Tidal Marsh Restoration Benefits Leopard Sharks (*Triakis semifasciata*) in South Bay Salt Pond Restoration Project Ponds.

Final Report

Prepared by:

James Hobbs, Jonathan Cook, & Patrick Crain, Biologist
Department of Wildlife, Fish, and Conservation Biology
University of California-Davis

Prepared for:

Resource Legacy Fund
555 Capitol Mall, Suite 1095
Sacramento, Ca 95814
&

South Bay Salt Pond Restoration Program Don Edwards San Francisco Bay
1 Marshlands Rd.
Fremont, CA 94555

INTRODUCTION

The South Bay Salt Pond Restoration Project (SBSPRP) is one of the largest tidal marsh restorations in the United States. Located within the southern arm of the San Francisco Estuary, the project is returning approximately 15,000 acres of former commercial salt production ponds to a rich mosaic of tidal wetlands and other habitats (**EDAW et al. 2007 EIR/EIS Report**). The restored ponds provide habitat for large numbers of migratory waterfowl and shorebirds, as well as several ESA listed species such as the Western snowy plover, California clapper rail, and the salt marsh harvest mouse (**Takekawa et al. 2001; Warnock et al. 2002; Strong et al. 2004**). To support diverse number of avian species and aquatic organisms the SBSPRP is using a mosaic design that incorporates local physical processes to support the structure and function of existing habitats, while restoring salt ponds to tidal wetlands. Pond restoration types include fully-tidal ponds, muted-tidal ponds and managed-ponds. Fully-tidal ponds were opened to tidal influence by excavating one, or in some cases multiple, breaches in the surrounding levee. These naturally fluctuating ponds typically de-water to outer channels (adjacent sloughs) on low tides and then completely fill, overtopping the marsh plane within the pond on high tides. Muted-tide ponds utilize water control structures to maintain pond levels at a minimum depth. The purpose of muting the tidal depth is to maintain habitat for wading birds and waterfowl while allowing tidal exchange with adjacent sloughs and the SF Bay. Water control structures are large enough to allow passage of mobile aquatic organisms, including fish, shrimp and crabs, to access the ponds, thereby creating opportunity for tidal refuge from the full exchange of the prevailing tides.

A variety of aquatic organisms, including invertebrates and fishes, have been found within restored salt ponds. Lonzarich and Smith (**1997**) found fifteen species of fish, including leopard shark, three species of annelid, seven species of crustaceans and a single species each of mollusk and insect. During a 2006 study, Saiki and Mejia (**2009**) reported 18 species of fish within the newly restored ponds A19, 20, 21, also known as the “Alviso island ponds.” Mejia et al. (2008) observed Leopard shark and other species moving from adjacent sloughs into and through managed wetlands, using water control structures for passage. Restored ponds have been found to contain high abundances of benthic invertebrates and small fishes, which provide abundant prey resources for leopard sharks entering the ponds.

Leopard shark (*Triakis semifasciata*), is one of the most abundant shark species of in the San Francisco Estuary (**Ebert 2003**; **Flemming 1999**). Their distribution ranges from Oregon to Mexico and they are found in nearly all benthic habitat types (**Ebert 2003**). In general, they are omnivorous, opportunistic benthic feeders, eating marine invertebrates, algae, vascular plants and fishes (**Russo 1975**; **Talent 1976**). Their diet also includes elasmobranch juveniles such as bat ray (*Myliobatis californica*), spiny dogfish (*Sqaulus acanthias*) and the brown smoothhound (*Mustelus henlei*) (**Russo 1975**; **Talent 1976**). Leopard shark take part in daily migrations onto shallow mud flats during flooding tides to feed (**Ebert 2003; Ackerman et al. 2000**; **Carlisle and Starr 2009**). This strategy is bio-energetically effective in shallow tidal habitats; however, recent restoration sites in San Francisco Bay have been designed with muted-tidal action, which may change leopard shark tidal movement behavior.

Few studies have been conducted on responses of fishes to habitat restoration in the San Francisco Estuary, and none have surveyed across different restored tidal pond configurations. (**Mejia et al 2008**; **Saiki and Mejia 2009**). As a part of the South Bay Salt Pond Restoration Projects adaptive management plan this study was undertaken to determine if leopard sharks utilize recently restored salt ponds in South San Francisco Bay. Monthly surveys were conducted among restoration sites with fully tidal, muted ponds, and adjacent tidal slough habitat. The survey examined the abundance leopard shark, other fish and invertebrate prey taxa, as a measure of response to restoration actions. In addition the diets of leopard sharks were evaluated in a fully-tidal pond, (pond E9-Eden Landing Marsh), a muted-tide pond (SF2-Ravenswood Marsh), a restored slough (Mt. Eden Creek) and a reference slough (Old Alameda Creek) to determine if the type 0f restoration influences diet composition, fullness, and condition factor.

STUDY AREA

South Bay Salt Pond Restoration Project.

The South Bay Salt Pond Restoration Project in collaboration with the California Department of Fish and Wildlife is restoring former salt production ponds to a mosaic of fully-tidal, muted and managed ponds in South San Francisco Bay. The restoration sites consisted of four tidal marshes which were acquired from Cargill Salt Inc. in 2000.

Eden Landing Marsh

The E9 complex, which consists of ponds E9, E8A and E8X, covers an area of approximately 630-acres and was reconnected to the bay in September 2011, during Phase I of South Bay Salt Pond Monitoring Program. The pond complex is fully tidal and bay water is exchanged through a breach located on its northwest corner. The breach opens to a channel which connects to Mt. Eden Creek, located just north of pond E9. Mt. Eden Creek was recently restored, with the main channel being widened and deepened (Enclosure 2, 2009). Old Alameda Creek is located south of E9 and was used as flood control for adjacent urban areas. As part of the restoration of the E9 complex, Pond A8A (south of E9 and hydrologically connected) was breached to tidal flows from Old Alameda Creek. In this study, Old Alameda Creek is a reference to recently restored sites, E9 and Mt. Eden. Eden Landing sampling was conducted at three main locations: the breach of E9, the mouth of Mount Eden Creek and Old Alameda Creek (Figure 1).

Ravenswood Marsh

SF2 is a 240-acre muted-tide pond designed to enhance habitat for foraging and roosting waterbirds (Figure 1). The complex consists of three distinct sections, with the landward section kept dry to provide nesting habitat for threatened Western snowy plovers. The middle and bay-ward sections contain 30 artificial nesting islands, designed to provide protected roosting and nesting habitat. Two water control structures connect the pond to the bay and maintain a minimum depth of ~1.5 m, by lessening the full range of the tides. Sampling for leopard sharks was focused around the most northern water control structure.

Bair Island

Bair Island, located on the western edge of San Francisco Bay, is a 3000-acre restoration site which consists of three main island components: Inner, Middle and Outer Bair (Figure 1). The complex of islands is surrounded by tidal mudflat and separated by three sloughs: Corkscrew, Steinberger and Redwood. Sampling for this project occurred at three sites, Outer Bair at the bay-ward breach, Corkscrew Slough and adjacent Steinberger Slough.

Alviso Marsh

Alviso Marsh is the largest restoration area associated with the South Bay Salt Pond Restoration Program (Figure 1). It consists of 9 former salt production ponds that are now reconnected to the bay. Ponds A21, A6 and A8 were chosen for sampling due to their close proximity to the bay and previously documented presence of leopard sharks. A6 is located on Alviso Slough and is approximately 330 acres in size. It was breached in fall of 2010 and is now fully tidal. Breached in March of 2006 Pond A21 is part of the 500 acre island pond complex, which includes A20 and A19. Fish sampling occurred at the breach, closest to the Bay, which connects A21 to Coyote Creek. The A8/A7/A5 complex is a series of managed ponds covering 1400 acres that was opened in fall 2009. Tidal control structures on A8 are operated seasonally to protect federally threatened Central California Coast steelhead. The water levels inside of A8 were managed for the majority of this study.

METHODS

Leopard sharks were sampled among the four tidal marsh systems, with each marsh having at least one restoration pond/type sampled 1 to 2 days per month from July 2010 to July of 2014. We were unable to sample all marshes in all months and most sites within marshes were sampled at random intervals. To capture large adult leopard sharks, multiple sampling techniques were used including gillnetting, hook-and-line sampling, and otter trawls were used to capture small juveniles. The otter trawl employed a four-seam design with a 1.5 m by 4.3 m opening and an overall length of 5.3 m. The mesh size was 3.5 cm in the body and 0.6 cm in the cod end. During sampling it was deployed and towed against the prevailing tide at a speed of approximately 5 kph for either 5 or10 minutes, with 10 minute tows occuring within major slough channels. At the completion of each tow, all organisms were collected and placed in a 50 gallon tub containing water from the sampling site.. Fish were identified to species, counted and the first thirty of each species measured for standard length. Macro-invertebrates (>1mm) were identified to the lowest possible taxa and counted. Three macro-invertebrate taxa (mysids shrimp, isopods and amphipods) were too numerous to count, thus rank abundance was recorded as and index where (0=no catch, 1 = 1-3 individuals, 2 = 2-10, 3 11-50, 4 = 51-100, 5 > 100, 6 >1000).

When sampling with gillnets an experimental net design with increasing mesh size was used to effectively target fish of multiple size classes. The specifications of its construction were as follows: overall the gillnet was 27.4 m long by 2.4 m deep, with 6 panels composed of 13 mm, 25 mm, 38 mm, 51 mm, 64 mm and 76 mm mesh. During a sampling event 1 to 3 gillnets were deployed, with their orientation perpendicular to the channel edge, with the smallest mesh in closest proximity to the shore. Sampling duration was 60-90 minutes, with timing centered around mid-ebb tide.

Hook-and-line sampling was conducted using frozen squid or baitfish as bait that was embeded on a 5/0 Gamakatsu circle hook and fished for approximately 1-3 hours. Angling efforts were focused near or within pond restoration levee breaches or water control structures to maximize predatory fish detections. Large predatory fish species tend to congregate around narrow contriction points where prey items are funneled during times of strong tidal flow.

After capture, leopard sharks were placed in an aerated holding tub and processed immediately to minimize handling stress. Weight, total length and sex were recorded. The stomach contents of a subset of fish were obtained by gastric lavage. Our lavage method utilized a hand pressurized sprayer filled with bay water. The tube of the sprayer was inserted into mouth of the shark and the stomach contents were washed into a fine mesh dip net. Contents were then placed into glass mason jars and fixed with 70% ethanol. Leopard sharks were marked using an anchor type identification tag (Floy) attached to the anterior portion of the dorsal fin. Each tag contained a unique numerical identification code that included a phone number for contact in the event of outside retrieval.

Catch data analysis

Catch among sites and between years was calculated using catch per unit of effort (CPUE) for each gear type. For hook and line sampling or angling, we report the CPUE as:

$$CPUE_a = \frac{\sum number\ of\ leopard\ sharks\ caught}{\sum total\ angler\ hours}$$

For gillnetting we use:

$$CPUE_g = \frac{\sum number\ of\ leopard\ sharks\ caught}{\sum\ total\ soak\ time\ (hrs)}$$

For otter trawling we use:

$$CPUE_o = \frac{\sum number\ of\ leopard\ sharks\ caught}{\sum total\ trawl\ time\ (hrs)}$$

Sampling effort and gear efficiencies differed among gear types and sites; therefore we report CPUE as an index only and did not conduct statistical analysis on count data. The presence or absence of leopard sharks in our sample was combined for all gear types as our catch metric and data is reported as occurrence or frequency of occurrence in the remainder of this document. The result is a loss of overall abundance data, but the species presence provides a catch variable that is less biased by gear efficiency, sampling effort and differences among habitat features that likely influence the ability to capture leopard sharks. Sampling locations within the tidal marshes were classified by restoration types (full-tidal, muted-tidal and adjacent sloughs) and combined by restoration type for comparisons.

Modeling

A generalized linear model using a binomial distribution and logit link function was used to test the null hypothesis that restoration type had no effect on leopard shark occurrence. The full model included 3 restoration types (muted, full-tidal and sloughs), survey year, season (4 levels), salinity (ppt), temperature (°C), and dissolved oxygen concentration (mg/L). Variables were analyzed for collinearity and removed prior to modeling. Models were analyzed sequentially beginning with the full model. Non-significant terms were dropped and the model was re-run until a best fit was determined. Akaike's information criterion (AIC) (Akaike 1969) was used to determine the best effects model, without interaction terms, using all possible subsets. It was not possible to add all interactions terms as sampling by survey month, year and site type were sampled unevenly. A separate model with survey month as a main effect revealed seasonal differences between winter months (Jan-Mar) and all other months for salinity, temperature and dissolved oxygen. The final model included season as a factor with only two levels, winter (Jan-Mar) and all other months (Apr-Dec), and pond restoration type (fully-tidal muted and slough habitat). Sex and size (length, weight) were not recorded for surveys conducted prior to 2013, as a result sex was not included as a variable in the full model. In addition Leopard shark occurrence was examined in response to water quality variables, including salinity, temperature and dissolved oxygen concentration, using a generalized additive model with a binomial distribution and a cubic spline smoothing function.

Diet

Diets from a subset of leopard sharks, collected between December 2012 and October 2013 from ponds SFS2 and E9, as well as Mt. Eden Creek and Old Alameda Creek sloughs were analyzed. Diet items were identified to the lowest taxonomic level possible and the wet weight of prey items was measured to the nearest 0.1g on an Acculab EC-4100 digital scale. Prey items were not individually enumerated in order to avoid introducing over-counting bias that may occur due to items differing in their degree of digestion. Prey identity, mass and frequency of occurrence were used to measure dietary composition and to calculate indices of dietary importance among restoration sites. Frequency of occurrence (FO_i%) was calculated by summing the number of individual shark stomachs that contained one or more individuals of each prey taxa (*i*) divided by the overall number of shark stomachs sampled per study site (*j*)(**Hyslop 1980**). Percent mass (M%) was calculated by summing the mass each prey taxa (*i*) divided by the total of all mass of all contents per study site (*j*) (**Hynes 1950**). These metrics were then combined to obtain the feeding importance index (FI_i%)

(**Kawakami 1980**). The feeding importance index (FI_i%) is an indication of the contribution of each item in the diet of the species, according to the following equation:

$$\boldsymbol{FIi}\% = \frac{\boldsymbol{FOi}\% * \boldsymbol{Mi}\%}{\sum_{i=1}^{n} \boldsymbol{FOi}\% * \boldsymbol{Mi}\%} * \mathbf{100},$$

where ***FIi%*** represents the feeding importance of an item *i*, ***FOi%*** the frequency of occurrence of the item *i* and ***Mi%*** the percent mass of the item *i*. The FI% index was also calculated for leopard shark diets collected in Elkhorn Slough in the 1970's (Talent 1976) and 1990's (Kao 2000).

The degree of dietary overlap between restoration sites was calculated using Morisita's index of similarity

$$\boldsymbol{C} = \frac{2 \cdot \sum_{i=1}^{n} p_{ij} \cdot p_{ik}}{\sum_{i=1}^{n} p_{ij}^2 + \sum_{i=1}^{n} p_{ik}^2}$$

Where p_{ij} = the proportion of prey *i* at site *j*, p_{ik} = the proportion of prey *i* at site *k*, and *n* = the number of prey categories for *j* and *k*. The degree of dietary overlap between restoration sites was determined using Langton's (1982) scale, for which values 0-0.29, 0.3-0.59 and ≥ 0.60 are ranked low, medium and high, respectively.

A relative stomach fullness index was calculated by dividing the mass of all stomach contents by the mass of the fish. Stomach fullness was compared between restoration sites with a generalized linear model using a poisson distribution. Due to relatively low sample sizes and unevenness of diet samples collected across sampling locations, season and restoration type, no interaction terms were included in the model. T-tests were used to test for differences in length among gear types and sexes of fish collected for diet analysis. An analysis of variance (ANOVA) was used to detect differences in length of leopard sharks (pooled sexes and gear types) among restoration sites. A chi-square test for goodness of fit (X^2) was used to determine if sex ratios were evenly distributed among restoration sites. All statistical models were performed in R version 3.0.2.

RESULTS

Catch

We collected 445 leopard sharks between 2010 and 2014 with a total of 358 hours of sampling using gillnets, angling and otter trawls for an overall CPUE of 1.2 fish per hour of sampling (Table 1). Gillnetting and angling via hook-and-line yielded similar CPUE while otter trawling collected fewer fish. Inter-annual patterns in CPUE were apparent among marshes. Sampling was unevenly distributed and inadequate in many marshes and between years to derive trends. In Alviso Marsh, the CPUE was lower in 2011 and 2012, where wetter years with higher freshwater flow and lower salinity may have precluded leopard sharks. In Eden Landing Marsh CPUE increased from 2011 to 2013, a period that spanned the restoration of pond E9. Overall CPUE was highest at Eden Landing, followed by Bair Island, Alviso and Ravenswood. To reflect leopard shark utilization of restored and adjacent slough habitats all sampling efforts for leopard shark were conducted either within pond restoration breaches or just outside pond breaches along the scoured channels created by pond discharge.,

Modeling

The model that best predicted the occurrence of leopard sharks within restoration site types included restoration type and season as factors. This model accounted for 74% of the overall variance in leopard shark occurrence and had the lowest AIC = 291.56 (Table 2). The main effects, restoration type and season, with just winter months (Jan-Mar) distinguished from the remainder of the calendar months, were highly significant model terms ($p < 0.001$). Year and Salinity variables were not significant and were removed from the model, while temperature and dissolved oxygen were highly collinear and varied seasonally (Figure 2). Leopard shark occurrence in our restoration sites was strongly influenced by season, with mean occurrence being lower during the winter months compared to the spring through fall months (Figure 3). This seasonal effect was also apparent across all restoration types. Leopard shark mean presence was approximately three times greater in full-tidal restoration ponds and adjacent tidal sloughs compared to tidally muted-ponds. In winter, full tidal ponds had the highest incidence of leopard sharks, while tidal sloughs exhibited the greatest change in mean presence between seasons.

A GAM model using salinity, temperature and dissolved oxygen as predictor variables explained 28.8% of the deviance of occurrence with a UBRE score of -0.66.

Salinity had a highly significant effect (p <0.001) on the occurrence of leopard shark with their occurrence dropping to zero at salinity values below 14-ppt.Temperature had a smaller, marginally significant effect (p = 0.02), with fish not found below 9.4 °C. Dissolved oxygen concentration was not a significant variable influencing the occurrence of leopard sharks, although leopard sharks were not encountered at dissolved oxygen concentrations below 4-mg/L (Figure 3).

The size distribution of leopard sharks varied among sampling methods, with otter trawls collecting smaller individuals ($\bar{x}$ = 303 mm TL, ±175 s.d.) than gillnetting and angling ($\bar{x}$ = 619 mm TL, ±238 s.d.) (ANOVA, F = 23.7, p < 0.001). Length distributions between gillnetting and angling were not significantly different (Bonferroni pairwise p = 0.26), nor were there differences between length distributions of either sex (t = -0.49 p = 0.62). This allowed us to pool all samples collected by gillnetting and angling, regardless of sex, to represent the size distributions of leopard sharks collected at our restoration sites (Figure 5). Overall length frequency distributions ranged from 37 cm to 109 cm TL (TL = total length) and were similar among restoration sites (ANOVA, F= 1.74, p = 0.16). The sex ratios (F:M) among the restoration sites were similar (X^2 = 1.3, p = 0.73). Condition factors were also similar between restoration sites and sexes (Two-Way ANOVA; restoration type F = 1.98, p = 0.12, sex F = 0.14, p =0.7) (Table 4)

Recruits

The lengths of leopard sharks collected by survey revealed a bi-modal distribution between February and December. Fish less than 40 cm were most likely young-of-year, born in the study area. Kushner et al. (1992) using validated vertebral increments, established the length of young-of-year leopard sharks in Elkhorn Slough to be less than 40 cm TL, thus fish less than 40 cm TL were considered young-of-year. The lengths of fish from monthly survey at all sites exhibited an increasing trend for the smallest size classes (< 40 cm) from March through October reflecting growth of young-of-year leopard sharks (Figure 6). Young-of-year recruits were found at all sites and were collected from March through October in Alviso Marsh.

Diet

A total of 165 individual leopard sharks diet contents were collected from four restoration sites; 80 shark diets were collected at Eden Landing Marsh from fish collected at the breach of pond E9, a fully-tidal pond, 18 sharks from Mt. Eden Creek, a recently restored tidal slough adjacent to pond E9, 32 sharks from Old Alameda Creek a reference slough and 35 sharks from the Ravenswood Marsh pond SF-2, a muted-tide pond. A total of 26 diet items were identified, which included multiple taxa which were not identified to species (Table 5). The most abundant organisms in the diets were multiple species of Polychaete worms, the majority of which were the *Streblospio benedicti*. Brachyuran crabs were the second most dominant taxa with the yellow shore crab, *Hemigrapsus oregonensis,* making up the majority of crabs. Fish taxa contributed to the diets with the longjaw mudsucker, *Gillichthys mirabilis,* being the dominant fish prey item by mass (Table 7).

Tidally Muted- SF2

Diets from 35 leopard sharks collected in December of 2012 contained high percentages of Polychaetes, (FI*i*%) 69%, making up the majority of prey items consumed, while fish contributed 12%, crabs 10% and shrimps 6%. The contribution of fish to the leopard sharks diets was greatest at the SF2 site relative to the other restoration sites.

Tidal-E9

Diets from 80 leopard Sharks were examined that were collected inside the breach at pond E9 between February and September of 2013 Two of the sharks were collected from February, 4 from March, 4 from April, 2 from May and the remaining 70 in September 2013. The inclusion of sharks caught from Feb-May had no influence on the overall diet patterns. Polychaetes and crabs made up over 80% of the diet, with dissolved organic material contributing 7% and all other taxa, except opisthobranchs, contributing to the diets.

Slough Samples- Mount Eden Creek and Old Alameda Creek

Diets were examined from 18 leopard sharks collected in Mt Eden Creek, just upstream of the E9 channel breach in Eden Landing, 7 sharks were collected from April, 4 from August and 8 from October 2013. The diet composition of leopard sharks from Mt. Eden Creek was dominated by polychaetes and crabs, while overall species composition was similar to the E9 site. Diets were examined from 25 Leopard sharks collected at Old Alameda Creek, 4 collected in April, 1 in May, 12 in August and 15 in October 2013. Diet composition differed the most for Old Alameda Creek with 60% of the diets consisting of crabs and 30% polychaetes, while other prey items contributed only 2-3%. Diet composition among restoration sites were similar, with values for the Morisita's Index of Dietary Overlap ranging from 0.97 between Mt. Eden Creek and E9, to 0.58 between Old Alameda Creek and the SF2. Diet composition was less similar between this study and previous leopard shark diet studies conducted in Elkhorn Slough in the 1970s by Talent (1976) and in the 1990's by Kao (2000). Diets examined in the earlier study were dominated by brachyuran crabs, principally the yellow shore crab, *Hemigrapsus oregonensis*, while in the later study to the dominant diet item was the fat innkeeper worm, *Urechis caupo* (Figure 7). Morisita's Index of Dietary Overlap was much lower between the present study and the Elkhorn Slough studies, with an index of 0.58 between this study and the 1970's study and an index of 0.23 for the 1990's study.

DISCUSSION

Leopard sharks, *Triakis semifasciata*, are dependent apon California coastal estuaries for nursery habitat, and are often the most abundant species of shark found within them (**Ebert 2003**). In the San Francisco Estuary, leopard sharks commonly occur throughout the mesohaline (>18-ppt) areas in the deeper channel habitats and shallow shoal habitats of North, Central and South Bays (**Ebert 1986**; **Flemming 1999**). In other estuaries leopard sharks are commonly found in eelgrass beds and in shallow sloughs of tidal marshes (**Ebert 2003**). The San Francisco Estuary lost the majority of its eelgrass beds and tidal marshes when these habitats were converted to salt production ponds in the early 1900's (**Atwater 1979**). Restoration of these salt production ponds has resulted in a variety of novel habitat types including fully-tidal ponds that are primarily intertidal and muted-tide ponds, which are managed to keep these habitats inundated during low tides. We encountered leopard sharks utilizing fully and muted-tidal ponds as well as adjacent sloughs in the Alviso, Bair Island, Eden Landing and Ravenswood Marshes. Leopard sharks were more abundant in the Eden Landing and Bair Island Marshes where salinity and dissolved oxygen concentrations were higher. High freshwater flows in winter reduced salinity below 14-ppt in Alviso Marsh, making it uninhabitable for leopard sharks during this season. Low dissolved oxygen concentrations (< 4mg/L) in summer may limit habitat availability as well. When leopard sharks were encountered in Alviso Marsh during summer, it was always in bay-ward stations containing higher salinity and higher dissolved oxygen levels.

Fully-tidal restoration ponds and adjacent sloughs supported similar abundances of leopard sharks, while in muted-tide ponds, leopard sharks were encountered less frequently. Leopard sharks were often found residing within the scoured channels created by the breaching of levees of fully-tidal ponds. Narrow levee breaches create constriction points where ebbing waters leaving the restoration site providing a “hot-spot” for predators to ambush prey species. This type of restoration design may facilitate trophic transfer of production up the food chain. Leopard sharks are mobile predators that eventually migrate out of the estuary, effectively connecting the pond restorations to the coastal ecosystem. The creation of predation “hot-spots” may also facilitate exploitation by non-native piscivorous species. During our surveys we encountered other predator species, including bat rays (*Myliobatus californica*), another native elasmobranch and striped bass (*Morone saxatilis*), an introduced sport fish. Striped bass are the top predator in the San Francisco Estuary and were extremely abundant in recent history (**Skinner 1962**). Their possible impact on native fishes is still a concern, despite recent declines in their population. Striped bass are known to be very opportunistic predators, taking advantage of man-made structures as ambush points, and are known to make frequent migrations between freshwater and the ocean. If they continue to congregate near levee breaches and water control structures they may potentially compete with leopard sharks or prey upon young of year sharks that are utilizing these ponds as nursery habitat.

Leopard sharks were encountered less frequently in muted-tide ponds. SF-2 in the Ravenswood Marsh is only 240-acres in size and is smallest muted-tide pond associated with this restoration project. The available habitat for leopard sharks in SF2 is likely even less, as much of the pond is only 1-m deep and fish were confined primarily to the remnant borrow ditch from construction of the original levee. The amount of available habitat, or lack thereof, may have influenced leopard shark abundance. A8 in Alviso Marsh is the largest pond restoration site (~1,400 acres) and is located at the base of the Alviso Marsh where freshwater from the Guadalupe River can lower salinity within the pond. Salinity in the wet winter of 2011-2012 was below 10-ppt and would have precluded leopard sharks from occupying the pond. Leopard sharks would also have to find passage through the water control structures to access the muted-tide ponds. This may explain the lower numbers of leopard sharks encountered. Flow through the water control structures of muted-tidal ponds is lower than that which occurs in fully-tidal breaches and may not attract leopards sharks into the vicinity of areas that were sampled. Lastly, dissolved oxygen concentrations in muted-tidal ponds during summer months can often drop below 4-mg/L nightly making these habitats inhospitable to leopard sharks. In Elkhorn Slough leopard sharks have been found to avoid habitats when dissolved oxygen concentrations drop below 4-mg/L (**Carlisle and Starr 2009**). Leopard sharks were encountered during summer months in muted-tidal ponds when dissolved oxygen concentration is typically low. They likely exit muted ponds via water control structures if and when conditions in adjacent sloughs become better than those within the ponds.

Leopard sharks are known to be omnivorous apex predators in estuaries and we found them to feed on a wide variety of taxa including several species of fish (**Russo 1975**). Diets were dominated by polychaete worms and brachyuran crabs across restoration types. A recent study found high densities of benthic invertebrates, including polychaetes, in SF2 (**Murphey 2013**). Brachyuran crabs and polychaetes were commonly encountered when sampling with otter trawls (*unpublished data*) and this trend was reflected in the diets examined in this study. Leopard sharks of similar size ranges fed on very similar prey taxa in the Elkhorn Slough Estuary (**Talent 1976**; **Kao 2000)**. We found a high degree of dietary overlap among restoration types and with diets of leopard sharks in Elkhorn Slough from the 1970’s. Fishes made up a higher proportion of prey items in diets collected from tidal-muted restoration ponds. The increased importance of fish in the diets in muted-ponds may be associated with a variety of physical variables, including reduced tidal flows, increased water clarity or overall reduced refuge habitat for prey fish. Prey fish abundance in the habitats we sampled for diet composition were generally low compared to other sites monitored, thus it is likely physical factors were more responsible for the increased importance of fish in leopard shark diets.

Leopard sharks give birth to young in shallow waters between March and June within the San Francisco Estuary (**Smith and Abramson 1990**). During the duration of this study, we encountered small leopard sharks (approx. 20cm) in otter trawls at Alviso Marsh and Bair Island Marsh beginning in March and peaking by June. Young-of-year leopard sharks were found in fully-tidal and muted-tidal restoration ponds. Large pregnant females captured in muted-tidal ponds highlighted the putative nursery habitat functions provided by the restoration actions. During summer months however, water conditions within ponds may prove too stressful for young leopard sharks. Thus the actual function of muted-tidal ponds as nursery habitat remains unconfirmed.

CONCLUSION

From July 2010 to July 2014 we conducted over 500 hours of sampling using gillnets, angling and otter trawls over 50 months and caught 445 leopard sharks from multiple age classes during all months of the year. Leopard sharks occurred more frequently in tidal sloughs and fully-tidal restoration ponds compared to muted-tidal ponds. Diet compositions, condition and stomach fullness was similar among restoration sites. This suggests restoration of former salt production ponds has provided approximately 2,000 acres of novel nursery habitat for leopard sharks. We caution further construction of muted-tidal pond restorations due to frequent hypoxic conditions during summer months when young sharks would be utilizing these habitats. The sharks' ability to escape these ponds when conditions become stressful is currently unknown. Furthermore, muted-tidal ponds can have significant effects on habitat conditions where physical factors can increase the likelihood of large fluctuations in salinity, temperature and dissolved oxygen concentrations.

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TABLES

Table 1. Summary of catch results for the three gears used to capture Leopard Sharks from 2010 to 2014 at the four restoration marshes.

	Angling			**Gillnet**			**Otter**			
	N	Effort (Hr)	$CPUE_a$	N	Effort (Hr)	$CPUE_g$	N	Effort (Hr)	$CPUE_o$	Σ CPUE
2010										
Alviso	11	6	1.8	-	-	-	3	5	0.7	2.5
Bair	4	4	1.0	-	-	-	4	2	1.7	2.7
Eden	-	-	-	-	-	-	0	0	0.0	0.0
Ravenswood	-	-	-	-	-	-	0	0	0.0	0.0
2011										
Alviso	6	10	0.6	1	12	0.1	0	20	0.0	0.7
Bair	8	6	1.3	12	5	2.4	3	4	0.8	4.5
Eden	2	1	2.0	2	1	2.0	0	1	0.0	4.0
Ravenswood	-	-	-	9	13	0.7	0	0	0.0	0.7
2012										
Alviso	2	9	0.2	3	40	0.1	0	25	0.0	0.3
Bair	19	10	1.9	31	20	1.6	2	6	0.4	3.8
Eden	7	5	1.4	44	7	6.3	0	1	0.0	7.7
Ravenswood	-	-	-	34	6	5.7	-	-	-	5.7
2013										
Alviso	15	17	0.9	12	26	0.5	4	17	0.2	1.6
Bair	1	3	0.3	0	2	0.0	0	1	0.0	0.3
Eden	80	17	4.7	165	24	6.9	7	4	1.8	13.4
Ravenswood	-	-	-	2	5	0.4	-	-	-	0.4
2014										
Alviso	14	5	2.8	0	10	0	10	10	1.05	3.9
Gear-type Total	169	93	1.82	315	171	1.84	33	94	0.35	

All Years	$\bar{x}$ CPUE
Alviso	8.9
Bair	11.4
Eden	25.1
Ravenswood	6.8

Table 2. Generalized linear models and explanatory variables for leopard sharks encountered in South Bay Salt Pond Restoration Project studies.

Model	Parameters	df	Res Dev	AIC
1	Year + Season + Restoration Type + Salinity	223	316.74	293.66
2	Season + Restoration Type	225	283.76	292.38
3	Season (W,Sp-F) + Restoration Type	226	279.66	283.56

Table 3. Model parameter estimates for pond restoration type and season.

Parameters	Estimate	Std. Error	z value	Pr(>|z|)
Muted-Tide Pond	-1.21	0.36	-3.36	0.00078 ***
Slough	1.50	0.44	3.45	0.00057 ***
Full-Tide Pond	1.52	0.41	3.74	0.00019 ***
Season (W, Sp-F)	-1.42	0.48	-2.93	0.00342 **

Table 4.. Leopard sharks numbers, condition, sex ratios and number of fish tagged during diet surveys at four restoration marshes in South San Francisco Bay.

Complex	Number Caught	Mean Size(cm)	K (1sd)	Female	Male	Sex Ratio F/M	Tagged	Recaps
SF2	35	68	0.04 ±0.01	17	18	49/51	35	0
E9	80	69	0.05 ±0.01	42	38	53/48	58	1
Mt. Eden Creek	18	71	0.03 ±0.02	9	9	50/50	14	0
Old Alameda	32	74	0.04 ±0.01	14	18	44/56	13	0
Total	165	70.5	0.04 ±0.01	81	75	1	120	1

Table 5. Leopard shark prey items identified from stomach content analysis and frequency of occurrence (FO%), mass (M%) and feeding imporance index (FI%) summed across all leopard sharks examined.

Prey Identifications	Scientific Name	Prey Categories	FO%	M%	FI%
Polychaetes		Polychaetes	73.94	27.99	50.660
Yellow shore crab	*Hemigrapsus oregonensis*	Crab	63.03	24.10	37.179
Dungeness crab	*Metacarcinu magister*	Crab	5.45	11.82	1.578
Longjaw mudsucker	*Gillichthys mirabilis*	Fish	2.42	9.32	0.553
ghost shrimp	*Neotrypaea californiensis*	Ghost Shrimp	3.64	5.56	0.495
Dissolved organic material		DOM	46.67	4.67	5.336
Bay shrimp	*Crangon franciscorum*	Shrimp	25.45	4.21	2.623
Topsmelt	*Atherinops affinis*	Fish	1.82	4.08	0.182
Yellowfin goby	*Acanthogobius flavimanus*	Fish	4.85	2.76	0.328
Fish fragments		Fish	6.06	2.56	0.379
Plant material		Plant	26.67	0.66	0.433
Shrimp fragments		Shrimp	3.03	0.60	0.045
Algae		Plant	16.36	0.44	0.175
Eggs		Eggs	0.61	0.29	0.004
Crab fragments		Crab	2.42	0.23	0.014
Northern anchovy	*Engraulis mordax*	Fish	0.61	0.23	0.003
Clam fragments		Clam	1.82	0.15	0.007
English sole	*Parophrys vetulus*	Fish	1.21	0.12	0.004
Pacific herring	*Clupea pallasii*	Fish	0.61	0.05	0.001
Oriental Shrimp	*Paleomon macrolepitodus*	Shrimp	0.61	0.05	0.001
Pacific staghorn sculpin	*Leptocottus armatus*	Fish	0.61	0.04	0.001
Opistobranchia		Opisthobranch	0.61	0.04	0.001

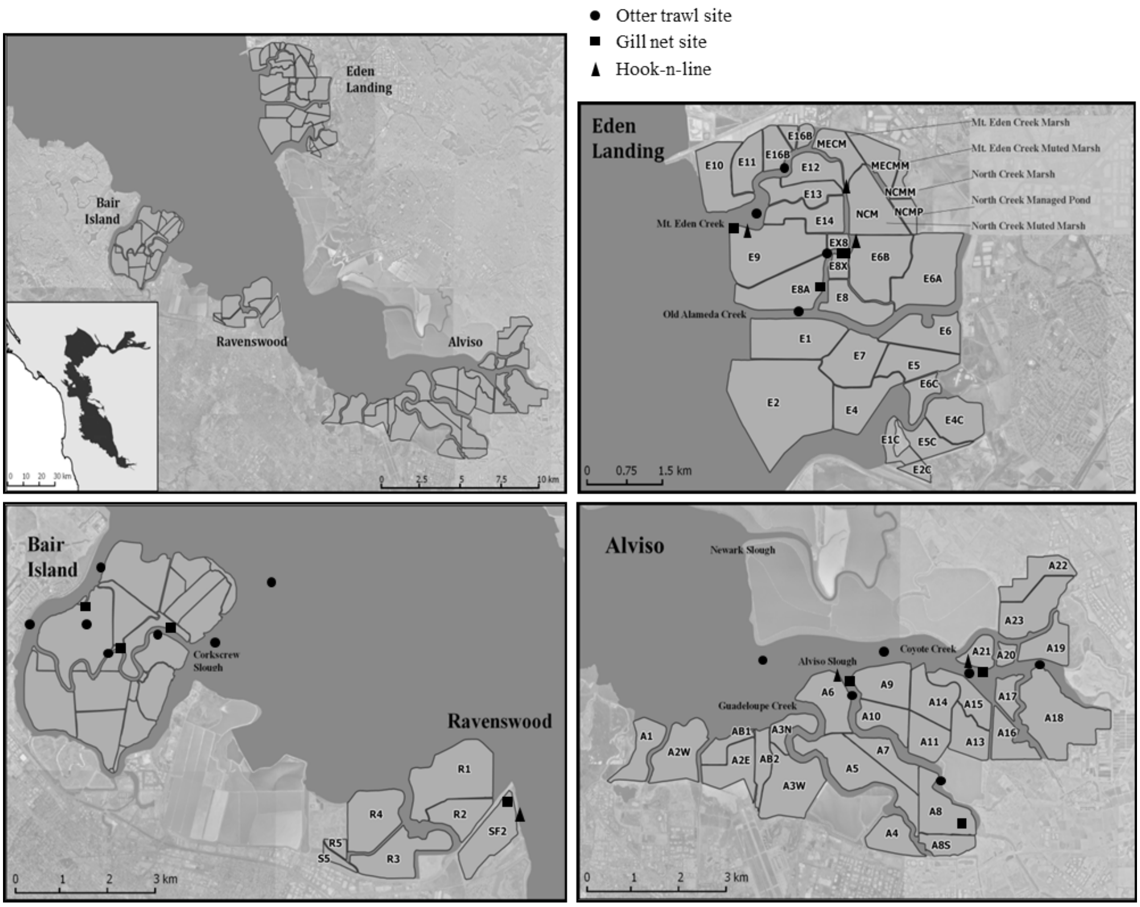

Figure 1. Map of the South Bay Salt Pond Restoration Project marshes (Top Left). Eden Landing Marsh (Top Right), Bair Island and Ravenswood Marsh (Bottom Left) and Alviso Marsh (Bottom Right). Gear types and sampling sties depicted by black symbols.

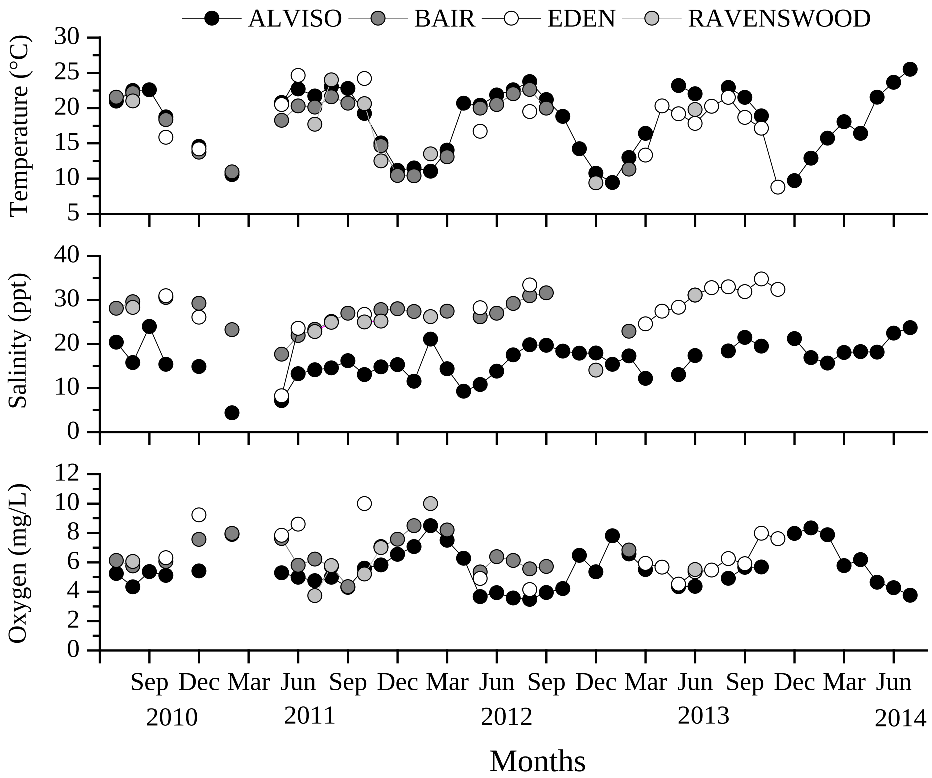

Figure 2. Monthly averages of water quality parameters, water temperature °C, salinity ppt, and dissolved oxygen mg/L among the South Bay Salt Pond Restoration marshes from July 2010 to July 2014.

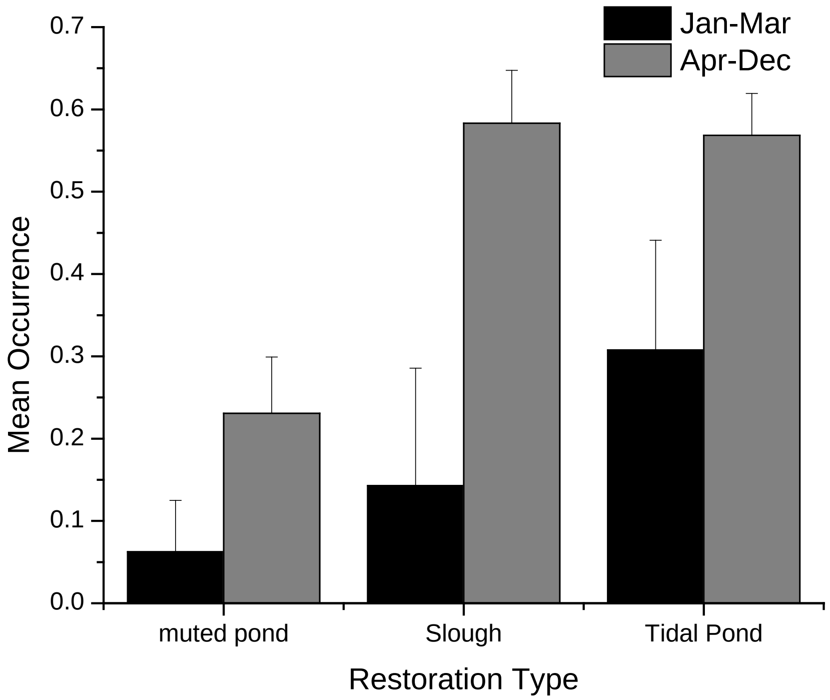

Figure 3. Mean occurrence of leopard sharks from combined otter trawls, gillnetting and angling efforts from July 2010 to July 2014 among restoration types and seasons aggregrated into winter months (January through March) and all other months spring through fall.

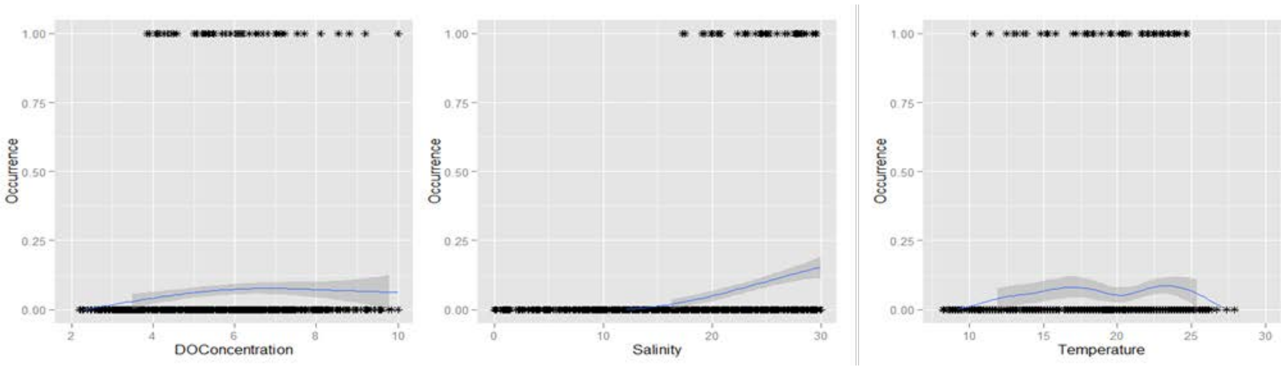

Figure 4. The presence (1) or absence (0) of leopard sharks in restoration sites and the water quality variables measured during each sampling survey. Data includes all gear types and all stations within the four restoration marshes from July 2010 to July 2014. N -1,225 samples.

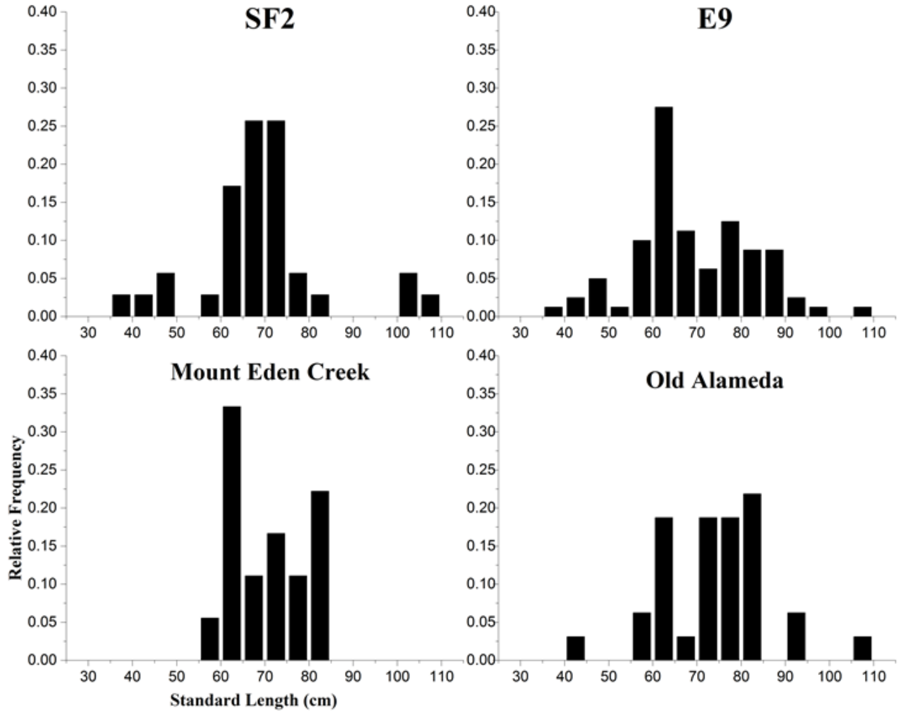

Figure 5. Length-Frequency distribution for leopard sharks collected at the four diet study locations.

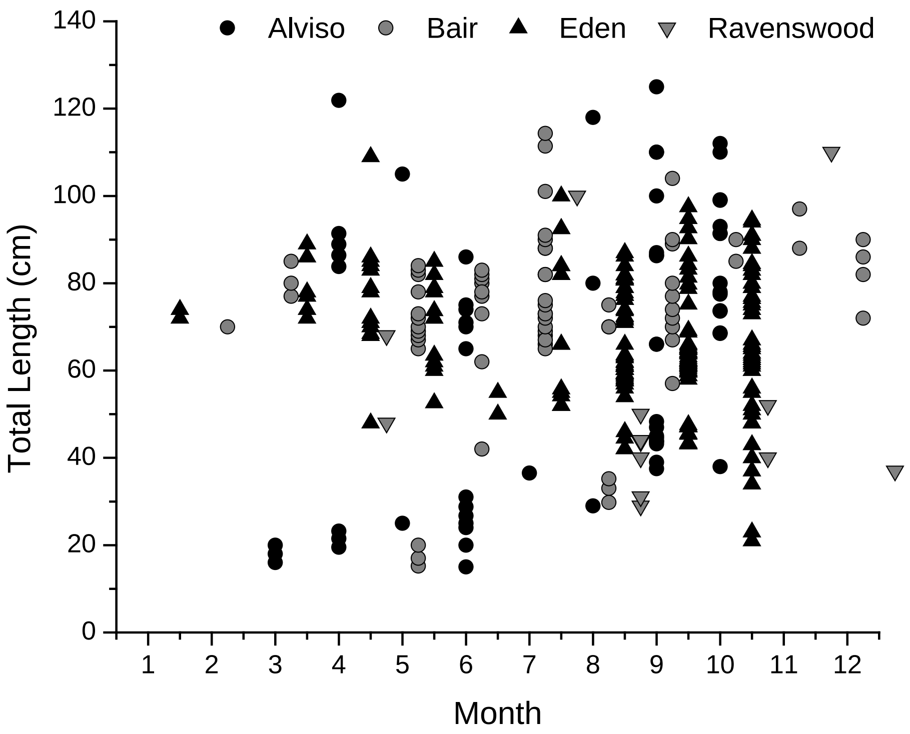

Figure 6. Lengths (cm) of leopard shark collected at the four marshes using all gear types in the study. Fish less than 40-cm are young-of-year recruits. Marshes are jittered to facilitate viewing all datapoints.

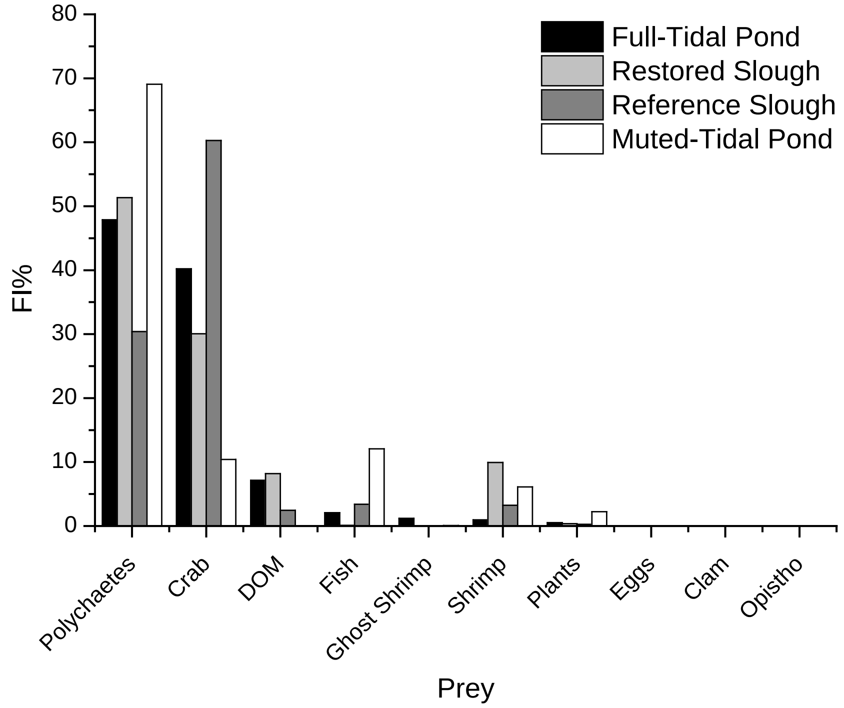

Figure 7. Feeding importance index of prey categories for leopard shark collected at the four restoration in the South Bay Salt Pond Restoration Project .

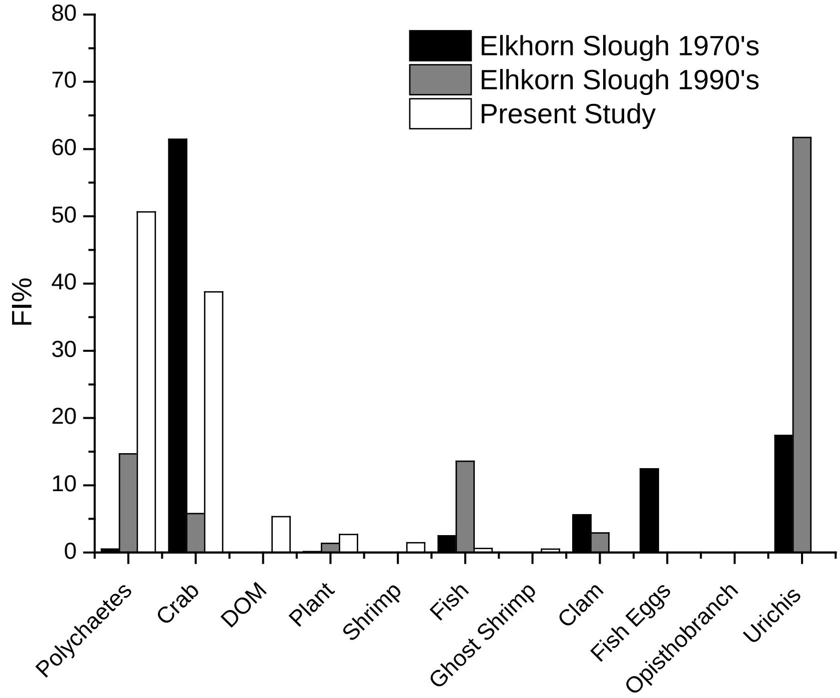

Figure 8. Feeding importance index of prey categories for leopard shark collected this study and for leopard sharks collected in the 1990’s (Kao 2000) and 1970’s in Elkhorn Slough, Ca (Talent 1976)

APPENDIX

Tag return

We tagged a total of 120 individuals during sampling surveys at Ravenswood and Eden Landing in 2013. A single leopard shark was recaptured by a recreational fisherman at Candlestick Point in San Francisco on July 12th, 2014. The male shark, measuring 75-cm TL, was tagged on October 19, 2013 at the Old Alameda Creek site. The distance between the tagging and capture locations was over 27-km (Figure A1). This individual grew 14-cm in length while at liberty. Based on the data reported by the recreational angler, the approximate growth rate would have been ~1.5 cm per month. However, there is some question as to the accuracy of this estimate as previous work has reported growth rates to be much slower than those found here and we cannot confirm the size of the fish at recapture.

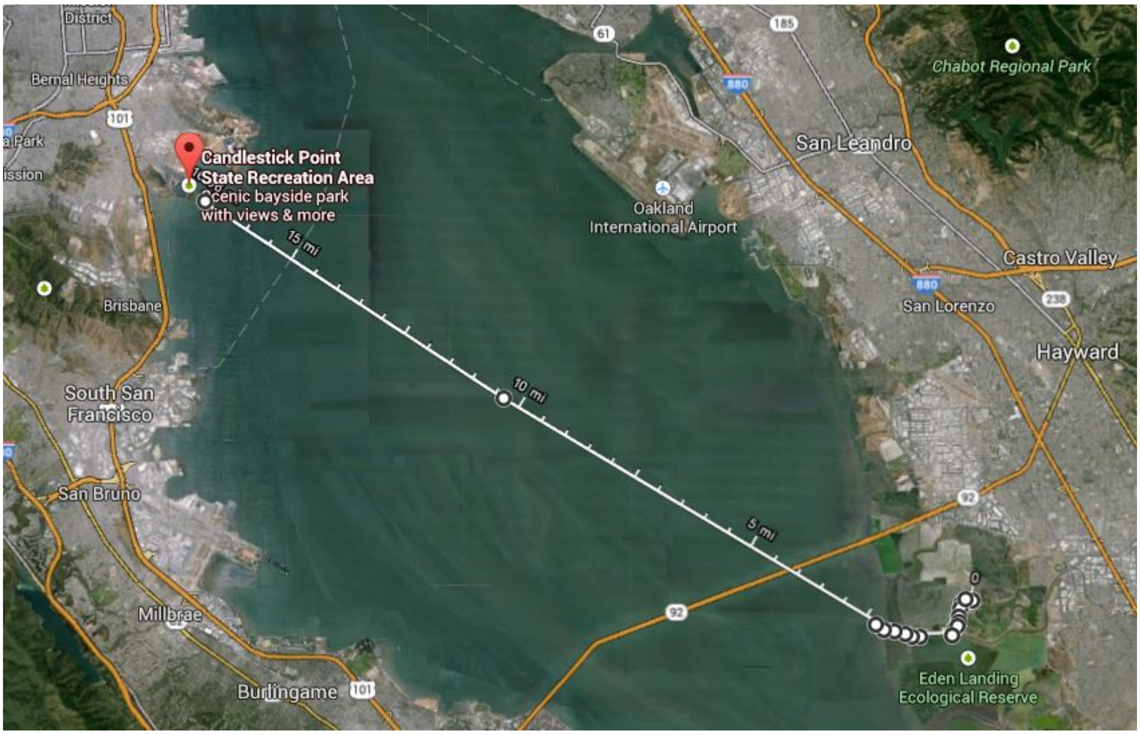

Figure A1: Depiction of Leopard Shark movement. It was tagged on October 19th, 2013 and recaptured on July 12th, 2014. Total distance ~27km.