levels of the sex steroids 11-ketotestosterone and 17 β -estradiol in male carp from Las Vegas Bay compared to those from a reference site (Callville Bay in Lake Mead). Low concentrations of sex steroids can cause premature sexual maturity, reduced gonad size, and

reduced secondary sex characteristics (Munkittrick *et al.* 1992). Male carp from Las Vegas Wash and Bay also had significantly higher concentrations of the protein vitellogenin than did males from the reference site. Vitellogenin, an estrogen-controlled egg protein, is usually at or below the lower analytical detection limit and has no known function in males. Female carp from Las Vegas Wash had significantly higher levels of 11-ketotestosterone compared to controls. The ratio of 17 β -estradiol to 11-ketotestosterone in female carp from Las Vegas Bay was comparable to male carp. The ratio of 17 β -estradiol to 11-ketotestosterone was significantly higher in male carp from Las Vegas Bay compared to the reference site.

Goodbred *et al.* (1999) examined sex steroid hormone concentrations and ratios, vitellogenin concentrations, and physiological indications of endocrine disruption in carp from eight sites along the lower Colorado River from Pierce Ferry in upper Lake Mead to Lake Mohave. Concentrations of 11-ketotestosterone again were significantly lower in Las Vegas Bay male carp and the ratio of 17 β -estradiol to 11-ketotestosterone was more than 80 percent higher in Las Vegas Bay male carp than controls. Forty percent of the male carp from Las Vegas Bay had a ratio that exceeded 1 (male ratios are typically < 1, females > 1). Vitellogenin was detected in male carp from five of the eight sites, but was not found in male carp from Las Vegas Bay. Ethynylestradiol, an estrogen used in commercial birth control pills, was detected in carp from Las Vegas Bay and Calville Bay. Concentrations were significantly higher in male carp from Las Vegas Bay. Sperm viability was significantly lower in Las Vegas Bay carp. The mean gonadosomatic index (ratio of gonad weight to total body weight) of female carp from Las Vegas Bay was 30 percent lower than predicted based on the stage of egg maturation. Reduced gonadosomatic indices have been associated with contaminant exposure in other studies (Dey and Bhattacharya 1998; Shukla and Pandey 1985).

Assessment of Lake Mead carp and largemouth bass reproductive physiology and functionality by the USGS and Service is ongoing. Preliminary data do not indicate a substantial difference in hormone concentrations or physiological measurements in female carp collected from Las Vegas Bay and a Lake Mead reference site (Overton Beach). Observed differences appear to be attributable to seasonal asynchrony between sites. Several differences were found in male carp, including reduced 11-ketotestosterone concentrations in blood plasma, skewed

ratios of 17 β -estradiol to 11-ketotestosterone, decreased gonadosomatic index values, and decreased sperm motility and viability. Reduced 11-ketotestosterone concentrations in blood plasma and skewed ratios of 17 β -estradiol to 11-ketotestosterone were also found in male largemouth bass from Las Vegas Bay as compared to the reference site. Female largemouth bass from Las Vegas Bay had higher 11-ketotestosterone concentrations in blood plasma than controls. Results of these investigations are expected to be reported in 2003.

Razorback suckers in Lake Mead also exhibit indications of endocrine disruption. Concentrations of 17 β -estradiol were significantly higher ($p = 0.015$) in male razorback suckers from Las Vegas Bay than those from Echo Bay (Figure 10). Concentrations of 11-ketotestosterone were lower, with the difference approaching significance ($p = 0.068$). The ratio of 17 β -estradiol to 11-ketotestosterone was also significantly higher in Las Vegas Bay males than in Echo Bay males. The mean ratios were 1.01 and 0.23 in males from Las Vegas Bay ($n = 11$) and Echo Bay ($n = 10$), respectively. The mean ratio and ratios in 5 of 11 male razorback suckers exceeded 1 (Figure 9). Ethynylestradiol was detected in blood plasma of razorback suckers collected from Las Vegas Bay (Tim Gross, USGS Florida Caribbean Science Center, pers. comm. 2001).

Due to the limited sample sizes, statistical comparisons of hormone concentrations and ratios were not attempted for female razorback suckers. However, the 17 β -estradiol concentration in a single female from Las Vegas Bay was substantially lower than concentrations observed in females from Echo Bay (Figure 9). Consistent with this, the ratio of 17 β -estradiol to 11-ketotestosterone was substantially lower in the Las Vegas Bay female.

Effects of EDCs to razorback sucker physiology and functionality are uncertain at this time. Due to their endangered status and limited numbers in Lake Mead, razorback suckers were not sacrificed for assessment of gonad histology, gonadosomatic index, or organosomatic assays. Attempts to assess sperm motility and viability of male razorback suckers were not successful in 2000, due to the absence of an appropriate sperm extender (necessary for microscopic observation of sperm). Preliminary data from 2001, indicate that sperm motility and viability are lower in male razorback suckers from Las Vegas Bay. Results of this component of the investigation will be reported in 2003.

Hormone concentrations in fish can vary with season and reproductive status. All razorback suckers evaluated in our investigation were collected near known spawning areas during the time period that razorback suckers spawn. Additionally, all but one male at each site

expressed milt when palpated. Therefore, differences in reproductive status are not believed to account for differences in hormone concentrations and ratios observed between razorback sucker populations in Lake Mead.

CONCLUSIONS

Razorback suckers in Lake Mead are exposed to and accumulate contaminants. Several contaminants have the potential to compromise reproduction and survival of the Lake Mead populations. The highest selenium concentration was in a single whole-body razorback sucker sample from Echo Bay. The concentration in this sample was sufficient to reduce reproductive success. Other trace elements do not appear to be at levels of concern to razorback suckers. Several OC compounds were detected in the razorback sucker sample from Las Vegas Bay but none were detected in the Echo Bay sample. PCBs and metabolites of DDT accounted for the majority of the combined OC concentration in the Las Vegas Bay razorback sucker.

The Lake Mead razorback suckers examined during trammel netting did not exhibit overt external indications of contaminant exposure and effects. Condition factor values of Las Vegas Bay and Echo Bay razorback suckers were similar. Additionally, no difference in indications of exposure to genotoxic compounds was found between the two Lake Mead razorback sucker populations.

As compared to Echo Bay, Las Vegas Bay razorback suckers exhibit evidence of endocrine disruption. Male razorback suckers in Las Vegas Bay had higher concentrations of 17β -estradiol and skewed ratios of 17β -estradiol to 11-ketotestosterone. Based on a limited sample size, female razorback suckers in Las Vegas Bay appear to have depressed 17β -estradiol concentrations and skewed ratios of 17β -estradiol to 11-ketotestosterone. Efforts to initiate a cooperative study to determine potential reproductive effects in fish at the population level are ongoing.

ASSESSMENT OF DOWNSTREAM CONTAMINANT EXPOSURE AND EFFECTS

INTRODUCTION

Water quality monitoring has demonstrated that a variety of potentially toxic constituents are introduced to Lake Mead from Las Vegas Wash. The occurrence of elevated perchlorate concentrations in the Colorado River downstream of Hoover Dam demonstrates that contaminants from Las Vegas Wash affect water quality in the lower Colorado River. Analysis of biological samples provides evidence that fish and wildlife in Las Vegas Bay are exposed to and accumulate some of these constituents. Biochemical and physiological differences between fish collected from Las Vegas Bay and sites upstream of the Bay raise concern for potential effects to fish, wildlife, and their habitat downstream of Las Vegas Wash. The largest razorback sucker population in the lower Colorado River basin occurs in Lake Mohave and the Colorado River downstream of Hoover Dam. Additionally, these areas also support endangered bonytail chub and southwestern willow flycatcher. This portion of the investigation was designed to assess the occurrence of contaminants and biological effects in the Colorado River downstream of Hoover Dam and in Lake Mohave.

METHODS

Semi-permeable membrane devices were employed to assess the occurrence of selected OC compounds in the water column in Las Vegas Bay and areas downstream of Lake Mead. SPMDs, which consist of low-density polyethylene tubes filled with triolein or other lipid materials, are effective at sequestering lipid soluble compounds dissolved in water (Huckins *et al.* 1990). SPMDs were attached to anchor chains of channel buoys in Las Vegas Bay (equivalent to site 3 in Bevans *et al.*, 1996), near the USGS gaging station on the Colorado River downstream of Hoover Dam, near the Willow Beach boat launch area, and in Cottonwood Cove on Lake Mohave. At each location, SPMDs were affixed at a depth of approximately 1 m for a five week period from early June to mid July, 1998. The time period and length of time SPMDs were set coincided with SPMD sampling conducted in 1995 (Bevans *et al.* 1996). SPMDs and deployment devices, along with dialysis and cleaning services upon retrieval were provided by

Environmental Sampling Technologies (St. Joseph, Missouri). SPMD extracts were submitted to the Mississippi State Chemical Laboratory for analysis of selected OC and PAH compounds.

Lake Mead carp were collected in 1995 (Bevans *et al.* 1996). Organochlorine pesticide and PCB residue concentrations in that study were determined in 5-cm vertical cross sections of Lake Mead carp taken mid-body by the USEPA Region 9 Laboratory in Richmond, CA, using dual column gas chromatography with electron capture detectors. Additional analyses were performed by atomic emission detector with identification and confirmations by ion trap detector. Carp from Lake Mohave and Lake Mead were collected in 1998. Whole carp carcasses were stored and shipped frozen to PACF, Laurel, MD for determination of OC residues. Residue concentrations were quantified with gas-liquid chromatography (GLC) equipped with a ^{63}Ni electron capture detector. The GLC column used was a 30m MEGABORE coated with a 1.0 micron film of 7 percent cyanopropyl 7 percent phenyl polysiloxane. Residues in 10 percent of the samples were confirmed by gas chromatography/mass spectrometry (GC/MS). The nominal lower limit of detection was 0.01 ppm for pesticides and 0.05 ppm for PCBs based on a 10 g aliquot, ww. Only analytes common to both groups (alpha BHC, alpha chlordane, beta BHC, dieldrin, endrin, gamma BHC, gamma chlordane, heptachlor epoxide, *p,p'*-DDD, *p,p'*-DDE, *p,p'*-DDT, PCBs resembling Aroclor 1254, and toxaphene) were included in subsequent figures.

In 1998, a total of 46 adult carp were collected from Lake Mead near Hoover Dam (Hemenway Beach area, $n = 7$), the Colorado River between Lake Mead and Lake Mohave (Willow Beach area, $n = 19$), and Lake Mohave (Cottonwood Cove area, $n = 20$) to assess contaminant accumulation and effects in areas downstream of Las Vegas Wash. Carp were collected by boat-operated electrofishing techniques, temporarily held in an on-board tank, then transferred to an in-situ livebox for 1 to 3 hours prior to necropsy. Each fish was lightly anesthetized with MS222, struck on the head, rapidly examined for external abnormalities (gill, eye, skin), weighed, measured for total and fork length, and bled from the caudal peduncle with a heparinized syringe. The blood sample was used for a blood smear (methanol fixed, Leishman-Giemsa stain) and aliquots then centrifuged (10,000 RPM, 10 min.) for measurement of hematocrit, leukocrit, and collection of plasma. Blood smears were examined at 1000X magnification and the first 100 leukocytes counted as lymphocytes, thrombocytes (elongated forms only), neutrophils, eosinophils, or monocytes (Roberts 1989, Witten *et al.* 1998). Clot activation during the collection of the blood sample can result in “spent” thrombocytes which

resemble small lymphocytes (Roberts 1989). As these atypical thrombocytes are easily confused with small lymphocytes, a combined lymphocyte + thrombocyte count was compared with the granulocyte count (neutrophil, eosinophil, and rarely monocytes).

Plasma was frozen on dry ice and later assayed for total protein (spectrophotometer assay using Pierce Chemical Company bicinchoninic acid reagent and standard) and electrophoretic profile. Plasma protein electrophoresis was performed with a 7 FL sample run on a CIBA agarose gel (1M barbital buffer, 90V for 45 min.). The stained gels were scanned and the percent area of each fraction determined with Seprascan™ software. Analysis of variance was performed on percent area values for each fraction (or combined fractions).

The peritoneal cavity was opened and internal organs were examined for gross abnormalities. A sterile swab was inserted into the kidney and used to inoculate a trypticase soy agar plate. After the gonad was removed and weighed, kidney samples were collected for *R. salmoninarum* antigen enzyme-linked immunosorbent assay and viral assay on both fathead minnow (FHM) and CHSE214 cell lines (five fish pooled samples cultured for 15 days at 15 °C). Samples of kidney, liver, and gonad for histological examination were collected and fixed in Davidson's fixative for 24 hours then transferred to 70 percent ethanol. These tissues were processed for 5 mm paraffin sections and stained with hematoxylin and eosin. A subset of sections from each sample group were stained with Perl's Iron stain (hemosiderin), Acid fast stain (ceriod/lipofuscin), and Sudan Black B (ceriod/lipofuscin).

Blood plasma samples were also submitted to the Florida Caribbean Science Center for analysis of 17β-estradiol, 11-ketotestosterone, ethynyl-estradiol, and vitellogenin. Analyses followed methods described in Goodbred *et al.* (1997).

Three composited crayfish samples were collected from backwater ponds used as rearing ponds for larval razorback suckers. Each sample consisted of a composite of a minimum of five crayfish. Samples were placed into clean glass jars and submitted for OC and trace element residue analysis to PACF in Laurel, Maryland.

RESULTS AND DISCUSSION

Semipermeable Membrane Device Monitoring

Bevans *et al.* (1996) detected a variety of organic compounds in extracts from SPMDs set in Las Vegas Wash (1 site), Las Vegas Bay (3 sites), and Calville Bay (1 site) in Lake Mead.

Semipermeable membrane devices sequester lipid-soluble contaminants from the water column, thus allowing time-integrated sampling of a variety of organic contaminants. The number of detected OC compounds and combined concentrations were significantly higher in Las Vegas Wash and Bay compared to sites below Hoover Dam. Combined concentrations of tetrachlorodibenzo-*p*-dioxins and tetrachlorodibenzofurans were also higher in Las Vegas Wash followed by Las Vegas Bay. Polycyclic aromatic hydrocarbons were elevated at all sites. We identified 12 OC compounds in extracts of an SPMD placed in Las Vegas Wash (Figure 11). Compounds detected in 1996 and 1998 included hexachlorobenzene, cis-chlordane, trans-chlordane, trans-nonachlor, dieldrin, *p,p'*-DDD, *p,p'*-DDE, and PCBs. The combined concentration of these compounds in 1998 was lower than that found by Bevans *et al.* (1996) (Figure 11). The DDT metabolites DDD and DDE accounted for about one-half of the combined concentration of all OCs (0.222 and 0.433 $\mu\text{g/g}$, respectively), with PCBs accounting for about one-quarter (0.100 $\mu\text{g/g}$). Although not reflected in Figure 11, *o,p'*-DDD and *o,p'*-DDE were also found at a combined concentration of 0.08 $\mu\text{g/g}$ in 1998. Organochlorine compounds were not detected in extracts of SPMDs placed in the Colorado River immediately downstream of Hoover Dam, the Colorado River at Willow Beach, or Cottonwood Cove of Lake Mohave.

Trace Elements in Carp

Unless otherwise noted, trace element concentrations in the following discussion are given on a dry weight basis.

Arsenic, beryllium, and molybdenum were at or less than analytical detection limits (0.33, 0.07, and 1.3 $\mu\text{g/g}$, respectively) in all whole-body carp samples. We are not aware of information to evaluate concerns with boron in whole fish. Boron concentrations in all samples, ranging from less than 1.3 to 42 $\mu\text{g/g}$, were well below a 300 $\mu\text{g/g}$ concentration associated with reduced reproductive success in birds (Smith and Anders 1989). Similarly, we found no information on the implications of cadmium in whole fish but concentrations in all carp samples (<0.06 to 0.8 $\mu\text{g/g}$) were less than a 2.0 $\mu\text{g/g}$ concentration associated with biochemical changes in birds (Furness 1996) and well below a 20 $\mu\text{g/g}$ concentration associated with histopathology in birds (Cain *et al.* 1983).

With the exception of a single fish from Willow Beach which had a chromium concentration of 3.9 $\mu\text{g/g}$, chromium concentrations in carp were well below (# 1.2 $\mu\text{g/g}$) a 4.0 $\mu\text{g/g}$ concentration identified as a concern level by Eisler (1986). Chromium concentrations

were similar in whole carp collected from Hemenway Beach, Willow Beach, and Cottonwood Cove.

Ohlendorf (1998) indicated that a copper concentration of 9.8 $\mu\text{g/g}$ in whole rainbow trout was associated with a slight increase in mortality and a concentration of 13.3 $\mu\text{g/g}$ was associated with significant mortality. In our study, copper concentrations in whole carp samples from all sites ranged from 1.2 to 15.2 $\mu\text{g/g}$. Concentrations in 5 samples (~ 10 percent) exceeded the 9.8 $\mu\text{g/g}$ effect level. Only a single sample exceeded 13.3 $\mu\text{g/g}$. Concentrations were similar among sites.

Lead concentrations were less than the 0.33 $\mu\text{g/g}$ analytical detection limit in 30 of 46 (65 percent) whole-body carp samples in our study. The highest concentration was 2.3 $\mu\text{g/g}$. We were unable to locate information on effect levels of lead in whole-body fish. Concentrations in all carp samples were well below a 25 $\mu\text{g/g}$ dietary concentration associated with biochemical effects and a 125 $\mu\text{g/g}$ dietary level associated with reduced growth and abnormal development in birds (Finley *et al.* 1976, Hoffman *et al.* 1985).

Mercury was detected in 4 of 7 samples (~57 percent) from Hemenway Beach, 1 of 19 samples (5 percent) from Willow Beach, and 10 of 20 samples (50 percent) from Cottonwood Cove. The highest concentration, 0.48 $\mu\text{g/g}$, was found in a Hemenway Beach sample. Snarski and Olson (1982) reported reduced reproduction of fathead minnows (*Pimephales promelas*) when whole body concentrations exceeded 0.6 $\mu\text{g/g}$, ww. Kania and O'Hara (1974) reported diminished predator-avoidance behavior in mosquitofish (*Gambusia affinis*) when whole-body mercury concentration exceeded 0.7 $\mu\text{g/g}$, ww. Wet weight concentrations in all of our whole-body carp samples (# 0.07 $\mu\text{g/g}$, ww) were below these effect levels.

Selenium concentrations in whole-body carp in our study ranged from 2.4 to 12.6 $\mu\text{g/g}$, with a mean of 4.9 $\mu\text{g/g}$ for all sites combined (Figure 11). Lemly (1996) identified a concentration of 4 $\mu\text{g/g}$ in whole-body fish as a threshold of toxic effects to fish. Toxic endpoints commonly include histopathology, physiological effects, reduced reproduction, and reduced survival. Selenium is recognized as a powerful teratogen, and reproduction is generally considered the most sensitive and significant toxic endpoint. Selenium concentrations exceeded this threshold in approximately 60 percent of the whole fish samples in our study and, as such, likely represents a threat to fish reproduction in the lower Colorado River. Additionally, 17 percent of the whole fish samples exceeded a critical avian dietary threshold of 5.0 $\mu\text{g/g}$ identified by Skorupa and Ohlendorf (1991). Therefore, selenium also represents a threat to

piscivorous birds in the study area. Selenium has been identified as a concern throughout the Colorado River basin (Hamilton 1999). Anthropogenic sources, particularly agricultural drainage, were identified as the primary cause of elevated concentrations in the lower basin.

Organochlorine Compounds in Carp

We detected a total of 2, 10, and 5 OC compounds at Hemenway Beach, Willow Beach, and Cottonwood Cove, respectively. Bevans *et al.* (1996) found nine OC compounds in carp from Callville Bay and 17 compounds in carp from Las Vegas Bay. Of the 13 OC compounds common to both data sets, the number of detected OC compounds detected in carp (Figure 12) and the combined concentrations were statistically higher ($p < 0.00$, $p = 0.02$) in Las Vegas Bay carp (Figure 13). With the exception of Cottonwood Cove where endrin was elevated in about one-third of the samples, total DDT (combined concentrations of *p,p'*-DDD and *p,p'*-DDE) and PCBs (resembling Aroclor 1254) accounted for the bulk of the total OC concentrations. Total DDT concentrations were highest in carp from Las Vegas Bay followed by carp from downstream sites (Figure 14). Concentrations in Las Vegas Bay carp were statistically higher ($p \neq 0.02$) than Callville Bay, Hemenway Beach, and Cottonwood Cove, but not Willow Beach ($p = 0.29$). PCBs resembling Aroclor 1254 were detected in all but one carp from Las Vegas Bay but in fewer than one-half of the fish at other sites. PCB concentrations were lower than known effect concentrations for fish (Niimi 1996).

Carp Health

Generally, carp evaluated at Colorado River sites downstream of Las Vegas Wash and Bay (Hemenway Beach, Willow Beach, and Cottonwood Cove) were in good condition (Foott and Harmon 1999). Total length of sampled carp (412 - 442 mm) were similar at all sites (Figure 15). Weights were greater in females than males (Figure 15) as were associated condition factors (Figure 16). These differences were likely attributable to the gravid condition of the females (Foott and Harmon 1999). Fulton's condition factor values for female carp from Cottonwood Cove were significantly lower ($p = 0.03$) than those from Hemenway Beach. Although no difference was found for gonadosomatic index values for females among sites, there was uncertainty in these data due to the loss of eggs during capture and necropsy. Gonadosomatic index values were significantly lower in male carp from Willow Beach than at

other sites (Figure 17). The difference may have been due to cooler water temperatures and associated delayed maturation at the Willow Beach site.

Several differences were found in blood cell characteristics in carp from Hemenway Beach as compared with Willow Beach and Cottonwood Cove (Foott and Harmon, 1999). Carp from Hemenway Beach had both lower percentages of white blood cell volumes (e.g., lower leukocrit values) and elevated eosinophil counts. Lymphocyte to monocyte/granulocyte ratios for carp from Willow Beach and Cottonwood Cove were within the normal range for carp but were significantly lower in fish from Hemenway Beach. Exposure to a variety of contaminants, such as ammonia, metals, and PCBs, have been associated with depression of lymphocytes and corresponding elevation of granulocytes (Modra *et al.* 1998). The stress hormone cortisol may also induce decreased lymphocyte levels in carp (Weyts *et al.* 1998). No statistically significant differences in plasma protein composition were found among sites or between sexes.

No obvious trends in bacterial or viral infections were noted at the Colorado River sites downstream of Las Vegas Wash.

Several histological abnormalities in kidney and hepatopancreas (e.g., liver) tissue were common to most carp examined at all sites. Of note were hemosiderin deposits in the majority of fish examined from all sites. Apoptotic macrophage aggregates were often observed in association with hemosiderin deposits. Hemosiderin, a golden-brown pigment composed of aggregates of ferritin micelles, forms when there is an excess of iron in tissue (Cotran *et al.* 1989). In fish, hemosiderosis (an excess of hemosiderin in tissue) is usually associated with hemolytic anemia or excessive intake of iron in diet or directly from water (Thiyagarajah *et al.* 1998). Lipopigments, including lipofuscin and ceroid, form as a result of peroxidation of cellular lipids and, therefore, may provide an indication of oxidative stress in fish in this reach of the river. Endogenous pigments were also observed in kidney and hepatopancreas tissue of fish collected from Overton Bay in Lake Mead (Foott *et al.* 2000). Other histological abnormalities observed in carp from all sites include hyperplastic musculature surrounding the bile ducts, thyroid follicles of various sizes within the kidney interstitium, and calcium oxalate crystals in the distal kidney tubules. The prevalence of these conditions at all sites would suggest that the possible cause or causes are related to general conditions in this reach of the Colorado River and is likely not attributable to contaminants associated with Las Vegas Wash.

Endocrine Disruption Biomarkers

Assessment of the principle androgen or estrogen in carp did not reveal significant abnormalities in concentrations and ratios in female carp collected downstream of Las Vegas Wash. Concentrations of 17 β -estradiol and vitellogenin in female carp were significantly lower ($p=0.05$) at Willow Beach as compared to Hemenway Beach (Figures 18 and 21, respectively). The difference between Hemenway Beach and Cottonwood Cove was not significant. Cooler water temperatures (13°C at Willow Beach versus 20°C at Hemenway Beach) and temperature-mediated differences in reproductive status likely accounted for differences. Between site differences in 11-ketotestosterone (Figure 19) and ratios of 17 β -estradiol to 11-ketotestosterone (Figure 20) were not significant ($p>0.06$).

Like females, significant abnormalities in the principle androgen or estrogen concentrations or ratios were not apparent in male carp collected downstream of Las Vegas Wash. Although based on a limited sample size of male carp from Hemenway Beach ($n = 2$), the gonadosomatic index values for male carp from Willow Beach were significantly lower than those for male carp from Hemenway Beach and Cottonwood Cove (Figure 17). Temperature-mediated differences in reproductive status are again suspected. The lowest concentrations of 11-ketotestosterone were in male carp from Hemenway Beach (Figure 19). Concentrations were significantly lower ($p = 0.25$) than concentrations in males from Cottonwood Cove. Concentrations of 17 β -estradiol (Figure 18) and ratios of 17 β -estradiol to 11-ketotestosterone (Figure 20) were not significant among sites ($p\leq 0.14$). The ratio of 17 β -estradiol to 11-ketotestosterone was less than one (male values below one are considered normal) for all male carp at all sites. Vitellogenin was detected in three males from Willow Beach and three males from Cottonwood Cove (30 percent of the samples, each). Vitellogenin was not detected in either of the male carp from Hemenway Beach (Figure 21).

Contaminants at Razorback Sucker Recovery Sites

Razorback sucker recovery strategies include the rearing of razorback sucker larvae in predator-free backwater areas adjacent to Lake Mohave. Juvenile razorback suckers are later released back to the lake when they have attained sufficient size to avoid predation. We assessed contaminant concentrations in environmental samples collected from backwater areas to determine if contaminants have the potential to reduce larval survival or otherwise compromise the suitability of backwater areas for use as nursery habitats. Several of the contaminants associated with Las Vegas Wash and the mainstem of the lower Colorado River have a

propensity to bioaccumulate in organisms and concentrate in food chains. Therefore, crayfish were selected as a sample medium because they reflect exposure to contaminants in water and diet and have the ability to accumulate contaminants.

The highest concentrations of most trace elements of concern were found in crayfish from Davis Cove (Table 4). Beryllium, mercury, molybdenum, and selenium were less than the analytical detection limits at all sites. Analytical detection limits were lower than known biological effect levels for these elements. Concentrations were less than effect levels for whole-body fish and avian or fish dietary effect levels for arsenic (Gilderhus 1966), boron (Smith and Anders 1989), cadmium (Furness 1996), chromium (Eisler 1986), and lead (Hoffman *et al.* 1985).

None of the 22 target organochlorine compounds were detected in crayfish from the sampled coves (Table 5) suggesting that OC compounds are not of concern in these razorback sucker recovery sites.

CONCLUSIONS

Elevated perchlorate concentrations in the lower Colorado River indicate that contaminants introduced from Las Vegas Wash affect water downstream of Hoover Dam. This portion of the investigation was initiated to evaluate the potential for environmental contaminants from Las Vegas Wash to adversely affect reproduction and survival of aquatic organisms in Lake Mohave and the Colorado River downstream of Hoover Dam.

Sampling with SPMDs suggest that lipid-soluble contaminants did not pass Hoover Dam in significant concentrations during the period sampled. The period in which SPMDs were set coincided with the maximum extent of intrusion of Las Vegas Wash water into Lake Mead (LaBounty and Horn 1997). The detection of organochlorine compounds in fish downstream of Hoover Dam indicate that fish are exposed to and accumulate organochlorines in the Colorado River downstream of Hoover Dam and in Lake Mohave. However, the detected organochlorine compounds at the concentrations observed do not appear to represent a significant threat to fish reproduction or survival. The source of these contaminants downstream of Hoover Dam remains uncertain.

Fish collected downstream of Hoover Dam contained selenium at concentrations that have the potential to compromise reproduction and survival of aquatic organisms and birds.

However, elevated selenium concentrations were detected upstream of the inflow of Las Vegas Wash into Lake Mead indicating that selenium was a concern throughout Lake Mead and Lake Mohave. Previous investigations have identified selenium input to the upper Colorado River as a threat to fish and wildlife throughout the lower Colorado River basin. Other trace elements did not appear to represent significant threats to fish and wildlife downstream of Hoover Dam.

Fish collected in Lake Mead exhibited indications of tissue damage. Observed histopathology was consistent with chronic exposure to sublethal contaminant concentrations. Again, similar tissue damage was observed in fish collected upstream of the inflow of Las Vegas Wash suggesting that histopathology was a regional concern and not specifically attributable to the input of contaminants from Las Vegas Wash. No significant concerns with fish pathogens were identified in the study area. Overt indications of endocrine disruption were not identified downstream of Hoover Dam.

Significant contaminant concerns were not identified in backwater areas adjacent to Lake Mohave that are used as rearing areas for razorback sucker larvae.

GENERAL CONCLUSIONS

Degraded water quality conditions and environmental contaminants associated with Las Vegas Wash and Bay have the potential to adversely affect fish, wildlife, and habitat quality in Lake Mead. This study was initiated to assess the extent and magnitude of contaminant threats to trust resources occurring from Las Vegas Wash to Lake Mead and the Colorado River downstream of Hoover Dam. Concerns to trust resources have been identified due to elevated nutrient, trace element, pathogen, and pesticide concentrations in water from Las Vegas Wash and Las Vegas Bay. Low dissolved oxygen concentrations are a concern to aquatic species in portions of Las Vegas Bay. Potentially toxic compounds that have been identified in Las Vegas Wash and Las Vegas Bay water and/or sediment, include metals and other trace elements, perchlorate, pesticides, organochlorine compounds, dioxins, furans, polycyclic aromatic hydrocarbons, phthalates, phenolic compounds, and suspected EDCs (e.g., ethynylestradiol). Many of these contaminants were identified at levels known to be toxic to fish and wildlife. With the exception of selenium, which occurred at concentrations of concern throughout the study area, degraded habitat conditions and contaminants at concentrations of concern appeared to be localized in the Las Vegas Wash and Bay areas. Elevated organochlorine compound

concentrations were found in fish and avian eggs in the Las Vegas Wash and Bay areas. Selenium was elevated in fish tissue throughout the study area. Observed biological effects included abnormal hormone concentrations and ratios and tissue damage in fish. Fish endocrine system effects appear to be localized to the Las Vegas Wash and Bay area while histopathology occurred throughout the study area. This study adds to the growing literature that indicates that certain contaminants are present in Las Vegas Wash and Bay biota at levels known to adversely affect fish and wildlife.

ONGOING EFFORTS AND RECOMMENDATIONS

Since 1998, several steps have been taken to address water quality issues in Las Vegas Wash and Bay. Perhaps most significant is the formation of two technical advisory groups, the Lake Mead Water Quality Forum and the Las Vegas Wash Coordination Committee. The Service serves as an active participant on these committees, which have coordinated research and assessment efforts of multiple agencies, utilities, and private entities. Clark County and the cities of Las Vegas, North Las Vegas, and Henderson have initiated efforts to restore wetlands in the lower sections of Las Vegas Wash. Southern Nevada Water Authority, comprised of seven member agencies responsible for water delivery and wastewater treatment in southern Nevada, has also increased monitoring, assessment, and restoration efforts along Las Vegas Wash. However, successful habitat restoration requires careful design and implementation to prevent the increase of contaminant exposure and effects in created habitats and to ensure contaminant threats are not transferred to other areas.

Effective water quality management and habitat restoration are dependent on comprehensive monitoring programs that will identify adverse effects to fish and wildlife. As such, it is vital that monitoring programs assess the occurrence and severity of all contaminants and conditions of concern, including the presence of non-conventional pollutants (e.g., pharmaceuticals and personal care products) and toxic constituents associated with sewage effluent and urban runoff. Similarly, monitoring efforts should include assessment of habitat conditions and the diversity, abundance, health, and condition of fish, wildlife, plants, and invertebrates. Monitoring should include several areas where construction or restoration activities are planned for comparison of baseline to post-construction conditions. Southern Nevada Water Authority has already initiated surveys for reptiles, macroinvertebrates, and birds

to assess species diversity along Las Vegas Wash. A cooperative effort between the Service and SNWA to monitor trace element and organochlorine residues in bird eggs and fish along Las Vegas Wash is planned for the spring of 2003.

Integral to the success of the combined efforts to improve water quality and improve habitat is the establishment of appropriate water quality standards and criteria. In the absence of appropriate standards, Las Vegas Wash and Bay may not be identified as impaired waters under Section 303(d) of the Clean Water Act (CWA), despite evidence that certain conditions are not conducive to healthy populations of fish and wildlife. Adequate standards first require the designation of appropriate beneficial uses and reaches which are reflective of existing conditions and existing species assemblages. Examples where beneficial uses are not adequate include a segment of lower Las Vegas Wash where “propagation of fish” is excluded as a beneficial use in an area which has been designated as critical habitat and where an endangered fish occurs, as well as, where water contact recreation is prohibited in an area within the Lake Mead National Recreation Area.

Endocrine disrupting chemicals in wastewater flows and associated affects to wildlife are yet another water quality issue in Las Vegas Wash and Las Vegas Bay. The Service has been involved in ongoing negotiations with representatives from NDEP and wastewater treatment facilities for Clark County, City of Las Vegas, and City of Henderson to address this issue. The wastewater dischargers are collectively known as the Clean Water Coalition (CWC). Despite the fact that the State and CWC have developed a better understanding of Service concerns and the issue of endocrine disruption in general, no real progress has been made on how best to address EDCs and to what extent the CWC should be held accountable for the ecological impacts of wastewater flows. The Service maintains that there is overwhelming evidence that EDC’s are present in wastewater that comprises over 90 percent of Las Vegas Wash base flows, that scientific studies have demonstrated a relationship between certain constituents in wastewater and adverse effects to trust resources in the area, and that the best approach to assess population level effects is to conduct a cooperative reproductive endpoint study on fish exposed to wastewater. Although the CWC has agreed to allow access to their property for the proposed study, as of the writing of this report, the CWC seems unwilling or unable to assist in the financial support necessary for such an endeavor.

Over the last decade, the City of Las Vegas, Clark County, and City of Henderson have expended considerable time and money to improve the capacity and capabilities of their

wastewater treatment plants, often incorporating state-of-the-art technology. These efforts are to be commended and have undoubtedly improved water quality conditions in Las Vegas Wash. Additionally, the CWC is in the process of preparing an environmental impact statement for alternative sites to discharge future increases in wastewater flows from Las Vegas Valley, and possibly for diverting a portion of existing flows from Las Vegas Wash. Alternatives that are being considered include diverting flows to a wash along the north or south shoreline of Boulder Basin, diverting flows immediately above or below Hoover Dam, and enhancements to wastewater treatment to improve water quality within the existing discharge channel (Las Vegas Wash). At present, discharging above or below Hoover Dam appears to be favored by the CWC, raising concerns about potential impacts to razorback suckers occurring downstream of Hoover Dam including Lake Mohave. The Willow Beach National Fish Hatchery, located approximately 18 miles downstream of Hoover Dam in Arizona, collects adult razorback suckers from below Hoover Dam each year as part of the captive breeding and release recovery program. If a discharge above or below Hoover Dam is chosen as the preferred alternative, and wastewater flows are diverted in increasing volumes, EDCs and abnormal hormones in fish will likely occur in the Colorado River downstream, as has already been documented in largemouth bass, common carp, and razorback sucker from Las Vegas Wash and Bay. Reproductive impacts at the population level are uncertain, however, any loss in reproductive ability for an endangered fish could jeopardize recovery efforts.

Section 101(a) of the CWA establishes the objective to restore and maintain the chemical, physical, and biological integrity of the Nation's waters. Primary to achieving this objective are the goals of achieving water quality that provides for protection and propagation of fish, shellfish, and wildlife and recreation in and on the water. The establishment of water quality standards (Section 303) and the issuance of discharge permits under the National Pollutant Discharge Elimination System program (Section 402) are primary mechanisms to attain goals of the CWA. As required under the CWA, water quality standards should be reviewed and, if appropriate, revised every three years. Water quality standards for Las Vegas Wash and Lake Mead were last revised in 1998 and are overdue. Existing conditions in Las Vegas Wash and Las Vegas Bay seem to indicate that due to increasing wastewater flows (point and non-point), past water quality standards may have been inadequate to preserve the chemical and biological integrity of water.

The goals of the Clean Water Act are clearly consistent with the goals of the Endangered Species Act (ESA). Sections 4(d) and 9 of the ESA prohibit the taking of fish and wildlife species listed as endangered or threatened. “Take” is defined under the ESA as “... to harass, harm, pursue, hunt, shoot, wound, kill, trap, capture, or collect, or attempt to engage in any such conduct.” “Harm” is further defined to include significant habitat modification or degradation that results in death or injury to listed species by significantly impairing behavioral patterns such as breeding, feeding, or sheltering (50 CFR § 17.3). The results of our investigation concur with previous studies and indicate conditions in Las Vegas Wash and Bay that are not conducive to healthy populations of fish and wildlife, including an endangered fish and possibly two endangered bird species. These conditions appear to constitute take as defined under the ESA and may approach the definition of adverse modification of critical habitat. Section VIII. B. of the Memorandum of Agreement Between the Environmental Protection Agency, Fish and Wildlife Service, and National Marine Fisheries Service Regarding Enhanced Coordination Under the CWA and the ESA states “... If the Services present information to USEPA, or USEPA otherwise has information supporting a determination that existing State or Tribal water quality standards are not adequate to avoid jeopardizing endangered or threatened federally-listed species or adversely modifying critical habitat or for protecting and propagating fish, shellfish, and wildlife, EPA will work with the State or Tribe in the context of its triennial review process to obtain revisions in the State and Tribal standards...” We believe this report, supported by Bevens *et al.* (1998 and 1996), constitutes evidence of adverse effects to listed species. Water quality standards for Las Vegas Wash and Lake Mead should be revised, as needed, to address water quality concerns identified in wildlife. Formal consultation on the proposed standards by USEPA should accompany this process.

ACKNOWLEDGMENTS

We are grateful to the following people who made valuable contributions to this report. The investigations were conducted in partnership with the U.S. Geological Survey Biological Resources Division. Key USGS personnel include Steve Goodbred, Ken Covay, Tom Leiker, Tim Gross, Carla Wieser, and Shane Ruessler. Scott Foott, Damian Higgins, Stanley Wiemeyer, Chuck Minkley, and John Miesner of the Fish and Wildlife Service also made substantial contributions. Bill Burke of the National Park Service provided assistance with coordinating and facilitating sampling efforts on Lake Mead NRA. Jim Heinrich and Mike Burell of the Nevada Division of Wildlife and Paul Abate, Jack Ruppert, and Cy Fernandez of the Southern Nevada Water Authority provided field assistance. Dan Fischer and Randy Hadland provided technical assistance with City of Las Vegas water quality data. Many of the aforementioned provided critical review of the manuscript. Finally, Paul Matuska and Theresa Thomas provided warm lodging, cold beer, and impartial and insightful conversation on the project.

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