METHODS

Nutrient Dynamics and Quality Issues

Water Quality

Water quality sampling assessed basic field parameters and nutrient loading. Assessment of water quality parameters coincided with zooplankton and benthic invertebrate sampling periods between 1999 and 2001. The following variables were assessed: temperature (degrees Centigrade, °C), salinity (parts per thousand, ppt), dissolved oxygen (D.O.; milligrams per liter, mg/L), and pH. Sampling periods were intended to reflect water quality conditions during periods of summer low water; managed flooding during the fall; and winter rain-associated flooding in the spring (Table 3). Sampling occurred in the following monitoring units: Reclaimed Water, Reclaimed Water + Muted Tidal, Muted Tidal, Passive Hydrologic Management, Groundwater Pond, and Undiked Marsh (Table 4). A maximum of 22 locations were sampled depending on water conditions, with fewer samples collected during the summer when some shallow water areas were dry (Figure 5).

In addition, analyses for nitrates, nitrites, ammonia, unionized ammonia, dissolved organic carbon (DOC), and chlorophyll a were conducted in eight (8) of the 22 sampling areas during 2000-2001 only (Table 3). The number of sampling locations were limited due to the expense of laboratory analyses and limited funding. These water quality samples were collected from created ponds in Reclaimed Water (Management Units 1 and 3, and Upland Pond 7), Reclaimed Water + Muted Tidal (Ringstrom Bay), Undiked Marsh (Hudeman Creek/Slough), Groundwater Pond, and Muted Tidal (BS and TOH; Table 4) units.

In selected sampling locations where very low water pH (<5) was observed, water samples were analyzed for alkalinity (carbonate), chloride, sulfate, and 13 metals, specifically silver (Ag), aluminum (Al), arsenic (As), cadmium (Cd), chromium (Cr), copper (Cu), dissolved iron (Fe-D), total iron (Fe-T), mercury (Hg), manganese (Mn), nickel (Ni), lead (Pb), selenium (Se), and zinc (Zn). These analyses were performed because, in other systems, extremely low water and sediment pHs have often been found to be associated with decreases in alkalinity and pulses in nutrients and metals (Delaune and Smith 1985; Soukup and Portnoy 1986; Gambrell et al. 1991; Peverly and Kopka 1991; Satawathananout et al. 1991; Gambrell 1994; Anisfeld and Benoit 1997; Portnoy and Giblin 1997). Sampling periods are shown in Table 3. For comparison purposes, samples were also collected from areas with moderate to high water pH (7-8). Sampling areas were principally in the Passive Hydrologic Management (MU2) and Muted Tidal (BS and TOH) monitoring units, with one moderate to high pH reference location in a Reclaimed Water monitoring sub-unit (MU3; Table 4; Figure 5).

Sampling Methodology

As waters in most sampling areas were well mixed due to shallow water depths and strong winds, only one measurement or sample within the water column was collected. Temperature, salinity, D.O., and pH were assessed in situ using a YSI 85 (YSI Inc., Yellow Springs, Ohio) system and an Oakton pHTestr3 (Oakton Instruments, Vernon Hills, Ill.). Water samples were
### Table 3. Sampling periods for monitoring activities conducted during the Hudeman Slough Enhancement Wetlands Case Study.

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Shaded cells indicate sampling period.

### Table 4. Monitoring activities conducted within monitoring units during the Hudeman Slough Enhancement Wetlands Case Study.

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Shaded cells indicate sampling conducted within monitoring unit.
Water quality sampling locations within the Hudeman Slough Enhancement Wetlands Case Study area.

- Water quality sampling locations only
- Water quality sampled for nutrients and metals
- Water quality sampled for acidification-related release of metals

**Dates of Photos:**
- July 1993 (Napa County)
- June 2001 (Sonoma County)

**Monitoring Unit Type**

- Hydrologically Managed
  - Reclaimed
  - Reclaimed & Muted Tidal
  - Muted Tidal
  - Passive
  - Groundwater

- Hydrologically Unmanaged
  - Undiked Marsh
  - Diked Marsh
  - Seasonal Pond

Figure 5.
collected concurrently for laboratory assessment of nutrients and other parameters. All water samples were immediately placed in a cooler and kept on ice at an average temperature of 4°C. Water samples were delivered to the laboratory on the same day that sampling was conducted.

**Laboratory Analysis**

Laboratory analyses were conducted by Alpha Analytical Laboratory (Ukiah, Calif.), with the exception of chlorophyll a (Sequoia Analytical Laboratory, Inc., Morgan Hill, Calif., and Brelje and Race, Inc., Santa Rosa, Calif.).

**Data Analysis**

Means and standard errors were calculated for each monitoring unit, both for the entire study period and for individual sampling periods. Results from metals analyses were separated and presented on the basis of water pH and alkalinity. Nutrient concentrations were compared with concentrations considered characteristic of natural and recycled waters (Alexander Horne, professor, University of California, Berkeley, unpub. data). In addition, nutrient concentrations were further evaluated using information on mean ammonia, nitrate, phosphate, and zinc concentrations in effluent from the SVCSD treatment plant between 1999 and 2001 (SVCSD, unpub. data). Dissolved oxygen and unionized ammonia concentrations were assessed using water quality objectives established by the San Francisco Bay Water Quality Control Plan (Basin Plan; RWQCB 1995) and, for the latter, acute and chronic criteria for freshwater (U.S. Environmental Protection Agency (EPA) 1987) and saltwater (U.S. EPA 1989). Unionized ammonia was also evaluated using total ammonia concentrations. Metals concentrations were assessed using the California Toxics Rule water quality criteria (U.S. EPA 2000), specifically the 4-day salt water criteria, as sampling areas were generally saline (marine-derived) in nature.

**Sediment Nutrients**

Soil sampling evaluated the concentration of organic and inorganic nutrients, as well as other abiotic variables that can affect nutrient uptake and plant health. Variables included pH (on dried soils), salinity (ppt), soil moisture (percent), organic matter (percent), Total Kjeldahl Nitrogen (TKN; parts per million, ppm), nitrates/nitrites (mg/L), ammonium (mg/L), phosphorous (ppm), potassium (ppm), magnesium (ppm), calcium (ppm), sodium (ppm), and cation exchange capacity [CEC, milliequivalents/gram (meq/g)]. Sampling was scheduled to occur three (3) times annually: November (just after reclaimed water units were drained following fall flooding), April/May (following winter rain-associated inundation), and August (dry season; Table 3). Sampling was conducted at 27 sampling locations in the following monitoring units: Reclaimed Water, Reclaimed Water + Muted Tidal, Muted Tidal, Passive Hydrologic Management, Groundwater Pond, Diked Marsh, Seasonal Pond, and Undiked Marsh (Table 4; Figure 6).

**Sampling Methodology**

Sediment nutrient monitoring was conducted by collecting soil samples from each end of the 27, 60.6-meter (m) (200-foot) long vegetation transects established for vegetation monitoring. Four 30-cm cores were randomly removed from an approximately 4-m area surrounding the end of the transect by dividing the sampling area into quadrangles and tossing the corer in each quadrangle
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Figure 6. Sediment nutrient and vegetation sampling locations within the Hudeman Slough Enhancement Wetlands Case Study area.

Dates of Photos:
July 1993 (Napa County)
June 2001 (Sonoma County)
to indicate the point of sampling. In some areas, 30-cm cores could not be taken due to the presence of a hard panne or impermeable layer within 30 cm of the soil surface. The soil samples were bagged and placed in a cooler. Samples were then stored in a refrigerator at 4°C until they could be processed. When water was present, either as surface ponding or within the core hole (porewater), pH was taken in the field using the Oakton pHTestr 3.

Soil salinity was determined by placing a sample of saturated soil from the top 20 cm of the core into a needleless 10cc plastic syringe loaded with two (2) layers of #2 Whatman filter paper and expressing a drop of soil water onto a salinity refractometer (range 0-150 ppt; Pacific Estuarine Research Laboratory 1990). If the soil was not field saturated, a soil paste was created according to the methods described by Richards (1954). After soil salinity was assessed, the remaining soil was weighed wet, allowed to dry to a constant weight at ambient temperature using a fan to accelerate drying, and then weighed dry to assess moisture loss upon drying. Soil moisture is expressed as a percentage by dividing weight loss by the dry weight of the soil sample (Gardner 1986). The soil sample was then placed in a plastic bag and sent to the laboratory for analysis. For three sampling periods (April, August, and November 2000), a portion of the wet sample was used to prepare potassium chloride (KCL) extracts for assessment of inorganic nitrogen (nitrates/nitrites and ammonium). These extracts were kept frozen until processing was completed and then delivered to the laboratory.

**Laboratory Analysis**

Laboratory analyses of KCL extracts were conducted by Alpha Analytical Laboratory, Inc. (Ukiah, Calif.). Laboratory analyses of pH, organic matter, Total Kjeldahl Nitrogen, phosphorous, potassium, magnesium, calcium, sodium, and cation exchange capacity was performed by A&L Western Agricultural Soil Laboratory (Modesto, Calif.).

**Data Analysis**

Means and standard errors were calculated for each monitoring unit for the entire study period. Means for individual sampling periods were also generated for ammonium, nitrate, phosphorous, and Total Kjeldahl Nitrogen. Principal Components Analysis (SYSTAT 8.0, SPSS Inc., Chicago, Ill.) was used to informally explore the distribution of sampling locations in relation to soil variables. Principal Component Analysis is a form of multivariate indirect gradient analysis that examines relationships between sites or species and selected variables. Three principal components were retained for analysis. Varimax rotation enhanced interpretation of the components.

**Sediment Contaminants**

Sediment contaminant sampling evaluated the concentration of trace elements and organic pollutants in sediments. Design of the sediment sampling program was intended to maximize comparability with the sediment contaminant sampling program currently being conducted as part of the San Francisco Estuary Institute (SFEI)’s Regional Monitoring Program (RMP), which includes several sampling locations in the San Pablo Bay area. Sediment sampling was conducted in August 1999, February 2000, and January 2001 (Table 3). Sediment sampling focused on metals, organochlorine pesticides (DDTs, HCHs, chlordanes), organophosphorous pesticides, polynuclear aromatic compounds (PAHs and alkylated PAHs), and polychlorinated biphenyls (PCBs). A list of all analytes can be found in the laboratory datasheets in the
Technical Appendices. In addition, sediment grain size, total organic carbon (TOC), ammonia, total solids, total sulfides, and pH were assessed. It should be noted that only metals and organochlorine pesticides were assessed for the August 1999 sampling.

Sediment contaminant sampling was conducted in seven (7) locations during each sampling period: three to four locations in the Reclaimed Water monitoring unit (Management Units 1 and 3 and Upland Ponds 3 and 7), one location in the Muted Tidal monitoring unit (BS); one to two locations in the Passive Hydrologic Management monitoring unit (MU2 BD and MU2 OS); and one location in the Undiked Marsh (HC Undiked Marsh; Table 4; Figure 7) unit. A one-time sample was also taken in August 1999 from an upstream portion of Hudeman Creek to assess the potential role of adjacent land uses in contaminant input, but as sediment texture was vastly different from the other areas, this sampling location was eliminated (Figure 7).

Sampling areas were chosen using several criteria, including current hydrologic dynamics and potential for historic exposure to pollutants. As hydrologically managed monitoring units are diked historic baylands adjacent to uplands, sources of contamination prior to discharge of reclaimed water include tidal flow either prior to or after diking, run-off from adjacent farmed uplands, and on-site contributions from past land uses such as farming or dairy operations. To the extent possible, we selected sampling areas that were as similar as possible in terms of potential for contaminant contribution (i.e., distance from San Pablo Bay, distance to adjacent uplands, presence of creeks within area, and past land use for diked areas). It should be noted that differences in the route of tidal flow from San Pablo Bay (Sonoma Creek for Hudeman Slough Enhancement Wetlands/Ringstrom Bay and Napa River/Dutchman’s Slough for Huichica Creek/Buchli Station Units) may affect the potential for historic contaminant contribution.

The Hudeman Slough Enhancement Wetlands have been flooded with reclaimed water since 1991. As concentrations of pollutants could be expected to be highest near the outfall of reclaimed water discharge pipes, sampling in the Reclaimed Water monitoring units were conducted near the pipe outfalls at Management Units 1 and 3 and Upland Ponds 3 and/or 7 (Figure 7). Upland Pond 3 was only sampled once in January 2001. The pipe outfall is located at the upland end of ditches or channels that were constructed to convey reclaimed water to the center of the management units. Reclaimed water storage within the reclamation storage reservoirs may affect the concentrations of trace metals and organic contaminants within waters discharged to enhancement wetlands, because during the storage period, these compounds may fall out of solution and settle into the reservoirs’ sediments (Jim Flugum, engineer, Sonoma County Water Agency, pers. comm.). However, during the Study Period, reclaimed water was pumped directly into MU3, but water was typically stored for some time in the R1 reclamation storage reservoir before it was discharged into MU1.

Sampling in the Passive Hydrologic Management Unit (MU2) was conducted in a secondary borrow ditch paralleling Hudeman Creek, which was historically exposed to tidal flushing, but is now mainly inundated with precipitation, upland run-off, and possibly saline groundwater (Figure 7). As discussed earlier, Management Units 2 and 3 were not fully leveed until some time after 1951. The USGS topographic map suggests that this portion of the borrow ditch may have been part of a historic slough that is still visible in the unit (Figure 4). At least once,
Figure 7. Sediment contaminant sampling locations within the Hudeman Slough Enhancement Wetlands Case Study area.

Dates of Photos:
- July 1993 (Napa County)
- June 2001 (Sonoma County)

Hydrologically Managed
- Reclaimed
- Reclaimed & Muted Tidal
- Muted Tidal
- Passive
- Groundwater

Hydrologically Unmanaged
- Undiked Marsh
- Diked Marsh
- Seasonal Pond

Sediment contaminant sampling locations
Sampling was also conducted in the center of the marsh plain on the edge of the historic slough channel (Figure 7). Depth of the secondary borrow ditch is roughly equivalent to depth of the ditches and channels that are sampled within Management Units 1 and 3. The sampling location in the secondary borrow ditch is at least 500 m from the primary borrow ditch that conveys reclaimed water from MU3 to Hudeman Slough during the discharge period.

Sampling in the Muted Tidal monitoring unit was performed at the upland extent of a muted tidal borrow ditch/channel in the Buchli Station Unit (Figure 7). Samples in the Undiked Marsh monitoring unit were collected from the uppermost arm of Hudeman Slough in the Huichica Creek Unit in a tidal channel near the upland edge (Figure 7).

**Sampling Methodology**

Prior to sampling, sediment sampling equipment was washed with Liquid Alconox (Liquinox), rinsed repeatedly with distilled water, washed with a nitric acid rinse, and given a final rinse with distilled water. Following the final rinse, sampling equipment for each of the seven sampling locations was bagged independently into 3 mil-thick plastic garbage bags, and the garbage bags were tied securely. Each set of sampling equipment included a high density polyethylene (HDPE) bucket, a stainless steel mixing spoon, and a 2-inch-diameter HDPE liner used for coring and removing sediment samples. New pairs of sterile gloves were used at each sampling location for field personnel in contact with the sample. At each sampling location, the top 5-cm of soil was removed by inserting the liner into the soil surface to a depth of at least 8 to 10 cm. The soil core was then manipulated so that only the top 5 cm was placed in the HDPE bucket, which was covered with aluminum foil during sampling to minimize atmospheric inputs. Replicate cores were removed from within a 1-m to 2-m radius of the sampling point, placed in the bucket, and homogenized in the field using a stainless steel mixing spoon. The homogenate was divided into aliquots for analysis, with additional aliquots prepared for sediment bioassay analysis. All sediment samples were immediately placed within a cooler and kept on ice at an average temperature of 4°C. Sediment quality samples were delivered to the laboratory on the same day that sampling was conducted.

**Laboratory Analysis**

Laboratory analyses were conducted by Sequoia Analytical Laboratory, Inc., in Petaluma, Calif., or sister laboratories. Certain portions of the analyses were subcontracted to other laboratory companies, specifically analyses for arsenic (Huffman Laboratories, Inc.; Golden, Colo.), particle size analysis (Environmental Technical Services; Petaluma, Calif.), and pesticides, PCB congeners, and alkylated PAH analyses (Axys Analytical Services, Ltd., Sidney, British Columbia, Canada). Analytical reports are provided in the Technical Appendices, Section I.

**Data Analysis**

Means and standard errors were calculated for each monitoring unit, both for the entire study period and for individual sampling periods. Standard errors presented for the entire study period represent variability between sampling locations rather than variability between sampling periods. Results were compared with data from the RMP on sediment sampling in San Francisco Bay, specifically San Pablo Bay. There are two RMP sampling locations in San Pablo Bay: one on the Napa River near Mare Island (BD22) and the other at the southern end of the Petaluma River (BD15). At the time this report was written, the most current data publicly available was
from 1999 (SFEI 2001). Results were also compared with those from a study evaluating contaminant concentrations in San Francisco Bay and San Pablo Bay marshlands, specifically a RMP pilot study that sampled China Camp and Petaluma Marsh in 1995-1996 (Collins and May 1997).

Also incorporated into analyses are sediment quality guidelines developed by Long et al. (1995) and the SFRWQCB. In general, the Long et al. (1995) guidelines (National Oceanic and Atmospheric Administration’s [NOAA] Sediment Quality Guidelines) are used by SFEI’s RMP for informal screening purposes, as no Basin Plan for sediment contaminant concentrations exists for San Francisco Bay (SFEI 1999). The guidelines were developed using information on contaminants from numerous studies conducted in the United States and identified concentrations of contaminants that were associated with biological effects in the laboratory, field, or in modeling studies. The Effects Range-Low (ERL) and Effects Range-Median (ERM) values are the concentrations equivalent to the lower 10th percentile and the 50th percentile of the compiled study data (Long et al. 1995). Sediment concentrations below the ERL are interpreted as being “rarely” associated with adverse effects, while those in-between ERL and ERM are “occasionally” associated, and those above the ERM are “frequently” associated (Long et al. 1995). The SFRWQCB Ambient Sediment Criteria (ASC) guidelines target an upper limit for ambient or current background conditions of contaminants, using the 85th percentile of reference or ambient San Francisco Bay concentrations (SFEI 1999).

Cluster analysis was used preliminarily to explore whether monitoring units could be separated on the basis of sediment contaminants. A total of nine (9) sampling locations were classified according to 32 sediment contaminants, including 13 metals, total DDTs, total HCHs, total chlordanes, total LPAHs, total HPAHs, total alkylated PAHs, and total PCBs. As noted earlier, three (3) of the nine (9) sampling locations were only sampled once (Upland Pond 3, MU2 OS, and HC Undiked Marsh). Euclidean distance and the average linking method of hierarchical clustering were used to produce the best clustering results, which were defined as the least amount of sequentially added small clusters or individual sites that resulted in one or two large clusters. Cluster analysis was performed using SYSTAT 8.0 (SPSS Inc., Chicago, Ill.).

**Sediment Bioassays**

Sediment bioassays further evaluate the issue of sediment toxicity by exposing laboratory-grown invertebrates to sediments from monitoring areas to determine whether pollutant concentrations might affect survival. The RMP employs a sediment bioassay procedure that exposes amphipods (*Eohaustorius estuarius*) to whole, homogenized sediment for 10 days with percent survival as the endpoint. During two of the sampling periods (February 2000 and January 2001), the laboratory noticed large drops in pH (pH<5.0) of waters overlying the sediment in some of the samples. For these sampling locations, the laboratory performed an additional procedure in which bioassay tests were re-run with pH adjustment of both the sediment and overlying waters to screen for potential effects of low pH on survival of the organisms.

**Sampling Methodology**

Sediment samples for bioassays were collected simultaneously with sediment collected for contaminant analysis (see Sediment Contaminants above; Tables 3 and 4; Figure 7). All sediment samples were immediately placed within a cooler and kept on ice at an average