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Photo by Athena Macguire, California Department of Fish and Wildlife

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Notes from the Editor

This edition of the Fish and Game Journal brings two species notes and several marine articles to the scientific literature. One article, the inspiration for the cover photo, describes the economic value of the red abalone fishery. Other articles inform management of two economically important marine fisheries, the California halibut, and the Olympia oyster. The halibut was found to have differences in size and age of reproduction between the southern California population and the central California coast population which has implication for its management. Opportunities for restoration of the oyster were confirmed if suitable habitat is made available. Collectively, these articles highlight the importance of our natural resources to our economy and our society. Scientists investigate and report these co-benefits to managers who use the science to set management goals or to design strategies to enhance or augment natural conditions for the improvement of the resource. Without science we'd be guessing, and much more susceptible to population declines, over-exploitation, and extinction. It is therefore incumbent upon us as scientists, to keep working, keep studying, and keep reporting what we see and find. The contributions made to this journal help shape the future in many small but meaningful ways through publication of our scientific endeavor which will, it is hoped, have lasting benefits for future generations.

With the acquisition of new publishing software, discrepancies developed in the direction given to authors by Bleich et al. (2011), related to the format of tables and figures submitted with manuscripts to the Fish and Game Journal. For clarification, in-lieu of any previous direction provided to authors, tables and figures should be submitted as PDF, TIF or JPG formats. Microsoft PowerPoint format will not be accepted.

The California Fish and Game Journal is pleased to welcome Neil Clipperton, the Department's non-game bird coordinator, as a new associate editor.

Armand Gonzales
Editor-in-Chief
California Fish and Game

Front—. Red abalone (*Haliotis rufescens*). Photo by Athena Macguire, California Department of Fish and Wildlife. It is estimated red abalone has a worth of $24-$44 M in annual non-market benefits to recreational fishers in California (Reid et al. 2016).

Rear—. Desert tortoise (*Gopherus agassizii*). Photo by Jeff Mitchell. The desert tortoise is listed as threatened with extinction by both the State of California, Department of Fish and Wildlife, and the U. S. Fish and Wildlife Service.

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Assessment of length- and age-at-maturity for California halibut (*Paralichthys californicus*), including a histologically-based description of the reproductive cycle

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Estimates of length- and age-at-maturity for California halibut (*Paralichthys californicus)* have been reported for southern California, but not central California. To provide new estimates of length- and age-at-maturity for central California halibut, we macroscopically examined gonads from 635 fish caught between 2012 and 2014 and additionally examined ovaries histologically. We developed a detailed description of the reproductive phases and spawning states for California halibut, and assigned sex-specific length- and age-at-maturity to each individual. Males (n=333) ranged from 19.1 to 95.9 cm fork length (FL) and 1 to 16 yr of age and females (n=302) ranged from 18.6 to 111.0 cm FL and 1 to 19 yr of age. Males matured at younger ages and shorter lengths than females. The smallest mature male was measured at 25.7 cm (1 yr), 50% of males were mature by 27.0 cm (1.1 yr), and 100% were mature by 29.0 cm (3 yr). The smallest mature female was measured at 46.6 cm (2 yr), 50% of females were mature by 47.3 cm (2.6 yr), and 100% were mature by 51.3 cm (4 yr), according to histological criteria. Therefore, all California halibut examined were mature before reaching the commercial and recreational minimum legal size limit of 55.9 cm (22 in). When comparing central California maturity data with information from southern California, we found that central California halibut matured at larger sizes (both sexes) and older ages (females only) than southern California halibut, according to macroscopic criteria.

Key words: California halibut, *Paralichthys californicus*, length-at-maturity, age-at-maturity, reproductive cycle, minimum legal size limit, histology, spawning state, batch spawner, flatfish

California halibut (*Paralichthys californicus)* are most commonly encountered from Bodega Bay, central California to Bahía de San Quintín, northern Baja California (Rosales-Casián 1996), although their geographic range extends from the Quillayute River in the state of Washington, USA, to Cabo Falsa in southern Baja California, Mexico (Fitch and Lavenberg 1971, Feder et al. 1974, Allen 1990, Martinez-Muñoz and Ramírez-Cruz 1992). This large predatory flatfish has supported important commercial and recreational fisheries in California since the early 1900s (Frey 1971, Allen 1990, Kramer et al. 2001). Because of its great economic and ecological importance (Allen 1990), California halibut is considered a high priority species for life history research by the California Department of Fish and Wildlife (CDFW), which manages these fisheries for long-term sustainability.

California halibut are oviparous, broadcast spawners that exhibit external fertilization (Allen 1990). This reproductive strategy involves the release of gametes (i.e., eggs and sperm) directly into the water column (Cailliet et al. 1986) where, in the case of California halibut, fertilized eggs develop into pelagic larvae (Frey 1971, Allen 1988). California halibut are batch spawners (Caddell et al. 1990) that release hydrated (i.e., fully developed) ova during reproductive events, while less developed oocytes remain in the ovary and mature for release at a later date (Cailliet et al. 1986, Murua et al. 2003). Thus, ovaries of mature California halibut always contain oocytes in various developmental stages, even after a spawning event has occurred. Although data to evaluate spawning seasonality off the central California coast are limited, a peak in reproductive activity has been observed within the summer months between Monterey and San Luis Obispo (Barnes et al. 2015).

Biological data, including reproductive strategy and timing of maturation, aid fisheries managers in evaluating the effectiveness of minimum legal size limits (Reed and MacCall 1988, Maunder et al. 2011). Since the 1970s, take of California halibut measuring less than 55.9 cm (22 inches) has been prohibited in all California fisheries. One of the intended purposes of this minimum legal size limit was to allow at least 50% of the California halibut population to reach maturity before becoming susceptible to take. Although available estimates of California halibut length-at-maturity suggest that the minimum legal size limit effectively protects immature individuals from take by the fishery along southern California (Love and Brooks 1990), no maturity studies have been conducted for the central California region. Because spatially varying environmental conditions and different degrees of fishing pressure can result in different rates of maturation (e.g., Packer et al. 1999, Yoneda et al. 2007), it is important to construct region-specific estimates of length- and age-at-maturity throughout a species' range.

Here, we estimate length- and age-at-maturity for central California halibut and provide a preliminary assessment of regional differences in maturation by comparing data from central California with those previously collected along the Southern California Bight (Love and Brooks 1990). We also present the first detailed description of the reproductive cycle for California halibut based on histology and include a macroscopic guide for use in assigning spawning states during field-based research.

Materials and Methods

Sample collections.—California halibut were collected from recreational, commercial, and research fishing activities off central California (i.e., north of Point Conception). Almost all fish were collected inside San Francisco Bay and from nearshore waters adjacent to Santa Cruz, Moss Landing, Monterey, Morro Bay, and Port San Luis (Figure 1).

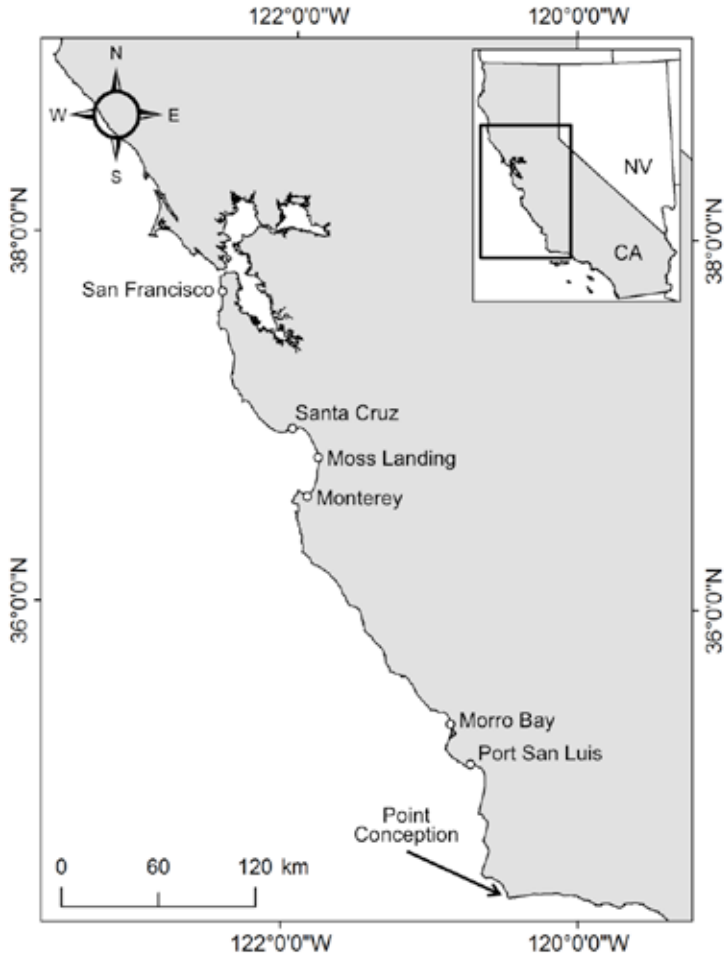

FIGURE 1.—Primary locations used to collect California halibut in nearshore waters off central California. Point Conception denotes the boundary between central and southern California.

In San Francisco Bay, California halibut were collected opportunistically by CDFW staff from July 2012 to November 2014. California halibut were not collected during the months of January and February, and only one sample was collected in December 2012. This was due, in part, to a lack of fishing effort during winter. Fish were obtained from two research trawl vessels, a commercial bay shrimp trawler, and a Commercial Passenger Fishing Vessel (CPFV) using hook-and-line gear, and fishery-independent researchers using hook-and-line gear.

From Santa Cruz, Moss Landing, Monterey, Morro Bay, and Port San Luis, California halibut were collected opportunistically between June 2012 and November 2013; these methods are further described in Barnes (2015). The majority of fish from these areas were caught during the summer months (i.e., June to August), when

California halibut are known to be reproductively active in these areas (Barnes et al. 2015). Samples from fish of legal size were collected from recreational fisheries that used hook-and-line and spear, and from the commercial hook-and-line fishery.

Laboratory processing.—California halibut were examined as fresh dead specimens. Fork length (FL, cm), body weight (kg), and eyed-side were recorded. Pre-filleted fork lengths for fish received as filleted carcasses were calculated using the equation: 0.137 + (0.99 * post-filleted FL); $R^2 = 0.999$, $P < 0.001$ (Barnes 2015). Fulton's condition factor (K) was calculated, whenever possible: 100,000 (body weight / fork length ^{3}) (Fulton 1902). Sagittal otoliths were processed into thin sections, a technique that has previously been used to age California halibut (MacNair et al. 2001) and is considered a reliable method for ageing longer-lived fishes (Christensen 1964, Power 1978, Beamish and McFarlane 1987). The methodology for thin sectioning was derived from the Committee of Age Reading Experts (CARE 2006) and the formation of one annulus per year was previously validated for California halibut using chemical marking (Pattison and McAllister 1990). California halibut were aged by two or three independent readers until agreement was reached, as described by Barnes 2015.

For both males and females, sex was initially assigned based upon macroscopic characteristics and, if necessary, histology was used to confirm sex. Gonads and livers were removed and weighed (g) for calculations of gonadosomatic index (GSI): 100 (gonad weight/gonad free body weight [g]) and hepatosomatic index (HSI): 100 (liver weight/liver free body weight [g]) (Le Cren 1951, Delahunty and de Vlaming 1980, de Vlaming et al. 1982). Ovaries were then preserved in 10% buffered formalin before transfer to 70% ethanol for storage.

Transverse sections of the preserved ovary were sent to an independent laboratory for histological preparation, where they were dehydrated, embedded in paraffin wax, thin-sectioned, mounted on a microscope slide, stained with hematoxylin and eosin (H&E), and returned for analyses. An initial batch of ovaries (n=18) was processed to determine if there was a difference in the most advanced oocyte stage among anterior, middle, and posterior sections of both blind- and eyed-side lobes. Because preliminary analyses demonstrated no difference by section or lobe, a single sample (i.e., the mid-anterior transverse section of the blind-side lobe) was analyzed for remaining females.

Reproductive phase, spawning state, and maturity assignments.—Prior to preservation, ovaries were macroscopically examined and described according to presence or absence of individual oocytes visible to the naked eye, color, and blood vessel configuration (e.g., color and amount of branching). Each ovary was also histologically examined to identify the most advanced stage of oocyte development. In order of least developed to fully developed, the oocyte developmental stages identified were chromatin nuclear (CN), perinucleolar (PN), cortical alveolar (CA), yolk granule (YG), final maturation (FM), and hydrated (HD), (Murua et al. 2003). CN and PN stages were considered primary growth oocytes and all others were considered secondary growth oocytes (Wallace and Selman 1981). Additionally, histological slides were examined for the presence of postovulatory follicles (i.e., evacuated follicles that collapse when a hydrated oocyte is released [POFs]) and atresia (i.e., resorption of oocytes that are not released). POFs were estimated as new or old and atresia was recorded as alpha atresia (i.e., fresh [*a*AT]) or beta atresia (i.e., old [*b*AT]) based upon the level of degradation (Hunter and Macewicz 1985).

Histological criteria and corresponding macroscopic characteristics were used to assign females to one of six reproductive phases: immature, developing, spawning capable, actively spawning, spent, and resting (Table 1; terminology modified from

Brown-Peterson et al. 2011). Considerations for macroscopically assigning reproductive phases were also outlined. Females histologically assigned to developing, spawning capable, actively spawning, spent, and resting reproductive phases were grouped as mature.

TABLE 1.—Histological characteristics used to describe reproductive phases for female California halibut. Oocyte developmental stages (CN = chromatin nuclear, PN = perinucleolar, CA = cortical alveolar, YG = yolk granule, FM = final maturation, HD = hydrated), other histological characteristics (aAT = alpha atresia, bAT = beta atresia, POFs = postovulatory follicles), and maturity assignments (0 = immature, 1 = mature) are indicated. Corresponding macroscopic characteristics and considerations for assigning reproductive phases based upon macroscopic assessments alone are also described.

Reproductive Phase	Histological Characteristics	Macroscopic Characteristics	Considerations for Macroscopic Assessments
Immature [0]	The most advanced oocytes are in CN or PN stages of development.	Individual oocytes are not visible to the naked eye. Ovaries are light pink to pale orange in color.	Separating this phase from the initial onset of maturity is difficult.
Developing [1]	The most advanced oocytes are in the CA stage.	Individual oocytes are not visible to the naked eye. Ovaries are bright orange in color. Red blood vessels are present.	This phase can be confused with the resting phase.
Spawning Capable [1]	The most advanced oocytes are in the YG or FM developmental stage. Old POFs may be present.	Individual oocytes are visible to the naked eye. Ovaries are yellowish-orange in color.	This phase can be confused with actively spawning and spent phases.
Actively Spawning [1]	HD oocytes and/or new POFs are present. Old POFs may also be present.	HD oocytes may be visible to the naked eye, are interspersed throughout the ovary, and can be accumulated near the oviduct. Ovaries are yellowish-orange in color.	This phase can be confused with spawning capable and spent phases if HD oocytes are not accumulated in the oviduct.
Spent [1]	The most advanced oocytes are in the YG stage. However, greater than 50% of YG stage oocytes are undergoing *a*AT. No POFs are present.	Individual oocytes may be visible to the naked eye. Ovaries are orange, bright orange, or purple in color.	This phase can be confused with spawning capable and actively spawning phases.
Resting [1]	The most advanced oocytes are in CA or PN stages. However, greater than 50% of all CA oocytes (if present) are undergoing *a*AT or *b*AT.	Individual oocytes are not visible to the naked eye. Ovaries are orange to bright orange in color. White (i.e., empty) blood vessels are present.	This phase can be confused with the developing phase.

Histological depictions of female reproductive phases were then used to illustrate the directionality of the California halibut reproductive cycle (Figure 2). In the earliest immature phase of the reproductive cycle, all oocytes can be classified as primary growth oocytes in the chromatin nuclear stage. This is a stage that the ovary never returns to or resembles again. In the next and final phase of immaturity, the most advanced oocytes remain in primary growth, but develop into the perinucleolar stage. The initial progression from the perinucleolar to cortical alveoli stage represents a transition into secondary growth oocytes, progression into the developing reproductive phase, and the initial onset of maturity. Once in the developing phase, the ovary never returns to an immature status. However, the final phase of immaturity can closely resemble the resting phase, which represents an unknown duration of reproductive inactivity that is identified by a return to primary growth stage oocytes. Ovaries were identified as 'early resting' if more than half of secondary growth-size oocytes were undergoing beta atresia, a possible sign of past reproductive activity (Hunter et al. 1992). Relatively large, old females that did not possess any histological signs of past reproductive activity were identified as 'late resting' and mature, even though ovaries resembled the final phase of immaturity. This was due to the assumption that they had previously spawned, based upon their relative size and age. In between the immature and resting phases, females release multiple

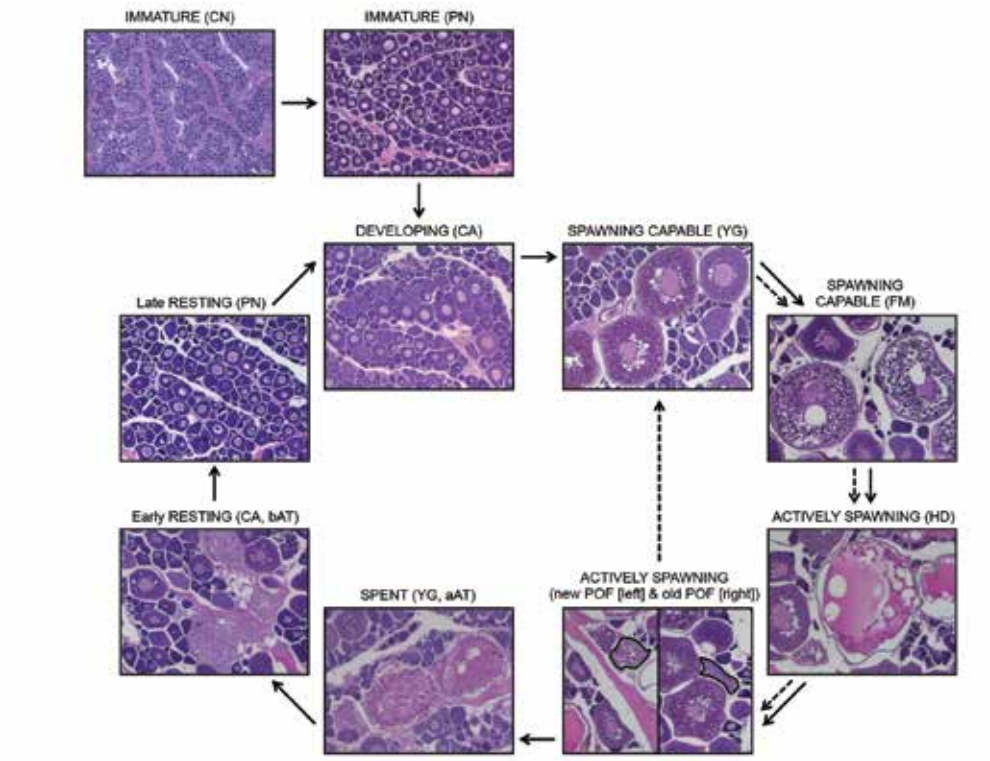

FIGURE 2.—Images of histological slides (50x, ImagePro Plus v7) depicting the most advanced oocyte stage (CN = chromatin nuclear, PN = perinucleolar, CA = cortical alveoli, YG = yolk granule, FM = final maturation, HD = hydrated) and other characteristics (aAT = alpha atresia, bAT = beta atresia, new and old POFs = postovulatory follicles) that were used to determine the reproductive phase for female California halibut. Solid arrows indicate the direction of the reproductive cycle. Dashed arrows indicate that females spawn multiple times by transitioning between spawning capable and actively spawning phases.

batches of eggs through transitions between spawning capable and actively spawning phases, before entering a relatively short spent phase evidenced by mass atresia. The reproductive cycle repeats when the individual leaves the resting phase and enters the developing phase.

Histological analyses were not conducted for male California halibut. Testes were macroscopically examined and described according to lobe shape and incidence of milt in the sperm duct. Males were assigned into one of three reproductive phases: immature, spawning capable, and actively spawning. Immature males had developing (i.e., oval-shaped) testes and no milt in the sperm duct. Spawning capable males had fully developed (i.e., triangular-shaped) testes, but no milt in the sperm duct. Actively spawning males had both fully developed testes and milt in the sperm duct at the time of capture. Males assigned to spawning capable and actively spawning reproductive phases were subsequently grouped as mature.

The incidence of milt in the sperm duct was used to assign an inactive or active spawning state for males. Females were macroscopically categorized into inactive and active spawning states based upon the incidence of individual oocytes visible to the naked eye. A female in an active spawning state was further identified as 'fully hydrated' if hydrated oocytes were accumulated in the oviduct at the time of capture. This macroscopic information was used to construct a guide for assigning spawning states during field-based assessments (Figure 3).

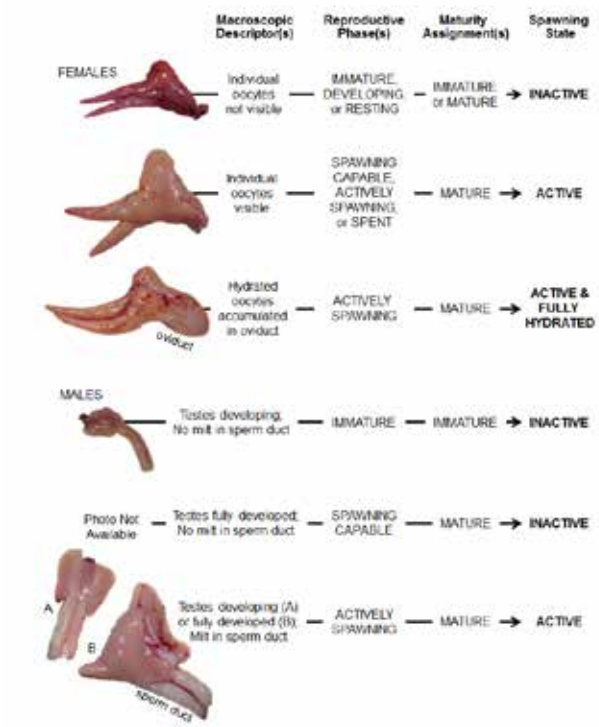

FIGURE 3.—Macroscopic guide to assigning sex-specific spawning states (i.e., inactive or active) to California halibut during field-based assessments. For each spawning state, possible corresponding photo(s), macroscopic descriptor(s), reproductive phase(s), and maturity assignment(s) are listed. The ephemeral 'fully hydrated' condition within the active spawning state for female California halibut confirms an actively spawning reproductive phase at the time of capture. Maturity cannot be macroscopically assigned to inactive females. The presence of milt in the sperm duct confirms an active spawning state at the time of capture for males. A photo was unavailable for mature males collected in an inactive spawning state due to the rare occurrence of this condition.

Maturity curves.—A histologically-based female maturity ogive and a macroscopically-based male maturity ogive were constructed for central California halibut. Data provided by Love and Brooks (1990) were used to represent southern California halibut in constructing maturity ogives for regional comparisons. Additionally, maturity classifications for central California halibut females were reassigned to be consistent with the macroscopic maturity criteria used for southern California females (i.e., based solely upon the incidence of individual oocytes visible to the naked eye). Therefore, developing and resting phase females from the histologically-based central California ogive (i.e., those with individual oocytes not visible to the naked eye) were reclassified as immature for regional comparisons. Methods used to construct male maturity curves for central California did not change because they were categorized macroscopically for both regions. Finally, unpublished data provided by the CDFW Bay Study were used to develop a conversion from total length (TL, cm), to FL for California halibut. The conversion to FL: (0.97 * TL) + 0.60 ($R^2 = 0.999$; $P < 0.01$) was only applied to southern California halibut because FL was initially recorded for all central California halibut.

For all maturity curves described above, relationships between the proportion of mature individuals and length (cm) or age (yr) were established using a generalized linear model (GLM) with a binomial distribution and logit link function (stats, R v3.2.2). Parameters a (slope) and b (intercept) from the equation $P_x = \frac{1}{1+e^{ax+b}}$ (where P_x is the proportion of mature individuals at a given age or length x) were calculated using sex- and region-specific models (Gunderson et al. 1980). Lengths and ages at 50% maturity were calculated from fitted models using the dose.p function and a proportion of mature individuals (p) set to 0.5 (MASS, R v3.2.2).

Reproductive phase, spawning state, and maturity comparisons.—A one-way ANOVA was used to test for differences in age (yr), length (cm), weight (kg), Fulton's K, GSI, and HSI among female reproductive phases, female spawning states, and male maturity assignments. The Tukey HSD post-hoc multiple comparisons test was used to evaluate relationships among significant (P<0.10) factors (SPSS v23). Finally, oocytes from a subsample of whole mount (i.e., preserved eggs) and histologically processed females were measured to evaluate potential differences in oocyte size by developmental stage (Appendix I).

Results

Sample collections.—A total of 635 California halibut (302 females and 333 males) were collected off of central California (Figure 4). The majority of fish collected from San Francisco Bay (92.5%) were shorter than the minimum legal size limit, whereas the majority of fish caught in Monterey Bay (i.e., Santa Cruz, Moss Landing, Monterey) and Morro Bay/Port San Luis (96.7%) were of legal size. As a result, the vast majority of immature fish were collected from San Francisco Bay, whereas mature fish were largely collected along the outer coast of central California. One additional female (82.0 cm; not shown in Figure 4) was collected from Half Moon Bay. Females ranged from 18.6 to 111.0 cm (1 to 19 yr) and males ranged from 19.1 to 95.9 cm (1 to 16 yr). The majority of fish were collected during the summer months (June through August, n=468), followed by fall (September to November, n=107) and spring (March to May, n=59). One fish was collected during winter (December through February).

Reproductive phase, spawning state, and maturity assignments.—Based on histological examinations, female California halibut were classified into six reproductive phases: immature (n=66), developing (n=27), spawning capable (n=109), actively spawning (n=77), spent (n=7), and resting (n=16). Histological assignments of female

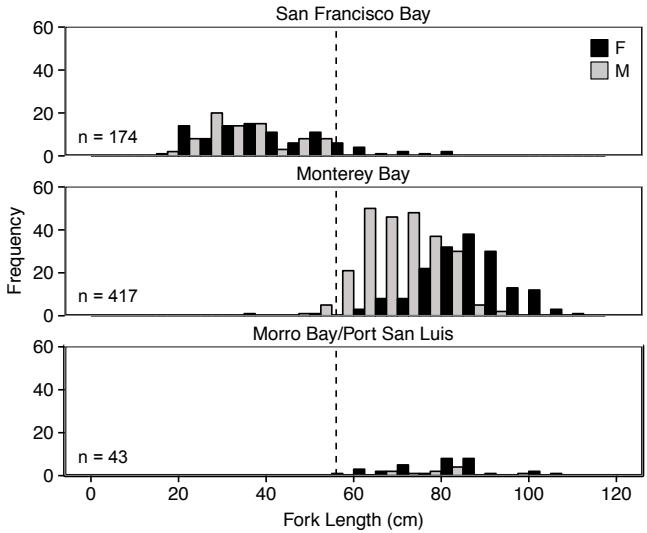

FIGURE 4.—Length frequency distributions for California halibut collected off central California, by sex and location. Black bars denote numbers of females and gray bars denote numbers of males. The dashed line indicates the minimum legal size limit (55.9 cm).

maturity resulted in 66 immature and 236 mature individuals. When reassigning maturity based solely upon macroscopic characteristics, we observed 109 immature females in an inactive spawning state and 193 mature females in an active spawning state.

Macroscopic evaluations of testes yielded three classifications for males: immature (n=19), spawning capable (n=6), and actively spawning (n=305). Malformed males (n=3) possessed testes that were oddly-shaped, more solidly textured, and without milt in the sperm duct. These individuals were excluded from analyses. Of those sampled, 98% of mature males were captured in an actively spawning state. Only a few mature males (2%) were classified as inactive because they possessed fully developed testes, but no milt in the sperm duct.

Maturity curves.— Female California halibut matured at greater lengths (cm) and ages (yr) than male conspecifics in central California, based on histological examination for females and macroscopic examination for males (Figure 5). All females had reached maturity by 51.3 cm (4 yr) and all males had reached maturity by 29.0 cm (3 yr). The fork length (± standard deviation [SD]) at which 50% of the samples collected were considered mature was 47.3±0.88 cm for females and 27.0±0.43 cm for males. The ages (±SD) at which 50% of the samples collected were considered mature was 2.6±0.10 yr for females and 1.1±0.10 yr for males. The smallest mature female was measured at 46.6 cm (2 yr) and the smallest mature male was measured at 25.7 cm (1 yr).

Using histological criteria to determine maturity for central California halibut females resulted in lower estimates of length and age at 50% maturity than those made using macroscopic criteria alone (Table 2). Comparisons of macroscopically-based maturity curves for central and southern California halibut showed that males and females matured at longer lengths off of central California (Figure 6). Females from central California were older at 50% maturity, but age at maturity did not differ for males between the two regions.

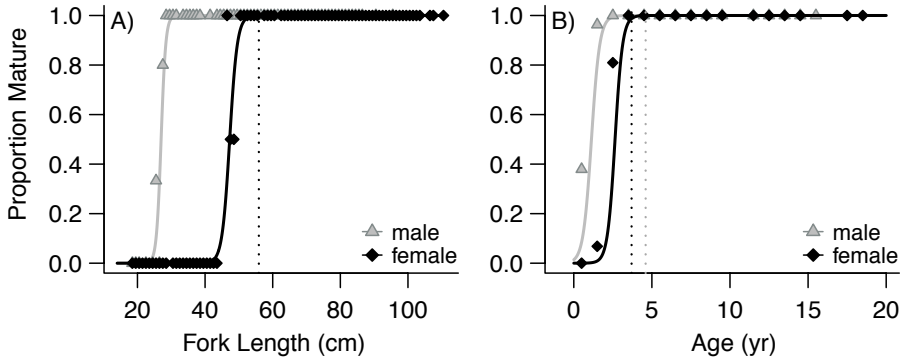

FIGURE 5.—Proportions of mature California halibut collected from central California, by fork length (cm; A) and age (yr; B). Males (classified macroscopically) are shown as gray triangles and females (classified using histological techniques) are shown as black diamonds. Dotted lines represent the single length (as determined by the minimum legal size limit of 55.9 cm) or sex-specific age (estimated by Barnes et al. 2015) at which California halibut become available to the fishery.

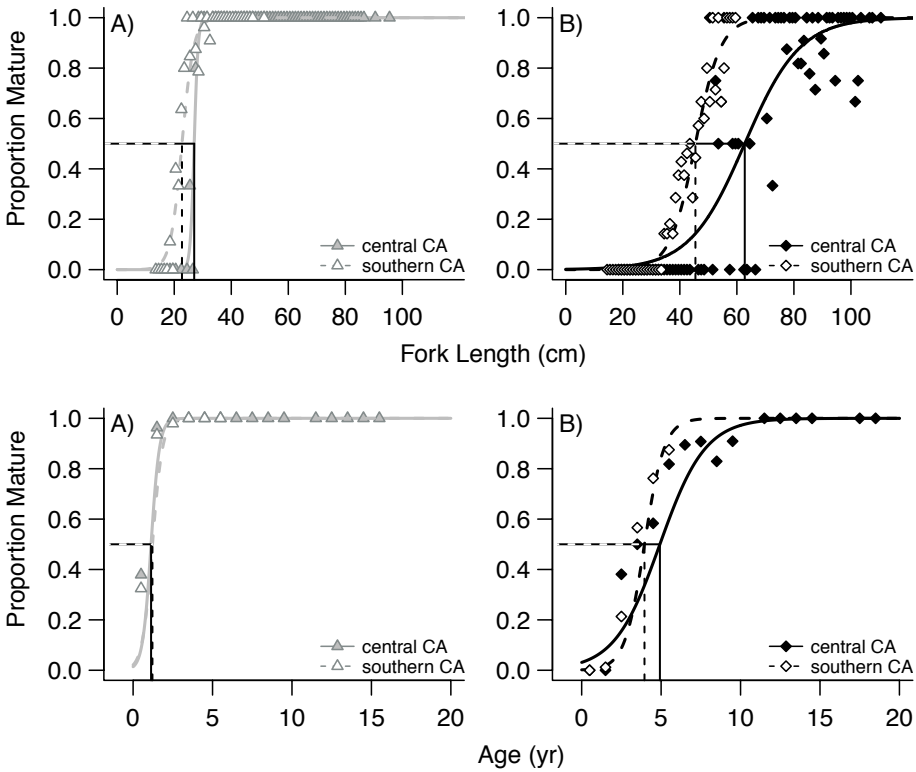

FIGURE 6.—Proportions of mature California halibut collected from central California (this study, shaded shapes and solid curves) and southern California (Love and Brooks 1990, open shapes and dashed curves), by fork length (cm; top) and age (yr; bottom). These curves are based on macroscopic examination only. Males are shown as gray triangles (A) and females are shown as black diamonds (B). Perpendicular lines illustrate region-specific estimates for length and age at 50% maturity.

TABLE 2.—Maximum likelihood estimates (and 95% confidence intervals) for parameters a (slope) and b (intercept) from generalized linear models relating sex- and region-specific proportions of mature California halibut to length (cm) and age (yr). Estimates based upon histological staging criteria developed as part of this study are denoted by asterisks. All other estimates resulted from macroscopic maturity criteria comparable to Love and Brooks (1990). Data for southern California halibut were provided by Love and Brooks (1990). Predicted lengths ($L_{0.50}$) and ages ($A_{0.50}$) at 50% maturity are shown in bold.

	Female		Male	
	central CA	southern CA	central CA	southern CA
Length (cm)				
a	* 0.9 (0.2 to 1.6)			
	0.1 (0.1 to 0.1)	0.2 (-0.1 to 0.3)	1.5 (0.5 to 2.5)	0.5 (0.4 to 0.6)
b	* -42.1 (-74.4 to -8.8)			
	-6.5 (-8.1 to -5.0)	-10.3 (-12.7 to -7.9)	-39.3 (-66.8 to -11.8)	-11.1 (-14.2 to -8.0)
$L_{0.50}$	*** 47.3 (45.6 to 49.0)**			
	62.8 (59.1 to 66.5)	**45.5 (44.8 to 47.2)**	**27.0 (26.2 to 27.8)**	**22.7 (21.8 to 23.6)**
Age (yr)				
a	* 4.1 (2.6 to 5.6)			
	0.7 (0.6 to 0.8)	1.6 (1.2 to 2.0)	3.8 (1.7 to 5.9)	3.1 (2.3 to 3.9)
b	* -10.9 (-14.9 to -6.9)			
	-3.4 (-6.0 to -2.6)	-6.4 (-7.9 to -4.9)	-4.3 (-6.8 to -1.8)	-3.8 (-4.9 to -2.7)
$A_{0.50}$	*** 2.6 (2.4 to 2.8)**			
	4.9 (4.4 to 5.4)	**4.0 (3.75 to 4.25)**	**1.1 (0.9 to 1.3)**	**1.2 (1.1 to 1.3)**

Reproductive phase, spawning state, and maturity comparisons.—Immature females were younger and smaller (both in length and weight $p<0.001$) than all other reproductive phases (Table 3). There were no significant differences in mean Fulton's K among reproductive phases. Estimates of mean GSI for spawning capable and actively spawning

TABLE 3.—Summary statistics for California halibut, categorized by reproductive phase and spawning state for females and maturity assignment for males. Female maturity assignments (0 = immature, 1 = mature) are also indicated for each reproductive phase. Mean, standard deviation, and sample size (parentheses) are indicated for each physical descriptor (age, length, weight, Fulton's K, GSI, and HSI). Different symbols represent statistically significant groups within a particular physical descriptor and reproductive category.

Female Reproductive Phase	Age (yr)	Length (cm)	Weight (kg)	Fulton's K	GSI	HSI
Immature [0]	1.7±0.6 (66) [*]	32.8±8.0 (66) [*]	0.47±0.30 (66) [*]	1.13±0.12 (66)	0.32±0.18 (65)	1.07±0.42 (60) [ч]
Developing [1]	6.2±2.6 (27)	72.0±16.7 (26)	4.95±3.75 (18)	1.15±0.13 (18)	1.09±0.29 (17) [∞]	1.48±0.39 (18)
Spawning Capable [1]	8.3±2.7 (105)	84.7±12.2 (109)	7.73±3.24 (60)	1.17±0.12 (60)	4.05±1.34 (57) [+]	1.68±0.35 (54) [♭]
Actively Spawning [1]	7.8±1.9 (77)	83.0±10.8 (77)	6.92±2.98 (41)	1.18±0.09 (41)	5.21±1.84 (39) [^]	1.71±0.36 (40) [♭]
Spent [1]	7.4±2.1 (7)	79.2±16.1 (7)	6.51±4.27 (5)	1.18±0.03 (5)	1.91±0.53 (5) [∞]	1.65±0.51 (5) [♭]
Resting [1]	4.9±2.2 (16)	66.0±14.4 (16)	3.23±2.45 (12)	1.14±0.09 (12)	1.23±0.24 (11) [∞]	1.12±0.30 (11) [ч]
Female Spawning State						
Inactive	3.3±2.5 (109)	47.2±21.5 (108)	1.65±2.57 (96)	1.13±0.12 (96)	0.57±0.44 (93)	1.16±0.43 (89)
Active	8.0±2.4 (189) [*]	83.9±11.8 (193) [*]	7.36±3.19 (106) [*]	1.17±0.10 (106)	0.57±0.44 (93) [*]	1.16±0.43 (89) [*]
Male Maturity Assignment						
Immature	1.1±0.2 (19)	23.3±3.0 (19)	0.15±0.05 (19)	1.11±0.09 (19)	0.14±0.11 (19)	0.97±0.35 (9)
Mature	6.7±3.0 (232) [*]	63.8±15.3 (311) [*]	3.08±2.23 (167) [*]	1.09±0.12 (129)	2.06±1.45 (127) [*]	1.02±0.36 (122)

females were different from one another and from all other reproductive phases ($p<0.001$). Results from the Tukey HSD post hoc test indicated that spawning capable and actively spawning females represented a relatively similar subset of individuals ($p=0.06$; $n=14.7$), distinct from all other reproductive phases. Mean GSI for females in the spent phase was different from immature ($p=0.03$), spawning capable ($p=0.001$), and actively spawning ($p<0.001$) phases, but not developing ($p= 0.71$) or resting ($p= 0.87$) phases. Statistical comparisons of HSI yielded various results across reproductive phases for female California halibut. However, mean HSI values for immature and resting females were most similar to one another, whereas mean HSI for spawning capable, actively spawning, and spent females were most closely estimated. HSI for developing females was not distinct from either grouping. When aggregating females into spawning states, active females were older, longer, heavier, and exhibited greater values of GSI and HSI ($p<0.01$) than those in an inactive state. Fulton's K was also greater for reproductively active females ($p=0.08$). Mature males were older, longer, heavier, and exhibited greater GSI ($p<0.001$), but there were no differences in mean Fulton's K ($p=0.72$) or HSI ($p=0.88$) from immature males.

Discussion

Male reproductive biology.—Because maturity for male California halibut can be assigned macroscopically, maturity curves may be constructed without the use of histology. In addition, the incidence of milt in the sperm duct can be used to determine the spawning state of male California halibut examined macroscopically in the field. Assigning a spawning state can lead to a better understanding about the proportion of males that spawn in a particular area over a given time period and should be documented.

Female reproductive biology.—Our histological analyses, which illustrated a co-occurrence of secondary growth oocytes (i.e., evidence that the ovary is preparing for another spawning event) and postovulatory follicles (i.e., evidence for recent spawning activity), support the idea that California halibut are batch spawners (Caddell et al. 1990). California halibut also demonstrate asynchronous ovarian development, as evidenced by the fact that the most advanced oocyte stage co-occurred with all preceding stages of oocyte development regardless of reproductive phase (Murua et al. 2003).

Assigning maturity based on histological examination for females.—We found that histological analyses were more accurate in determining maturity for California halibut than depending upon macroscopic characteristics alone. This is because histology reveals characteristics that are not identifiable to the naked eye (i.e., specific oocyte developmental stages, postovulatory follicles, and atresia). However, it remained difficult to histologically discern between immature and resting individuals (Hunter and Macewicz 2003), complicating the assignment of maturity for individuals in these phases.

Because our sampling design provided only a snapshot of reproductive activity, we cannot be certain that fish grouped as mature but collected in a phase other than actively spawning would have spawned within the cycle of capture. We also were unable to ascertain the amount of time it takes an individual found in the developing phase to become capable of spawning. The transition between developing and spawning capable reproductive phases can have substantial effects on estimates of length- or age-at-maturity, given that it may take up to a year to complete (Junquera et al. 2003). However, the time it takes for cortical alveoli oocytes (i.e., developing phase) to develop into yolk granule form (i.e.,

spawning capable phase) remains unknown for California halibut. Additionally, adverse environmental and/or ecological factors may cause a mature individual to forego spawning, undergo atresia, and redirect finite energy reserves toward maintenance (Rideout et al. 2005). There may also be an increased probability of failed reproduction during the first year of maturity (e.g., Jørgensen et al. 2006), which is not known for California halibut.

Assigning spawning state and maturity without histology for females.—Although histological processing is necessary to categorize female California halibut into specific reproductive phases, it is both expensive and time consuming. Therefore, macroscopic examination of the ovaries is highly preferred, especially during field-based assessments. There were some macroscopic similarities in ovary color and the prevalence of blood vessels; however, these characteristics alone were not enough to accurately assign an ovary to a specific reproductive phase. We found that the incidence of macroscopic oocytes [i.e., YG, FM, and HD stages; (Hunter et al. 1992)] was the most straightforward and accurate characteristic to use for field-based assessments of spawning state and maturity. Oocytes in earlier stages of development (i.e., CN, PN, and CA stages) are not individually visible to the naked eye and instead appear as a single mass.

Using this criterion, females can be placed into one of two spawning states: inactive or active. On a finer scale within the active category, a 'fully hydrated' state can be assigned if hydrated oocytes are accumulated in the oviduct and are released when pressure is applied to the organ cavity. This ephemeral state (Hunter and Macewicz 1985) suggests that the female was spawning at the location and time of capture. Although the resolution of these macroscopic assignments would not be as fine as those provided by histological analyses, it can lead to a better understanding about the proportion of females that spawn in a particular area over a given time period and should be documented. We assert, however, that macroscopic assignments should only be used to identify spawning state for females and not to assess maturity. Using macroscopic criteria alone could result in underestimations of proportional maturity, as demonstrated in this study when comparing assignments based on macroscopic and histological characteristics for central California females. If female maturity were to be assessed histologically in southern California, it would be expected that the curves would shift further to the left displaying that females mature at younger ages and smaller sizes than previously reported, provided that there were no temporal changes in maturation between studies. This is because resting females may be mistakenly classified as immature. Developing females, which are assumed to spawn soon after capture, may also be mistakenly categorized as immature individuals. Although assessing maturity macroscopically increases the chance for misclassification, we found that GSI can help guide categorizations of females. Thus, we highly recommend the collection of body and ovary weights, as long as they can be accurately measured.

Management considerations.—Although the first comprehensive stock assessment for California halibut separated the species into two distinct stocks, estimates of maturity were only available from fish collected south of Point Conception (Maunder et al. 2011). Our study is the first to estimate length- and age-at-maturity for the central California halibut stock, thereby providing region-specific data to inform upcoming assessments. We found that the lengths at 50% maturity for central California halibut (27.0 cm [1.1 yr] for males and 47.3 cm [2.6 yr] for histologically-assigned females), were well under the minimum legal size limit of 55.9 cm. This suggests that the minimum legal size limit likely meets the management objective of protecting immature individuals from removal by the fishery north of Point Conception, given current conditions.

When combining results from our maturity ogives with growth rate information from Barnes et al. (2015), we conclude that half of central California halibut males have the opportunity to spawn for 3.5 years before reaching the minimum legal size limit, whereas half of central California halibut females are capable of reproduction 1.1 yr before becoming susceptible to fishery take. For California halibut, reproductive success is related to optimal temperature conditions (i.e., warmer water is associated with better egg and larval survival within tolerance limits [Gadomski and Caddell 1991]) and may be affected by other ecological variables (e.g., prey availability). Varying environmental and ecological conditions can affect the number of successful spawning years available to California halibut prior to becoming susceptible to take by the fishery and should be considered when assessing productivity during different regimes.

Substantial effort and many different gear types (i.e., hook-and-line, spear, beach seine, trawl) were employed in an attempt to collect immature fish along the central California coast. However, the vast majority of immature California halibut were obtained within San Francisco Bay, an area that comprises the greatest proportion of estuarine habitat north of Point Conception. Additionally, California halibut utilize estuaries as nursery habitats (Haaker 1975, Allen and Herbinson 1990, Kramer 1990), and are known to benefit from occupying these environments during early life via increased growth and decreased mortality (Valle et al. 1999). Although the extent of California halibut migration out of this estuary to other portions of the central California coast is unknown, we think it is possible that some of the mature individuals collected along the central California coast were once juveniles inside San Francisco Bay. We do not believe that the sample locations of immature fish substantially affect our length- and age-at-maturity estimates at the regional level, although estimates may differ on a finer spatial scale.

Regional comparisons of length- and age-at-maturity.—There is an energetic tradeoff between growth and reproduction in marine fishes (Jones and Johnston 1977, Rijnsdorp 1990). Differences in growth between central and southern California halibut have been demonstrated, with central California halibut growing at faster rates than southern California conspecifics (MacNair et al. 2001, Barnes et al. 2015), which could be due to relatively early energy allocations toward reproduction over somatic growth and/or maintenance. With consideration that a substantial amount of time had passed between our study and that conducted by Love and Brooks (1990) off of southern California, we found differences in timing of maturation between central and southern California halibut. Our results showed a greater length and age at 50% maturity for central California females. Males from central California also exhibited a slightly greater length at 50% maturity, but there was no regional difference in age. However, similar ages at 50% maturity for central and southern California males could have resulted from a combination of relatively early maturation (between 1 and 3 yr) and rounding age estimates to the nearest year.

There are several potential explanations for the observed differences in length- and age-at-maturity by region. Biogeographic variation in environmental conditions (e.g., temperature, irradiance) and/or ecological interactions (e.g., prey availability, predation rates) have been attributed to intraspecific differences in the life history traits of many flatfish species (e.g., Witthames et al. 1995, Abookire and Macewicz 2003, Spencer 2008, Nissling and Dahlman 2010). Regional variation in fishing pressure can also lead to differences in timing of maturation (e.g., size-selective fishing can cause a shift to maturation at younger ages and/or smaller sizes) and has been documented in exploited fish populations (Rijnsdorp 1989, Bowering and Brodie 1991, Trippel 1995, Grift et al. 2003). Faster rates

of maturation may be problematic to fish populations in the long run because of negative impacts to reproductive potential (e.g., fecundity is known to increase with body size [Bagenal 1966, Wootton 1979]). In comparison with the earliest preliminary study on southern California halibut maturation (Higgins 1919), the data collected by Love and Brooks (1990) suggest that a population-level shift towards earlier maturation may have occurred.

Future studies.—A comparable study using the reproductive phase, spawning state, and maturity assignment criteria described herein should be conducted to re-assess sex-specific length- and age-at-maturity for southern California. Histological analyses would provide more precise estimates of female maturity for construction and comparison of maturity ogives. Relying on macroscopic characteristics alone is an inaccurate way to assess maturity because it only accounts for individuals in a reproductively active state. Because environmental conditions (e.g., PDO, [Chavez et al. 2003]) and relative fishing pressure (Maunder et al. 2011) have changed over the past three decades, it is also necessary to formulate contemporary estimates of length- and age-at-maturity (Rijnsdorp 1989). A study conducted in southern California that is similar to ours would enhance regional comparisons of length- and age-at-maturity, increase accuracy in evaluating the effectiveness of the minimum legal size limit south of Point Conception, and provide an opportunity to compare temporal variation in maturation of southern California halibut.

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Appendix I: Oocyte Measurements

Oocyte circumference (μm) and area (μm²) were measured from two types of ovarian tissue: preserved (i.e., whole mounts fixed in formalin and stored in ethanol; Table A) and histologically processed (Table B). Because California halibut oocytes are not perfectly spherical, estimates of diameter (μm) were calculated by dividing circumference (μm) by pi (π).

Table A.—Whole mount measurements (8x magnification, ImagePro Plus v7) obtained from actively spawning California halibut females (83.6±29.0 cm TL; n=5). Mean, standard deviation, and number of individual oocytes measured (parentheses) are listed by aggregated oocyte developmental stage. Because developmental stages are difficult to differentiate from whole mounts, oocytes in PN and CA stages were aggregated, as were those in YG and FM stages.

Developmental Stage	Diameter (μm)	Circumference (μm)	Area (μm^2)
perinucleolar (PN) or cortical alveoli (CA)	271±43 (51)	851±136 (51)	52,390±16,101 (51)
yolk granule (YG) or final maturation (FM)	491±73 (59)	1,543±229 (59)	169,971±46,923 (59)
hydrated (HD)	902±71 (68)	2,834±222 (68)	564,366±73,911 (68)

Table B.—Histological measurements (50x magnification, ImagePro Plus v7) obtained from spawning capable and actively spawning California halibut females (82.7±12.9 cm TL; n=12). Mean, standard deviation, and number of individual oocytes measured (parentheses) are listed by oocyte developmental stage. Damage caused by histological processing prevented the measurement of HD stage oocytes from histological samples.

Developmental Stage	Diameter (μm)	Circumference (μm)	Area (μm^2)
perinucleolar (PN)	89±20 (271)	279±61 (271)	5,056±2,217 (271)
cortical alveoli (CA)	200±57 (167)	628±180 (167)	26,245±16,025 (167)
yolk granule (YG)	365±62 (85)	1,147±196 (85)	80,661±29,418 (85)
final maturation (FM)	489±80 (42)	1,537±252 (42)	149,642±39,018 (42)

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Sexual development and symbionts of native Olympia oysters *Ostrea lurida* naturally settled on cultch deployed in San Francisco Bay, California

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Attempts to restore depleted oyster populations are taking place worldwide. The design of effective restoration programs can benefit significantly from knowledge of basic biological processes such as ontogenetic and seasonal reproductive patterns and the presence of potential agents of disease. In June 2007 we deployed oyster shell cultch in a series of mounds in San Francisco Bay, California, USA. In monthly sampling, sixty of the largest oysters recruited onto the cultch were examined histologically to track the reproductive development of the initial settlers. Symbiont presence was also recorded. Sexual development was already occurring in August 2007, 56 days after cultch deployment, with immature males comprising 18% of the sample. Mature sperm and oocytes were observed in September 2007, 92 days following cultch deployment. Brooded larvae were observed in October-November 2007 and April-June 2008, indicating a relatively long reproductive season and confirming that oysters that settle in late spring can reproduce as females by autumn. These results suggest the capacity for rapid population expansion when suitable habitat is available. The oysters were minimally affected by disease, in contrast to the native oyster (*Crassostrea virginica)* of the US Atlantic and Gulf coasts.

Key Words: *Ostrea lurida*, restoration, cultch, reproduction, disease

The native Olympia oyster (*Ostrea lurida)* was historically important in the ecology of intertidal estuarine communities along much of the west coast of North America. For thousands of years it was an important food source for humans (Baker 1995). The mid-nineteenth century California Gold Rush spurred intensive harvest of oysters in San Francisco Bay and subsequently in numerous other embayments in California, Oregon, and Washington (Baker 1995, Kirby 2004). Since the shells of their predecessors provided solid substrate that is often limited in such bays, the harvest of oysters resulted in a significant reduction of habitat. In San Francisco Bay, this harvest, along with siltation from hydraulic mining in the Sierra Nevada mountain range, the filling of tidal flatlands, and increasing numbers of competing non-native species, drastically reduced traditional oyster habitat while seawalls and armored shorelines provide potential new habitat. Despite no significant harvest for more than 100 years, oyster populations have failed to recover (Kirby 2004). The decline of *O. lurida* is far from unique, since oyster populations and oyster reefs have diminished or disappeared in many regions of the world (Kirby 2004, Beck et al. 2011, zu Ermgassen et al. 2012).

Over the past decade, interest in restoring native oyster populations has increased throughout the United States (McGraw 2009, Trimble et al. 2009, White et al. 2009, Beck et al. 2011, Kennedy et al. 2011, State Coastal Conservancy 2010, Wasson 2010, Wasson et al. 2015). Relative to the native eastern oyster (*Crassostrea virginica)*, the Olympia oyster is vastly understudied and active recovery efforts have just recently begun. Recruitment dynamics, habitat requirements, genetics, reproduction, disease, and the utility of deploying artificial reef structures are recognized as important information sets for implementing effective restoration strategies (McGraw 2009, State Coastal Conservancy 2010, Wasson 2010,Wasson et al. 2015). A number of recent studies examined *O. lurida* populations in Washington (Trimble et al. 2009, White et al. 2009), Oregon (Groth and Rumrill 2009, Pritchard et al. 2015), and Southern California (Polson and Zacherl 2009, Seale and Zacherl 2009). Populations are diminished from historical levels at all locations, and both reduced recruitment and limited habitat are commonly cited as key impediments to recovery.

Critical factors in recruitment success include reproductive output and larval survival. Members of the genus *Ostrea* employ a reproductive strategy that includes protandric hermaphroditism and larval brooding before release as veligers. The reproductive biology of *O. lurida* was first reported by Stafford (1913) in British Columbia, Canada, who observed that sperm develops in aggregates (later commonly referred to as sperm balls, morulae, or spermatozeugmata), larvae are brooded before release, and that "each individual is bisexual, hermaphroditic, monoecious." He also noted that younger individuals had sperm but no ova, i.e., protandry. The continuous cycling between male and female sexes was later described as rhythmical consecutive hermaphroditism (Mackie 1984). In contrast, members of the genus *Crassostrea* have monoecious gonads displaying alternative hermaphroditism in which adults develop a single sex that may or may not change during the subsequent season, with protandry being typical, e.g., younger animals tend to be male and older tend to be female (Mackie 1984).

Wesley Coe produced a series of reports (1930, 1931a, 1931b, 1932a, 1932b, 1934) after studying *O. lurida* of known approximate age that settled onto wooden or concrete blocks submerged for various lengths of time off a pier in La Jolla, California, during 1926-1931. The pier is located on the open coast, an unusual environment to encounter settling *O. lurida*. Coe stated that the initial male stage is followed by a female phase, another male phase, and a period of recuperation, but this cycle is suspended when temperatures

fall below 16 °C in fall and resumes the following spring when temperatures again reach 16 °C. The dramatic effects of elevated temperature on the rate of reproductive maturation in *O. lurida* were briefly reported by Santos et al. (1993). Adult oysters, presumably from a wild population in Washington State, were collected in January when water temperatures were 8 °C and the oysters were rapidly acclimated to 12, 18, or 21 °C. Those held at 21 °C produced large numbers of larvae after 2-3 weeks, followed by those at 18 °C after 3-4 weeks, while for the population at 12 °C a small number of brooders were noted at 8-9 weeks, at which time the experiment was terminated. More recently, Oates (2013) examined reproductive patterns in large (>30 mm shell length) *O. lurida* from two locations in Coos Bay, Oregon, conducting histology on 30 animals monthly from January to December 2012. Gonads were categorized as female, male, or hermaphroditic and predominantly female, predominantly male, or with equal representation of both sexes. Animals were also assigned a gonad maturity stage and oocyte diameter was measured. Gametogenesis was observed from May to September with brooded larvae seen from July to September. Differences in timing between the two sites was attributed to salinity stress (<15 ppt) at the site farther from the mouth of the bay. To our knowledge no other research on Olympia oyster reproduction has been published during the past seventy years.

By periodically examining a single set of the European flat oyster (*Ostrea edulis)* in England, Cole (1942) provided a detailed description of sequential sexual development in that species over time. His descriptions are quite similar to those of Coe in California, but with clearly defined animal ages and more detailed information on the sizes and numbers of oysters examined. Cole categorized the sequential stages of development summarized herein. In the indifferent or undeveloped stage the distal wall of each gonoduct is lined with ciliated epithelium and the wall closest to the digestive gland was lined with gonadal precursors, which even at this earliest stage could be identified as spermatogonia and oogonia. This is followed by a young male stage in which spermatogenesis commences, immature sperm balls develop, and oocytes along the follicle wall begin to expand, followed by a first male stage containing ripe and maturing sperm balls while oocytes along the follicle wall expand in size and number. The next stage is a male-to-female transition in which mature sperm balls are still present in follicles and ducts while developing oocytes completely line the follicle walls. The subsequent first female stage is characterized by follicles filled with mature oocytes interspersed with residual spermatocytes and occasional oogonia and spermatogonia lining follicle walls. Even at this stage residual mature sperm balls may still be present. Release of mature oocytes is rapidly followed by the second male stage, with developing sperm balls expanding in follicles until mature sperm balls again fill the cavity. Cole believed that such cycling continued regularly, with one female and one male phase being completed on an annual basis in Britain.

We conducted this study primarily to examine the timing and patterns of reproductive development of native Olympia oysters that settled onto planted cultch material in San Francisco Bay, California, USA. To track the initial settlers we examined the largest individuals present in monthly samples. After preliminary studies on California *O. lurida* and reviewing literature on oyster gonad categorization and maturation sequence, we concluded that the terminology and sequence used by Cole (1942) for British *O. edulis* provided excellent agreement with our species, although the timing was expected to be different based on water temperature and perhaps a variety of other factors. To the methods of Cole we added examination of maximum oocyte diameter and noted the presence of brooded larvae when apparent in histological preparations.

Disease has been shown to be one of the most important factors regulating animal densities in various oyster populations worldwide. Developing baseline knowledge of pathogen presence and distribution is essential toward gaining an understanding of their potential impacts on restored populations. Therefore in this study we recorded the presence of all symbionts, including potential pathogens.

Materials and Methods

Project location and description.—Crassostrea gigas left and right valves, dried at least two years, were deployed as cultch on tidelands in San Francisco Bay at the Marin Rod and Gun Club in San Rafael, California, USA (Figure 1) on 9 June 2007. Plastic mesh (2.5 cm) bags, approximately 70 cm in length and 25 cm in diameter, were filled with approximately 81 valves. Thirty bags were used to create pyramid-shaped mounds around a PVC pipe inserted into the mud substrate at approximately -0.6 m mean lower low water (MLLW) tidal height. We constructed 26 mounds in four rows (either six or seven mounds per row). Mounds within each row were 3.0 m apart with 3.0 m spacing between rows. About 15 of the lowest bags in each mound rapidly settled into the soft substrate, leaving about 15 upper bags available for colonization. Temperature and salinity data were obtained from instrumentation at the Romberg Tiburon Center near Tiburon, California, approximately 6.7 km south-southeast of the project site (Figure 1).

Figure 1.— Project location in San Francisco Bay, California, USA. Star shows cultch outplant location on tidelands at the Marin Rod and Gun Club, San Rafael. Circle indicates the Romberg Tiburon Center, 6.7 km from the project location, where temperature and salinity data were recorded.

Sample collecting and processing.—We collected samples of the deployed cultch on an approximately monthly basis for twelve months (Table 1). For each monthly sample we removed one randomly selected bag of exposed cultch from each of two randomly selected cultch bag mounds. After combining the cultch from both bags we selected 60 individuals in the uppermost size range for processing. This subset was selected in order to follow the individuals that settled soon after cultch was deployed, avoiding more newly-settled individuals. We measured the shell height and shucked each individual. Those with shell heights less than 2 cm were placed whole in a histological cassette. For larger individuals, a single cross-section was taken that contained digestive gland, gonad, gill, kidney, and heart. To reduce costs we placed multiple animals into one cassette when cross-sections were sufficiently small, with up to four animals per cassette. One exception was the first sample in July 2007 that consisted of only 25 very small animals; all were placed whole into one cassette. Cassettes were placed in Davidson's fixative (Shaw and Battle, 1957) for 24 hours followed by the routine production of 5 µm, hematoxylin- and eosin-stained tissue sections that were examined under a microscope.

Histological examination.—Using the classification method of Cole (1942) with minor modifications, we categorized the gonad for the first nine months (July 2007-March 2008) as Indeterminate (I) (in place of Cole's term 'indifferent'), Juvenile Male (JM) (in place of Cole's term 'Young Male'), First Male Stage (M1), First Male Stage to First Female Stage Transition (M1F1), First Female Stage (F1), or Second Male Stage (M2). Characteristics of each stage are described in the Results section. Slides from each sample were read in monthly order with knowledge of the sample date. One departure from the category descriptions provided by Cole (1942) was that we categorized some males as Juvenile Males despite the appearance of a very small number (one to several) of mature-appearing sperm balls in minimally developed gonad. For the monthly samples beyond March (April-June 2008) we found it impossible to confidently assign individuals into these and subsequent stages, and thus gonad stage data was only assessed through March 2008. For all individuals that contained female gonad we used an ocular micrometer to measure a maximum oocyte diameter after identifying the largest spherically-dimensioned oocyte in each section. We also recorded the presence of brooded larvae as well as potential pathogens and all other symbionts.

TABLE 1.—Sampling schedule showing date and number of animals processed for histological examination.

Date	Days After Deployment	Number of Animals
7/8/2007	29	25
8/4/2007	56	62
9/9/2007	92	59
10/9/2007	122	60
11/5/2007	149	61
12/17/2007	191	60
1/14/2008	219	60
2/11/2008	247	60
3/12/2008	277	60
4/17/2008	313	60
5/12/2008	338	60
6/11/2008	368	60

Results

Site description and environmental data.—Oyster shell cultch was deployed on mud-bottom private tidelands in northern San Francisco Bay owned by the Marin Rod and Gun Club (Figure 1). Native oysters are common but distributed in patches on low intertidal riprap throughout the region. Water temperatures and salinities recorded during the study period from sensors located 6.7 km from the deployment site are shown in Figure 2. The patterns are typical of this portion of San Francisco Bay, with nearly-oceanic salinity for most of the year, except during heavy winter and spring rains, and a low temperature in late winter rising to a peak in late summer to fall.

Reproductive development.—To investigate the rate at which the first settlers on the cultch became sexually mature, we histologically examined 60 oysters from the uppermost size range in each monthly sample. Based on our unpublished observations, from the second

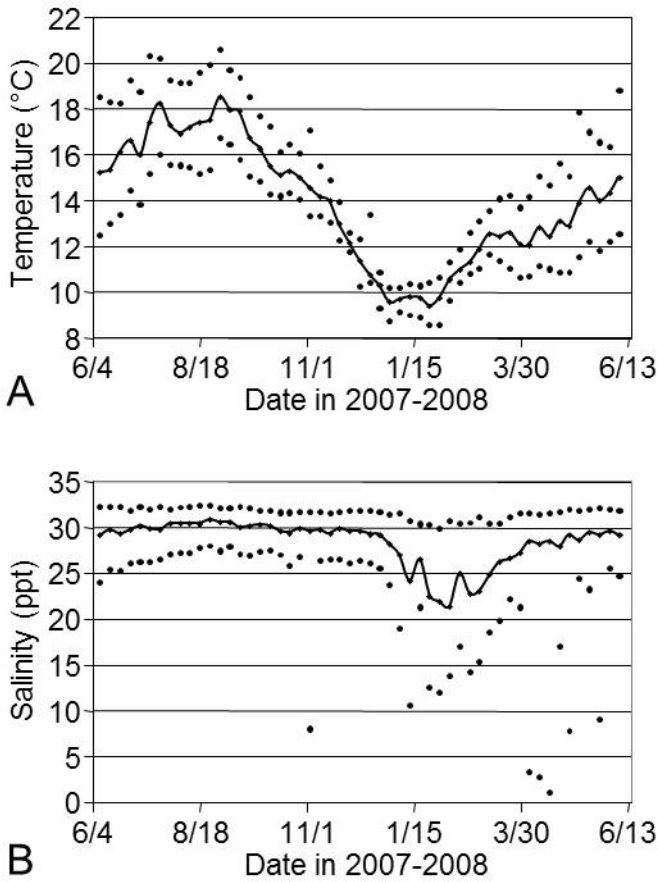

Figure 2.—Temperature (A) and salinity (B) in San Francisco Bay, California, USA over the study period, recorded 6.7 km from the project location. Line show weekly means and upper and lower dots show weekly maximum and minimum recordings respectively.

sampling onward, a total of 908 to 2,744 oysters were present on the cultch examined; thus our sample size of 60 resulted in subsets in the uppermost 2-7% of the size range present and should consist of the larger, fast-growing animals among those that settled soon after cultch deployment. The oysters grew steadily throughout the summer, then at a slower but fairly consistent rate from fall through late spring (Figure 3). Characteristics of the sequential stages of reproductive development are shown below.

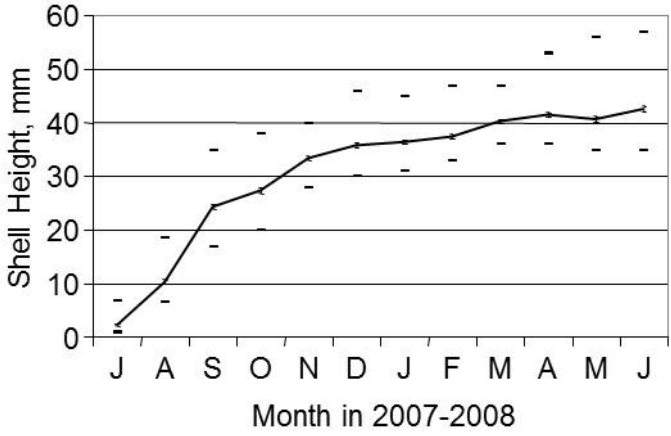

FIGURE 3.—Mean shell height (± standard error, barely visible) of Olympia oysters in each monthly sample. Dashes show minimum and maximum values in each sample.

Indeterminate (I). No gonad follicles present or follicles contain ciliated epithelium and/or gonad precursors of indeterminate sex (Figure 4A).

Juvenile Male (JM). Initial follicle is a mostly empty, arced, thin duct parallel to the anterior/posterior axis of the animal, with spermatogonia along walls and developing sperm balls in lumen. Oogonia appear along the perimeters. Ducts may expand into the body at right angles to the primary ducts as the number of sperm balls present increases. One to several mature sperm balls per follicle may be present but nearly all are immature (Figure 4B).

First Male Stage (M1). In earliest stages, a single layer of developing oogonia line follicle perimeters, followed by layers of developing sperm balls that surround mature sperm balls in the centers of follicle lumina. In later stages the proportion of mature sperm balls increases and they fill the follicle lumina, while the female gonad continues to develop along perimeters. In latest stages multiple layers of fully mature sperm balls are present in lumina and crowding gonoducts (Figure 4C).

First Male to First Female Transition (M1F1). Some mature and developing sperm balls are still present in follicle lumina and gonoducts. Greatly expanded oocytes line the perimeters one or more layers deep. In latest stages, the gonad is dominated by female tissue but mature sperm balls are still common (Figure 4D).

First Female Stage (F1). Multiple layers of developing or fully mature oocytes are present in follicle lumina, increasing in number and size as the stage progresses (Figure 4E-F). Residual spermatogonia, residual spermatocytes and mature or degraded sperm balls may be present in lumina and along duct perimeters. Early brooded larvae may be present in the mantle.

Second Male Stage (M2). In early examples, sperm ball precursors rapidly divide in follicle lumina while residual oocytes of variable size remain attached to walls of follicles that may be partially empty due to release of oocytes (Figure 4G-H). Degraded oocyte material and phagocytes are usually present and brooded larvae may also be present. Later stage follicles are dominated by mature sperm balls. Gonad volume is many times larger than that of the M1 stage.

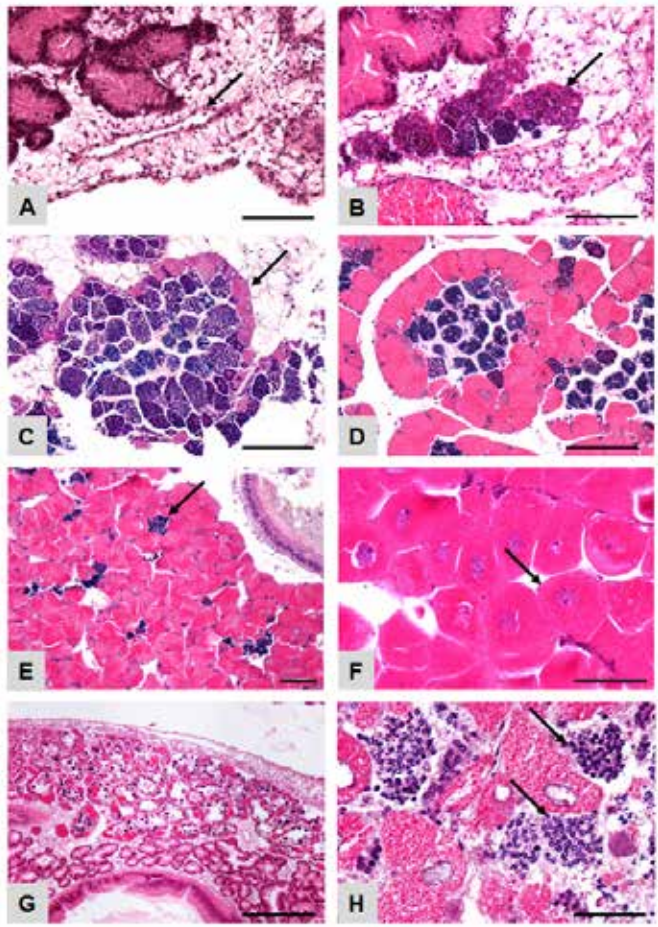

FIGURE 4.—Histology of Olympia oyster gonad development. A: Indeterminate (I) oyster from August 2007 sampling. Arrow points to undeveloped gonad duct. B: Juvenile male (JM), August 2007 sample. Small follicle (arrow) contains a few maturing sperm balls surrounded by loosely-organized developing female gonad. C: Male stage 1 (M1), September 2007 sample. Follicle dominated by mature and developing sperm balls with well-organized female gonad on the periphery (arrow). D: Male to female transition (M1F1), September 2007. Mature sperm balls present but most of the follicle volume contains maturing oocytes lining all or nearly all of the follicle walls. E: Mature female gonad (F1), September 2007 sample, with scattered pockets of residual spermatocytes (arrow). F: High magnification of female gonad, September 2007, showing uniformly fully mature oocytes (arrow). G: Early example of Male stage 2 (M2), November 2007 sample. The animal was brooding abundant larvae. H: Male stage 2, February 2008 sample. Rapidly-developing sperm balls (arrows) are replacing degraded oocytes. Scale bars are 100 µm in A, C, D, E, and F, 50 µm in B and H, 500 µm in G.

The emergence and presence of sequential stages of initial sexual development are shown in Figure 5. In August 2007, 56 days after the cultch deployment, 18% of the oysters were developing as juvenile males (JM) (Figure 5). By September 2007, 92 days after cultch deployment, all showed reproductive development, and oysters both with mature sperm and with mature oocytes were present, with 12% of the oysters in the F1 stage. In October the proportion in F1 increased to 22% and those in M2 appeared, comprising 10% of the oysters sampled. Throughout winter to early spring (November 2007-March 2008) the proportion of individuals in the F1 and M2 stages increased in preparation for spring spawning. In samples from beyond March 2008, it was not possible to confidently distinguish the M2 stage from potential M3 stages and the F1 stage from potential F2 stages with further complexity being present each month. However we did measure maximum oocyte diameter and noted the presence of brooded larvae and symbionts in the April to June 2008 samples.

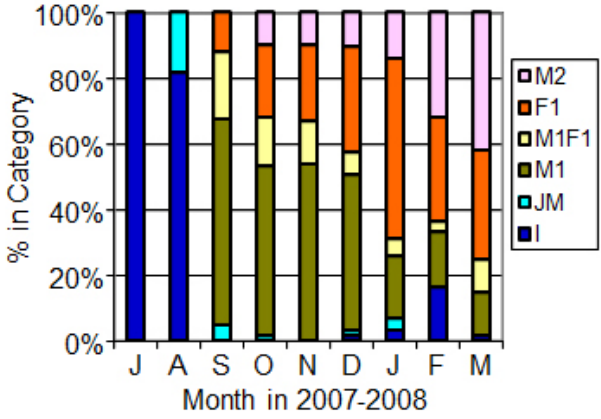

FIGURE 5.—Monthly categorization of sexual stages of Olympia oysters recruited onto cultch following outplant in June, 2007. Sexual stages are described in the Results section.

The maximum oocyte diameter in the oysters containing female gonad rose steadily from August through September 2007 and then remained fairly stable (Figure 6). These data agree with the first histological appearance of mature oocytes in September 2007.

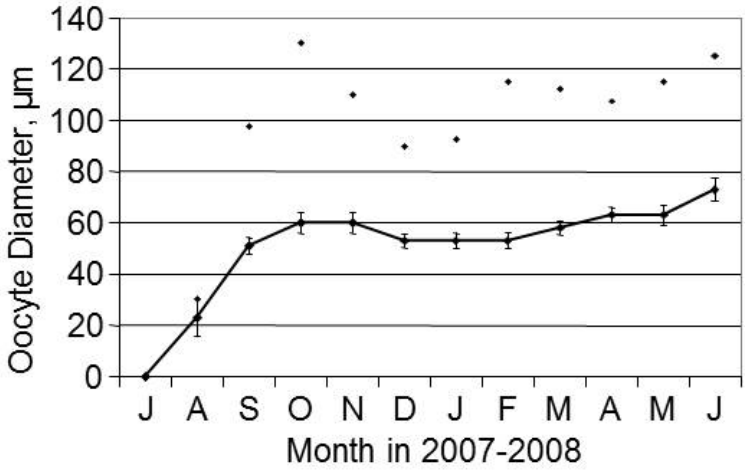

FIGURE 6.—Mean (± standard error) of the maximum diameter of oocytes in Olympia oysters that had female gonad tissue in each monthly sample. Dots indicate size of the largest oocyte measured in each sample.

Most larvae that were being brooded by oysters sampled in this study were probably lost during histological processing, yet brooded larvae were observed at time points when they would be expected to be present based on patterns of gonad status. Brooded larvae were first seen in five individuals in October 2007, following the first observations of mature sperm, mature oocytes, and post-spawn females in September 2007. We observed six brooders in November 2007, none from December 2007 to March 2008, and between two and four were present from April through June 2008. Brooded larvae were found in association with mantle and gill tissues (Figure 7A-C). In order to examine the temperature at which oocyte release occurs, we examined mean temperatures during the five days preceding sampling events for the five monthly samples that included individuals with brooded larvae (October and November 2007; April, May and June 2008). The mean temperatures ranged from 13.1 °C to 15.8 °C.

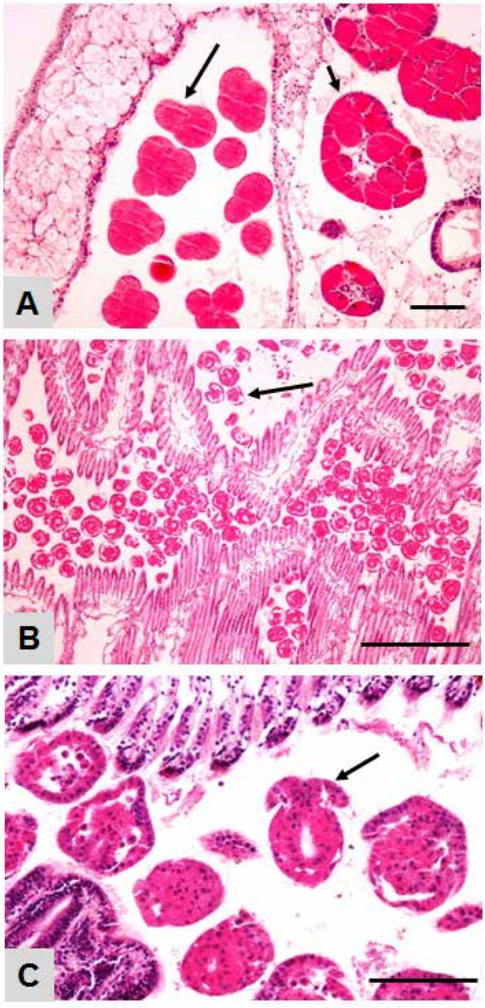

FIGURE 7.—Histology of Olympia oyster brooded larvae. A: Short arrow points to mature oocytes in female gonad and long arrow points to brooded 2-4 cell stages in a fold of the mantle, stage 1 female (F1), November 2007. B: Large numbers of early veliger larvae (arrow) brooded in gill tissue by a stage 2 male (M2), October 2007. C: Higher magnification of the larvae shown in (B). Arrow points to developing larva. Scale bars are 100 µm in A and C, and 500 µm in B.

Symbionts.—Pathological conditions and potential disease agents were rare throughout the study. The most commonly observed symbionts were *Echeneibothrium*-like cestode larvae in the gastrointestinal tract of 12 of the oysters (1.7% overall prevalence, Figure 8A). We first observed them in August 2007. In some instances there were associated localized tissue trauma and host responses at attachment sites to the gut epithelium. Two oysters (0.3% overall prevalence, one in each of the May and June 2008 samples) had a single *Urastoma*-like turbellarian flatworm present in association with gill tissue but no harm to the host was observed (Figure 8B). One oyster in the February 2008 sample had a *Mytilicola*-like copepod in the gastrointestinal tract (Figure 8C). Copepod egg masses were observed in gill tissue of another oyster in the same sample. One individual in the June 2008 sample was infected by a microcell protozoan with characteristics of *Bonamia* sp. (Figure 8D). The parasite was approximately 2.4 µm in diameter with a centrally located, 1.1 µm diameter nucleus. It was present in the cytoplasm of hemocytes and occasionally free in the hemolymph. Infected hemocytes were focally abundant and associated with widespread hemocyte recruitment.

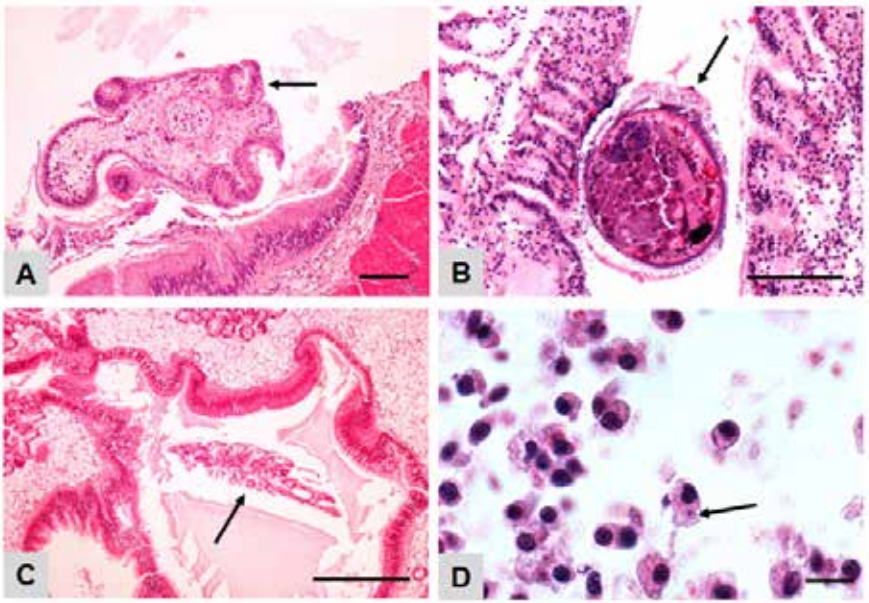

FIGURE 8.—Histology of Olympia oyster symbionts. A: Cestode larva (arrow) in gastrointestinal tract, May 2008. B: Turbellarian flatworm (arrow) between gill filaments, June 2008. C: *Mytilicola*-like copepod in gastrointestinal tract, November 2007. D: Presumptive *Bonamia* sp. microcell in a hemocyte, June 2008. Scale bars are 100 µm in A and B, 500 µm in C and 10 µm in D.

DISCUSSION

The deployment setting.—San Francisco Bay is the largest estuary on the west coast of North America. Human-mediated changes in water flow, filling of wetlands, and non-native species introductions have resulted in drastic alterations in habitat, environmental chemistry, and food webs (Cloern and Jassby 2012). Environmental conditions at our study site during the yearlong study were typical for northern San Francisco Bay, with winter

temperature lows coinciding with periodic, greatly decreased salinity during heavy winter rains. The native oyster *O. lurida* does well under these conditions except when salinities remain low for extended periods, which can result in dramatic mortality events (State Coastal Conservancy 2010, Cheng et al. 2015, Wasson et al. 2015). Our study location, like much of San Francisco Bay, has a fine sediment substrate, and the lower bags in each mound we created settled quickly into the substrate, while the higher bags remained stable. Deployment of the cultch at -0.6 m MLLW resulted in a low intertidal to subtidal setting, somewhat deeper than most natural populations of *O. lurida* in San Francisco Bay. This tidal height was chosen based on our unpublished observations of insignificant difference in recruitment between -0.8 m and -0.3 m MLLW, to minimize competition with non-native mussels *(Mytilus galloprovincialis)* that tend to be more abundant in deeper water, and for ease of access. The timing of our cultch deployment (June) was chosen to minimize colonization of non-oyster sessile invertebrates, particularly barnacles (*Balanus* sp.) that settle earlier in spring.

Onset of reproduction.—Coe (1930, 1931a, 1931b, 1932a, 1932b) described sexual development of Olympia oysters of known approximate age. He deployed wood or cement blocks off the Scripps Institution of Oceanography pier in La Jolla, California. Although important studies, his reports are somewhat unclear regarding the number of oysters examined, block type, block depth, and histological methodology. The central findings reported by Coe were that the oysters are hermaphrodites that first release male gametes (protandry) and then cycle between female and male stages, with developmental stasis at temperatures below 16 °C. Oysters that settled in spring could release male gametes as early as five months of age and release female gametes as early as one month later. Our data generally agree with this, with several exceptions. In our study some of the oysters that settled in early June had released gametes by early September (three months) and brooding hermaphrodites were present by early October (four months). Reasons for the discrepancy between our study and Coe's may include our size selection of the animals studied, although Coe also selected for larger animals (Coe 1932b) and/or differences in geographic setting, habitat, genetics, and sample sizes employed.

Cole (1942) provided a very thorough histological description of sexual development in *O. edulis*. Cole's review of previous studies and his own work collectively demonstrated *O. edulis* that settle in spring are capable of sperm production by fall of the same year, and that timing of reproductive stages is heavily influenced by temperature and latitude. He described a reproductive development pattern similar to that reported by Coe for *O. lurida*, i.e., an initial release of male gametes followed by release of female gametes and subsequent cycling. Studying *O. edulis* raised in Spain, da Silva et al. (2009) reported that among cohorts spawned in March to May 2001, all were indeterminate through October of the first year with a very small proportion of males by November. Males and then hermaphrodites and females grew in proportion through the following spring and summer, then over half of the animals became indeterminate again in November before the cycle repeated. This is a much slower and more synchronous sequence of development than reported for *O. lurida* despite reported temperatures (9-18.5 °C) similar to those in central California and Puget Sound, Washington. Millar (1964) and Wilson and Simons (1985) reported similar population synchrony in seasonal cycling of maximum oocyte diameter in *O. edulis* from Scotland and Ireland, respectively. We observed less seasonal synchrony, e.g. maximum oocyte diameters were relatively constant once the first females reached maturity (Figure 5).

Even though some oysters progressed to be producing and releasing oocytes during the first fall, we found that more than half remained in the first male stage through the end of the year (Figure 5). Thus following the initial male phase, significant proportions of the population consists of animals in either mature male or mature female stages at any particular point in time. This lack of synchrony suggests that founder populations would be capable of becoming self-sustaining more quickly than they would if gametogenesis showed a high degree of synchrony. For example, fertilization of newly spawned oocytes could be limited if nearly all of the oysters were simultaneously in the first female phase.

Reproductive categories and terminology.—Many studies on ostreid reproduction categorize each gonad with respect to the proportion of female and male tissue and stage of maturity. Although most of this research includes reasonably detailed definitions of sexes and stages, those definitions differ between studies. Thus Orton (1927), who studied gonad smears, considered *O. edulis* true hermaphrodites to be only those animals containing ripe sperm and mature eggs distributed evenly throughout the gonad, and had seven additional categories for female and male gonads containing various proportions and stages of each type. Most subsequent literature uses the term hermaphrodite to include any animal having female and male gonadal elements. Orton (1927) and later studies that utilized histology vary widely in the use of categories pure female/pure male or solely female/solely male. Orton (1927) considered pure females and pure males to be those individuals with gonads containing only entirely ripe ova and ripe or ripening sperm, respectively. Coe (1932b) did not use such terminology but noted that older *O. lurida* may tend to have one or the other sex dominate the gonad. Cole (1942) described *O. edulis* pure males as having only oogonia (the earliest identifiable female stage) with no later female stages present. Loosanoff (1962) described just three categories of *O. edulis* gonad, all hermaphroditic: ambisexual (with equal female and male representation) and predominantly female or male. Mann (1979) reported the presence of "totally male and totally female individuals" in laboratory-reared *O. edulis* without further description. Siddiqui and Ahmed (2002), studying two populations of *O. nomades* in Pakistan, included unisexual male and female categories in which all follicles contained only male or female tissue, and described the species as 'mostly hermaphroditic'. In his study of *O. lurida* in Coos Bay, Oregon, Oates (2013) included sole female and sole male categories for which follicles contain only female or male gonad material, although a representative micrograph of a female indicates the presence of what appear to be male spermatogonia. In our study, gonads overwhelmingly dominated with male or female tissue were fairly common in older animals, yet precursor cells of the alternate sex were always present. Collectively, these studies suggest that all ostreid oysters are protandric hermaphrodites with at least precursor cells of both sexes always present. We recommend that any use of categorical terms such as 'pure female' be accompanied by a thorough description of the gonad cell types present.

Several researchers observed asynchrony in reproductive stage among different parts of the gonad within individual *O. lurida* and other ostreids, particularly in young animals (Coe 1932b, Cole 1942, Loosanoff 1962). We observed this in a few individuals and in such cases the gonad was assigned a stage based on what was most common in the tissue section. We strongly agree with the conclusions of Coe (1932b) and Cole (1942) that tissue squashes or biopsies through holes drilled in the shell, as used in most studies prior to those of Coe, are inadequate to gain an accurate picture of the state of gametogenesis, particularly in early stages.

Seasonality and temperature-dependence of reproduction.—This study provides evidence of a relatively long reproductive season for Olympia oysters in San Francisco Bay. We identified brooding oysters as early as April and as late as November. Studies on seasonal settlement of *O. lurida* have reported a variety of ranges (Table 2), typically from spring to late fall with a peak in approximately June of each year. More restricted seasons occur at more northerly latitudes, presumably reflecting shorter periods of elevated water temperature. Hopkins (1937) reported the presence of brooded larvae in Puget Sound *O. lurida* beginning when waters reached approximately 13 °C (usually in May), peaking in late May to early June, with small numbers as late as October. Coe (1932a) stated that oysters spawned at La Jolla when waters were at least 16 °C, as early as April and as late as October. Seale and Zacherl (2009) studied *O. lurida* settlement at two southern California estuaries. At Upper Newport Bay, spawning occurred from May until November at temperatures of 16 °C or higher, similar to the results of Coe in La Jolla. However at Aqua Hedionda Lagoon, which is located between Newport Bay and La Jolla, temperatures rose above 16 °C in April, but spawning did not commence until June and continued into February, at temperatures as low as 14 °C. From these studies it is clear that the timing of the initiation, peak, and cessation of spawning show significant variation with latitude, location, and between years, but with a consistent peak in late spring to early summer subsequent to warming water temperatures. It is important for restoration activities that depend on natural spatfall to understand relationships between recruitment events and local environmental conditions, particularly temperature patterns.

By opening oysters and examining for the presence of larvae, Hopkins (1937) found that in a Puget Sound population of *O. lurida,* up to 55 % were brooding during the peak season in early June. The number of brooding oysters we observed by histology was very low; this was not unexpected, since brooded larvae are not physically attached to the mother oyster and most could be lost during histological processing. Our study would have benefited from examination of each opened oyster to determine brood presence before further processing and dissection. Nonetheless, identification of brooders provided unequivocal evidence of successful female gamete release and fertilization at particular points in time.

We calculated mean temperatures during the five days prior to sampling events for which brooded larvae were present. The five-day timeframe was based on Hopkins' (1937) report that brooded Puget Sound *O. lurida* larvae were early veligers at this time and all of the brooded larvae we observed were at this or earlier stages. These data indicated that San Francisco Bay Olympia oysters are capable of spawning at mean temperatures of about 13-16 °C, in accordance with the majority of previous studies on this species (Table 2).

TABLE 2.—Reported spawning season (presence of brooded larvae or settlement) for Ostrea lurida, arranged south to north.

Location	Spawning Season	Temperature	Citation
La Jolla, California	April-October	≥16°C	Coe 1932a
Agua Hedionda Lagoon, California	June-February	≥14°C	Seale and Zacherle 2009
Upper Newport Bay, California	May-November	≥16°C	Seale and Zacherle 2009
San Francisco Bay, California	April-November	≥13°C	This study
Coos Bay, Oregon	July-September	≥15°C	Oates 2013
Puget Sound, Washington	May-October	≥13°C	Hopkins 1937

Symbionts.—We found no significant impact of infectious disease in the population studied, in accordance with previous oyster surveys in San Francisco Bay (Friedman et al. 2005) and other bays in California (Moore et al. 2011). *Echeneibothrium*-like cestode larvae are commonly observed in the digestive tract of oysters in California (Moore et al. 2011) and can be found encysted in various clams (Sparks 1985). They have an elasmobranch as a definitive host; clams appear to be the true intermediate hosts while oysters appear to be accidental intermediate hosts. Turbellarian flatworms are uncommon in California oysters and are typically not associated with significant pathological effects. Although these and other metazoa can be difficult to identify to genus or species in tissue sections, the morphology of the two flatworms observed is consistent with them being members of the genus *Urastoma.* The intestinal copepod we observed in one oyster and copepod egg mass in another could not be identified further in our tissue sections although they are likely to be *Mytilicola orientalis*, which Bradley and Siebert (1978) reported was prevalent during spring to summer at 1-2.7 % in *O. lurida* at the Berkeley Marina in San Francisco Bay. The term "microcell" is used for tiny protozoan parasites that are members of the haplosporidian genus *Bonamia* or the taxonomically uncertain *Mikrocytos.* One oyster in our study was infected by a microcell parasite that had characteristics consistent with the genus *Bonamia*, i.e., several microns in diameter and located within hemocytes, and several types of *Bonamia* have been identified in native oysters and flat oysters *(Ostrea edulis)* from other California embayments (Hill et al. 2014). Friedman et al. (2005) reported a microcell in several *O. lurida* from San Francisco Bay that had characteristics more common to *Mikrocytos*. We saw no cases of the leukemia-like disease known as disseminated neoplasia (Elston et al. 1992); Friedman et al. (2005) reported it to be present in two out of 16 San Francisco Bay *O. lurida* populations they sampled. We also did not see haplosporidian plasmodia that were reported in *O. lurida* from Oregon (Mix 1974), nor a *Hexamita* flagellate described in native oysters from Puget Sound, Washington (Stein and Denison 1959), nor Rickettsia-like bacterial inclusions described in native oysters from British Columbia (Meyer et al. 2010). The young ages of the oysters in our samples likely contributed to the relative paucity of symbionts observed.

The minimal impact of disease on *O. lurida* populations from California to British Columbia contrasts with the situation for native ostreid and crassostreid oyster populations in many other parts of the world. Atlantic and Gulf coast populations of the native oyster *Crassostrea virginica* are significantly impacted by the parasitic dinoflagellate-like organism *Perkinsus marinus* (Smolowitz 2013) and Atlantic populations also are limited by presence of the protozoan parasite *Haplosporidium nelsoni* (Burreson and Ford 2004). European populations of the native flat oyster *Ostrea edulis* have been heavily impacted by the protozoan parasites *Bonamia ostreae* (Engelsma et al. 2014) and *Marteilia refringens* (Berthe et al. 2004).

Conclusions.—The design of effective restoration programs can benefit significantly from knowledge of basic biological processes such as ontogenetic and seasonal reproductive patterns. Oysters recruited onto outplanted cultch in San Francisco Bay in late spring rapidly matured, with significant numbers passing through first male and first female stages by fall of the same year. *O. lurida* appears to be minimally impacted by infectious disease. Collectively, these observations show promise for native oyster recovery in San Francisco Bay.

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The economic value of the recreational red abalone fishery in northern California

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There is a long tradition of recreational red abalone (*Haliotis rufescens*) fishing in northern California. The fishery is enjoyed by tens of thousands of fishers along Sonoma and Mendocino counties, but little is known about its economic value. Recreational fisheries are difficult to value because the catch is not sold commercially and the activity is dispersed along the coastline. For this study, we estimated the value to the fishers of the recreational red abalone fishery using the travel-cost estimation method, a non-market valuation approach. Using data for the 2013 season at more than 50 sites, we find that approximately 31,000 fishers derived between $24M and $44M per year of recreational value from the fishery. The lower figure was estimated based solely on fishers' driving costs, while the larger estimate results when also considering the time fishers spent on the activity. Examination of site-level variables influencing the choice made by fishers among the sites shows that key site selection criteria included 1) impacts of a harmful algal bloom in Sonoma County, 2) protection from northwest ocean swell, and 3) presence of amenities such as boat launches and restrooms. We show that the value of the fishery declined nearly $12M after stricter regulations were imposed in 2014

following a harmful algal bloom that killed thousands of abalone in Sonoma County. The economic value of the fishery clearly warrants investment in both the biological and economic sustainability of this important resource.

Key words: Economic Impact, *Haliotis rufescens*, Non-Market Value, Socioeconomics, Sport fisheries, Travel Cost Method

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California has the largest ocean economy in the United States with a gross state product of nearly $42B estimated for the year 2000 (Kildow and Colgan 2005). Recreational fishing is the third most popular water related activity after beach going and swimming. More than 2.7M people enjoy recreational ocean fishing annually in California (Leeworthy 2001). In California, it is estimated that recreational fishing generates an estimated $230M-$610M in direct expenditures per year (2010) (Pendleton and Rooke 2006). Estimates of the total non-market use value of recreational fishing is much higher and ranges between $342M -$2B for the year 2010 (Pendleton and Rooke 2006). As California grows in population, the number of people that participate in recreational fisheries is forecast to increase by 12% per decade (Leeworthy 2001) putting greater pressure on marine resources. Despite the importance of recreational fishing, estimates of market (money anglers contribute through spending) and non-market values (value fishers place on the resources they use) for individual recreational fisheries are scarce.

Red abalone (*Haliotis rufescens*) forms the basis for a recreational fishery in northern California yet little is known about the magnitude of its economic importance. Approximately 35,000 fishers (2000-2014), take 245,000 red abalone (2002-2014) per year (California Department of Fish and Wildlife [CDFW] unpublished data). The majority of the catch (95%) comes from Sonoma and Mendocino counties (Kashiwada and Taniguchi 2007). The recreational red abalone fishery in northern California is the only abalone fishery remaining open in the state. In 1997, commercial fishing was closed statewide and recreational fisheries for abalone were closed south of San Francisco due to declines in stocks (Karpov et al. 2000). The north coast fishery has been restricted to recreational users since 1949 and permits skin (breath-hold) diving only. The fishery is managed for sustainability under the Abalone Recovery and Management Plan (CDFW 2005), which aims to maintain abalone population densities to ensure productivity and consequently the economic viability of the fishery. The Marine Life Management Act (MLMA 1999) supports the management of California's fisheries to sustain, conserve and protect California's marine life including those with economic value.

Despite the recreational, cultural and economic importance of the red abalone fishery, little work has been done to estimate its economic value. Valuation of recreational fisheries is difficult since it is illegal to sell recreationally caught red abalone (aka illegal commercialization) in California (Rogers-Bennett and Melvin 2007). Commercial fisheries, on the other hand, are more easily valued by calculating income from ex-vessel landings. In this paper, the non-market economic value of the recreational red abalone fishery to the fishers, is estimated using the travel-cost method. The relative importance of site attributes at more than 50 sites is examined to determine site qualities used in site selection and the potential losses from a site closure. The non-market value of the fishery is estimated for eight years from 2003 to 2014. The gender and age of the fishery questionnaire respondents is reported to give an indication of demographics in this fishery. Finally, the economic value of the fishery is examined in light of prioritizing funding needs to sustain both the fishery and its economic benefits.

Materials And Methods

The travel-cost method (TCM) (Phaneuf and Smith 2005) is an economic approach used to assign monetary value to non-market goods such as recreational activities or resources. The model's premise is that travel costs are a proxy for the value of unpriced recreational sites, and that people for whom travel costs are lower will visit a site more frequently, mirroring the basic relationship between price and quantity demanded for normal goods. The TCM takes into account the various costs paid by a participant to engage in the activity. These include direct costs such as fees, and other costs such as the opportunity cost of time and vehicle operating costs. Using this information, a travel cost function and demand curve (Figure 1) can be estimated where the consumer surplus is representative of the economic value of the resource to the recreational users. Parsons (2003) provides a detailed overview of the method.

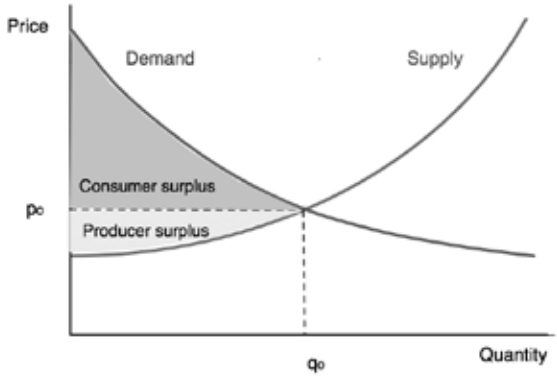

Figure 1. - Demand curve showing the marginal willingness to pay (WTP), with the area under the curve representing the total WTP.

Travel-cost studies follow one of two basic approaches: single-site models and multi-site models. Single-site models construct a demand curve based on the relationship between the cost of visiting a site and the frequency of visits. Multi-site models add in the element of choice from among a set of alternative sites for the same recreational purpose, and isolate the impact of site characteristics on the choice of sites, while also estimating the overall value of recreation. Given that abalone is taken at more than 50 different sites along the coast, a multi-site model was adopted for this study.

Data were drawn from the 2013 season CDFW database of 30,768 abalone report card holders, which represents the population of licensed harvesters, and a telephone survey of a random sample of this population. CDFW conducted the telephone survey of this group in 2014, with 516 responses regarding the 2013 fishing season. Information on the response rate to the telephone survey was unavailable. Respondents to the telephone survey provided demographic information and data on their fishing histories and habits. Of these 516 respondents, 392 also provided detailed catch information (which is not collected in the telephone survey) to the CDFW via its reporting system. Because we had both demographic and catch data from these 392 respondents, they were used as our sample for the travel-cost analysis.[1]

1 A representative sample (n) size is commonly obtained by solving $n=(Z^2pq)/e^2$ where n is sample size, Z is the value obtained from a normal curve at the desired confidence level (95%), e is the desired level of A representative sample *(n)* size is commonly obtained by solving $n=(Z^2\ pq)/e^2$ where n is sample size, Z is the value obtained from a normal curve at the desired confidence level (95%), e is the desired level of precision, p is the estimated proportion of an attribute that is present in the population, and q is $1-p$. A conservative approach assumes the maximum variance implying $p = q = 0.5$ with a confidence interval of 95% and a maximum sample error of 5%, then the optimal sample is 385 observations. Furthermore, the unit of analysis here is recreational trips and 392 individuals account for 1,520 trips.

There is a risk that this group is not representative of the overall population; those reporting may collect more or less than the average number of abalone, prefer certain kinds of sites, be demographically distinct or in some other relevant way diverge from the population. We do not have any information to indicate specific ways in which our sample may differ from the population at large.

In order to construct a database of trips, the unit of analysis in a travel-cost study, we examined respondents' reported abalone catch by date and fishing site from the report card database and cross-referenced the information with the number of trips they reported in the telephone survey. Analysis was performed on the resulting 1,537 trips.

The site attributes were chosen based on consultation with CDFW staff experts. Attributes selected were those perceived to impact where fishers choose to fish, to vary across sites and for which information exists for all sites. Some attributes found to be important in previous research, such as abundance and size (Chen et al. 2013), have not been measured systematically for all 51 sites (or even a significant subset of the most important sites), so we could not compare sites with respect to those variables. The two most important variables studied by Chen et al. (2013) and in this study were the ease of access to the water and the protection of sites from swells. While most of the site attributes (Table 1) were specific to that site and independent of neighboring sites (e.g., parking, bathroom facilities), two of the attributes influenced multiple neighboring sites. Protection from wave exposure by a headland may influence the number of days of accessibility to a number of neighboring sites. Also, a harmful algal bloom (HAB) in 2011 caused significant declines in abalone density within all of the Sonoma County area sites (Porzio 2014).

TABLE 1.—Site characteristics used for travel-cost analysis

Attributes	Variable name	Description	Type
Access	ACC	Difficulty of access to the water from parking area, often determined by steep terrain.	Category: 1-3 1 = easy, safe access 3 = most difficult or dangerous access
Boat launch	BL	Existence of a boat launch.	Dichotomous: 0 = no 1 = yes
Parking	Parking	The availability of parking.	Category: 1-3 1 = abundant parking 3 = very limited parking
Bathrooms	Bath	Existence of public bathrooms.	Dichotomous: 0 = no 1 = yes
Exposure to ocean swell	PROTEC	The degree of protection afforded by geographic features to prevailing NW swells.	Category: 1-3 1 = least exposed 3 = most exposed
Harmful algae bloom	HAB	Site affected by 2011 harmful algae bloom.	Dichotomous: 0 = no 1 = yes
Pay for parking	PAY	Whether parking requires payment of a fee.	Dichotomous: 0 = no 1 = yes

We assume that the welfare obtained by an individual *i* from a trip to the site *j* on decision occasion *t* is given by the following utility function:

$$U_{ijt} = \beta_1 TC_{ij} + \beta_2 ACC_j + \beta_3 BL_j + \beta_4 Parking_j + \beta_5 Bath_j + \beta_6 PROTEC_j + \beta_7 HAB_j + \beta_8 PAY_j + \mu_{ijt}$$

In this equation TC*ij* is the travel cost from each *i*-th individual´s origin to the destination *j*. Travel cost includes the cost of operating a vehicle, for which we used the federally specified rate of $0.565 per mile for 2013. Distances and travel times were calculated with Google Maps (V2), using respondents' home zip code as trip origin and the coordinates of the abalone site visited as the destination. To this we added the opportunity cost of time traveling and spent at the recreation site. Common practice (Cesario 1976; Parson 2003) is to use a fraction, which we set at 0.5, of the person's wage. We encountered a gap in the data because many of the respondents to the telephone survey declined to provide income information and no income data is contained in the report card database. The model was therefore estimated with two variants on the definition of travel cost. For those respondents without income data, we used the average income for their zip code of residence. We ran one regression using only the driving cost (TC1) in order to use the whole sample with consistent data for every trip. This approach underestimates the travel cost and, consequently, recreational value, representing therefore a lower bound. TC2 uses income data (both individual and zip code) and adds four hours spent at the dive site (in and out of the water) to calculate the travel cost.

Calculating willingness to pay (WTP) is complex with this kind of model and ours is especially involved since there are over 50 alternative choices for sites to collect abalone. The generic formula for WTP is known as the "log-sum" formula and is given by:

$$WTP = \frac{1}{\theta}\left[\ln \sum_{j=0}^{J} e^{V_j^1} - \ln \sum_{j=0}^{J} e^{V_j^0}\right]$$

Where j represents the recreation site, *j*=1, 2 ... J, and superscripts 0,1 represent the initial and final situations, respectively. θ is the coefficient on travel cost (in absolute value). The final situation is characterized by whatever policy (or, generically, change) we are evaluating, which could include a change in a site's attributes, that is, in elements of every V*j*, or elimination of one or more sites. In this latter case, the site(s) in question simply disappear from the sum of values of all the sites.[2]

On the other hand, if the quality of an attribute changes for all sites, the WTP is:

$$WTP = \frac{\beta_1 \Delta X}{\theta}$$

The coefficients βi capture preferences for various levels of the attribute. A positive and significant coefficient ($\beta i > 0$) means that the increase in the attribute results in a higher likelihood that the site is selected. The other relevant coefficient for calculating the WTP is θ, which captures the reduction in an individual's utility as the travel cost rises (or the marginal utility of income in absolute value). Regressions were run in the Stata software package (V12) using

2 In other words, we replace $\sum_{j=0}^{J} e^{V_j^1}$ with $\sum_{j=0}^{J_k} e^{V_j^1}$ in which $J_k < J$.

a conditional logit model. An additional regression to test the validity of results was run on the travel-cost-only data with a mixed logit model, which accounts for the possible independence of irrelevant alternatives (IIA) and captures the unobserved heterogeneity of the sample.

In addition, to gain an understanding of the trajectory of recreational value over the years, we applied the per-trip value calculated for 2013 to the years 2000-2012 and 2014. Total fishing trips for these years was calculated by multiplying the number of report card holders by the average trips per report card holder as reported in the telephone surveys for each year, including respondents who took no trips. Average trips figures were available for 2003-2006, 2008, 2012 and 2014, so these are the years for which total values were calculated. This extrapolation provides only a very coarse approximation; per-trip values can be expected to vary year to year with changes in regulations, abalone abundance, weather, economic conditions and other factors. Future research should use trip values specific to each year, work that was beyond the scope of this study.

Results

The per-trip recreational value of each site was estimated by two travel cost models (Table 2). The values appear as negative numbers because they refer to the loss that would result if a particular site were closed or otherwise no longer available. The sites for which the values are greatest are largely clustered between Albion and Fort Bragg on the Mendocino coast, with losses in the range of $2.50-$5.00 per trip. The modest figures are explained by the fact that divers can simply opt for another of the long list of sites if only one is closed; sites are partially substitutable. The impact of closing all sites simultaneously is a loss $219-$406 per trip, depending on the model chosen. The 2013 telephone survey reports 30,678 fishers take on average 3.6 trips per year. The total net recreational value estimated for the fishery in 2013 was between $24M based on the driving cost alone (TC1), and $44M when considering both driving cost and the time spent on the trip (TC2) (Figure 2).

Travel cost is shown to be significant at the 99.9 percent confidence level in all three models (Table 3). The results of the two regressions runs to generate the value estimates, plus, in the rightmost column, the mixed logit regression run as an additional test of the validity of the analysis are shown revealing the concordance of the 3 models (Table 3). Of the site characteristics, impact from the 2011 HAB, bathrooms, boat launch and exposure to swell (listed in descending order of their coefficients) were all significant at this level in all models and had the expected signs (negative or positive impact on utility). Ease of access to the water was significant at the 95 percent confidence level in the TC1 and TC2 models but not in the mixed logit. The requirement to pay for parking, on the other hand, was significant (99 percent confidence level) for the mixed logit only. The HAB attribute, which is associated lower abalone abundance, has by far the largest coefficient (impact on site choice). The affected Sonoma County sites received less visitation despite their closer proximity to the major population centers around San Francisco.

Extrapolating the per-trip values for 2013 to other years, we show an initial period of steady recreational values (2003-2005) near $40M, followed by a peak in value in 2006 of just under $50M (Figure 2). The values for 2008 and 2012 were similar to the estimate for 2013 ($44M). The slightly lower values in the early 2000s were due to a lower average number of trips taken per report-card holder. The value dropped dramatically in 2014 (~$32M) as report card sales fell by 16 percent, to their lowest levels within the 15 years for which we have data. Trips per fisher also declined, by 13 percent, in the 2014 season.

TABLE 2. —Recreational value by site shown as the economic wellbeing reduction per trip, in dollars, that would result from closing each fished site individually. Cs = consumer surplus; wtp = willingness to pay). Sites appear in order from north to south.

		Model 1: Driving costs only		Model 2: Driving costs and time	
COUNTY	**SITE**	Mean	Standard deviation	Mean	Standard deviation
Del Norte	Crescent City	-0.33	2.83	-0.54	3.44
Del Norte	Other Del Norte County	-0.25	0.88	-0.53	1.91
Humboldt	Trinidad	-0.54	1.93	-1.22	4.73
Humboldt	Punta Gorda	-0.20	0.37	-0.43	0.70
Humboldt	Shelter Cove	-0.88	1.07	-1.75	1.74
Humboldt	Other Humboldt County	-1.07	1.41	-2.14	2.28
Mendocino	Usal	-0.94	0.49	-1.86	0.88
Mendocino	Hardy Creek	-0.79	0.31	-1.51	0.56
Mendocino	Abalone Point	-1.03	0.38	-1.99	0.70
Mendocino	Westport	-0.73	0.27	-1.39	0.50
Mendocino	Bruhel Point	-0.27	0.10	-0.52	0.18
Mendocino	MacKerricher State Park	-1.37	0.45	-2.61	0.84
Mendocino	Glass Beach	-1.47	0.48	-2.78	0.89
Mendocino	Georgia Pacific Mill	-1.68	0.54	-3.14	0.99
Mendocino	Todd's Point	-1.30	0.41	-2.41	0.74
Mendocino	Hare Creek	-1.62	0.51	-3.06	0.95
Mendocino	Mitchell Creek	-0.64	0.15	-1.21	0.27
Mendocino	Jughandle State Reserve	-1.05	0.21	-1.95	0.39
Mendocino	Caspar Cove	-1.52	0.29	-2.88	0.55
Mendocino	Russian Gulch State Park	-2.70	0.49	-5.04	0.89
Mendocino	Jack Peters Gulch	-0.75	0.13	-1.41	0.23
Mendocino	Mendocino Headlands	-2.31	0.40	-4.29	0.73
Mendocino	Gordon Lane (Spring Ranch)	-0.46	0.07	-0.88	0.14
Mendocino	Van Damme State Park	-2.61	0.41	-4.86	0.77
Mendocino	Dark Gulch	-1.05	0.16	-1.97	0.29
Mendocino	Albion Cove	-2.99	0.45	-5.54	0.82
Mendocino	Salmon Creek	-0.83	0.12	-1.53	0.22
Mendocino	Navarro River	-1.98	0.30	-3.67	0.53
Mendocino	Elk	-2.45	0.42	-4.53	0.73
Mendocino	Point Arena Lighthouse	-0.90	0.19	-1.68	0.33
Mendocino	Point Arena (Arena Cove)	-3.60	0.84	-6.58	1.44
Mendocino	Moat Creek	-3.14	0.76	-5.74	1.32
Mendocino	Schooner Gulch	-1.03	0.26	-1.89	0.46
Mendocino	Anchor Bay	-1.14	0.33	-2.18	0.62
Mendocino	Robinson Point	-0.21	0.07	-0.40	0.12
Sonoma	Gualala Point	-0.34	0.11	-0.63	0.20
Sonoma	Sea Ranch	-0.58	0.19	-1.09	0.36
Sonoma	Black Point	-0.42	0.14	-0.79	0.27
Sonoma	Stewart's Point	-0.49	0.17	-0.93	0.33
Sonoma	Rocky Point	-0.22	0.08	-0.42	0.15
Sonoma	Horseshoe Cove	-0.60	0.21	-1.13	0.42
Sonoma	Fisk Mill Cove	-1.10	0.42	-2.07	0.81
Sonoma	Salt Point State Park	-1.07	0.41	-2.00	0.81
Sonoma	Ocean Cove	-1.11	0.43	-2.09	0.86
Sonoma	Stillwater Cove	-1.53	0.61	-2.87	1.20
Sonoma	Timber Cove	-0.99	0.39	-1.86	0.79
Sonoma	Fort Ross	-0.99	0.40	-1.85	0.80
Sonoma	Reef Campground (Pedotti)	-0.79	0.32	-1.47	0.65
Sonoma	Jenner	-0.41	0.17	-0.76	0.35
Sonoma	Bodega Head	-1.57	0.68	-2.92	1.47
Marin	Tomales Point	-0.92	0.41	-1.69	0.91
Sum CS per site		-58.97	0.85	-110.64	1.04
Total WTP for closure of all visited sites		-218.71	24.12	-405.84	43.36

TABLE 3. —Regression results.

	Model 1	Model 2	Model 3
	TC1	TC2	TC1 mixed logit
TC1	-0.0173***		-0.0221***
	(-18.73)		(-19.93)
TC2		-0.00919***	
		(-18.56)	
Access	0.114*	0.105*	-0.0815
	(2.41)	(2.23)	(-0.99)
Boat launch	0.574***	0.575***	0.692***
	(7.94)	(7.95)	(4.18)
Parking	0.0764	0.0847	0.0679
	(1.40)	(1.55)	(0.87)
Bathrooms	0.627***	0.626***	0.817***
	(7.40)	(7.38)	(6.47)
Exposure to ocean swell	-0.377***	-0.373***	-0.374***
	(-8.03)	(-7.99)	(-4.54)
Harmful algal bloom	-1.470***	-1.421***	-2.932***
	(-15.90)	(-15.58)	(-10.30)
Pay for parking	0.0758	0.0755	-0.516**
	(1.08)	(1.08)	(-2.85)
Number of tripstrips	15201520	15131513	15131513

t statistics in parentheses $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$

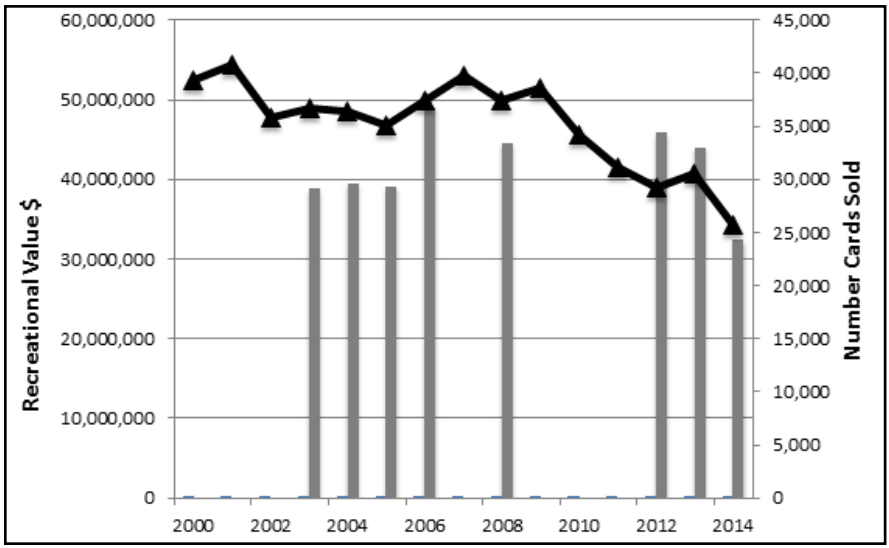

FIGURE 2.—Recreational value of the red abalone fishery in northern California for the years with data on the number of trips extrapolating the per-trip value from 2013 to the other years (2003, 2004, 2005, 2006, 2008, 2012, 2014) shown using the height of the bars. The second Y axis shows the total number of abalone report cards sold per year from 2000-2014 shown using the solid triangles. Note: the automated license system went into effect in 2010 reducing the possibility of illegally purchasing two cards in one year.

The 2014 fishing season was the first year marked by the full impact of the HAB event and associated regulation changes, such as the reduction in the annual bag limit, the new late start time (8:00AM) and the closure of the historically most heavily used site in the fishery – Fort Ross State Park.

Finally, we report descriptive statistics of the fishers from a sample to give a sense of respondent characteristics. We find that 95 percent of the sample was from California and 92 percent were male. The age distribution shows 73 percent over the age of 35, with an average of 15 years of abalone fishing experience (Figure 3). As noted above, the average number of trips was 3.6 and the average days fishing was just over 4.0, with an average of 8.4 abalone caught during the season. Note that these figures include respondents who purchased report cards but did not end up fishing.

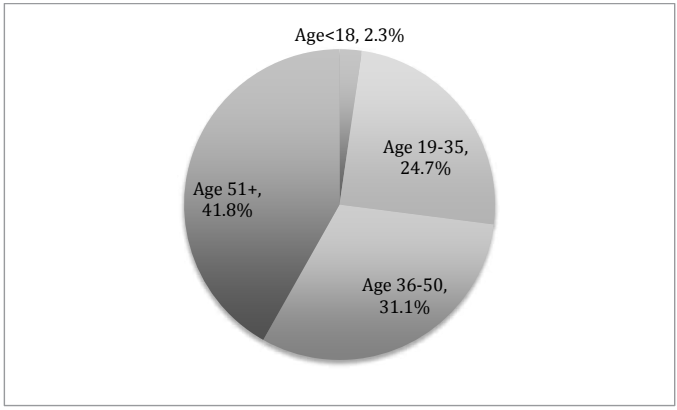

FIGURE 3.—Age distribution of 2013 abalone fishers included in sample.

DISCUSSION

The red abalone fishery is worth $24-$44 M in annual non-market benefits to recreational fishers (Figure 2). We consider these conservative estimates of the value of the fishery because they are based on travel and time costs alone, excluding other trip related costs (lodging and meals), as well as associated gear (e.g. wetsuits, abalone floats, irons and licenses). These results are based on the 392 respondents many of whom (>40%) are more than 50 years old. Chen et al. (2013) found, that abalone fishers spent an average of $193 on dive equipment, $167 on lodging and camping and $140 on food and beverages from stores, which adds up to 50 percent of overall expenditures. Transportation expenses (excluding the opportunity cost of time, accounted for 28 percent of spending). While their study was based on only 90 respondents, the results do suggest that collecting additional data for a fuller accounting of travel costs is warranted in future years to get a fuller picture of the economics of this fishery.

We recommend some modest changes in the routine annual data collection effort that would permit creating a more robust time-series of economic value for the fishery. The travel-cost estimation method as applied here requires data on trips taken by individual fishers, including the destination, associated spending and number of people traveling together for each trip, as well as demographic information on the fishers. We recommend that this sort of data on fisher trips (rather than fishing day) be collected directly through

the annual telephone survey of report card holders. To date, surveys have not collected data on individual trips. As a result, in this analysis, we reconstructed a profile of each trip based on location and date information reported on the capture of individual abalone, cross-referenced with the number of trips each respondent reported. Collecting specific trip data would save substantial time on analysis and permit inclusion of costs beyond driving expenses and the opportunity cost of time, allowing for a more comprehensive estimate of fishery value. This would avoid the strategy employed in this analysis, using per-trip values from 2013 and extrapolating these to other years. Finally, the recommendation we are making to collect trip data would facilitate an economic impact analysis.

The multi-site travel-cost estimation method is useful when weighing the economic effects of management actions which would open or close one specific fishing site or a group of sites. Multi-site information can be used to estimate the specific economic losses (or gains) from closing (or opening) sites, based on their attributes and levels of use. In this case the site information was useful in understanding the economic impacts of the regulation changes made following the HAB. The full impacts of the HAB and the associated regulation changes, including a reduction in the annual bag limit, the later start time, and the closure of Fort Ross took effect in the 2014 season. In 2014, the total value of the fishery dropped by $12M from $44M to $32M coincident with a 16 percent decline in report card sales and a 13 percent drop in average annual fishing days per fisher. Although we cannot assign causality, the figures do give managers a quantitative indication as to the economic dimensions of the HAB event and subsequent regulation changes.

Because similar valuations are lacking for other major marine recreational fisheries in California, we have little basis for comparisons. Most economic analyses of California fisheries have consisted of estimates of recreational expenditures or gross commercial revenue to fishers. These estimates are not comparable to the figures we have generated with the TCM, which is the net benefit–the consumer surplus–accruing to fishers of the fishery. Expenditures for the recreational spiny lobster (*Panulirus interruptus*) fishery in southern California, was calculated at $37M per year (Hackett et al. 2013). While, the two largest commercial fisheries in California (by ex-vessel value) are market squid (*Doryteuthis [Loligo] opalescens*) ($58M) and Dungeness crab (*Metacarcinus [Cancer] magister*) ($46M) from 2008-2012 (Rogers-Bennett and Juhasz 2014). Without venturing any speculations about the economic value of these fisheries–which is equal to the producers' profits plus consumer surplus–we simply note that their gross expenditures are of a similar magnitude as the economic value of the red abalone fishery. While these are apples-and-oranges comparisons, we can look at additional calculations to estimate comparable economic impact figures for red abalone. The total economic impact of red abalone recreational fishing from previous work was found to be $26.7M for the 2014 season (Reid et al. 2016). Direct expenditures, the figure most similar to the $37M estimated by Hackett et al. (2013) for spiny lobster, were found to be $18.6M for red abalone in California (Reid et al. 2016) (Figure 4).

Conclusions

The loss of both the recreational and commercial abalone fisheries in southern California in 1997 makes it clear that this resource is vulnerable to depletion and collapse. The economic value estimates presented here demonstrate that there are tens of millions of dollars in recreational benefits at stake if the North Coast recreational fishery were to suffer the same fate. The economic importance of the fishery provides policy-

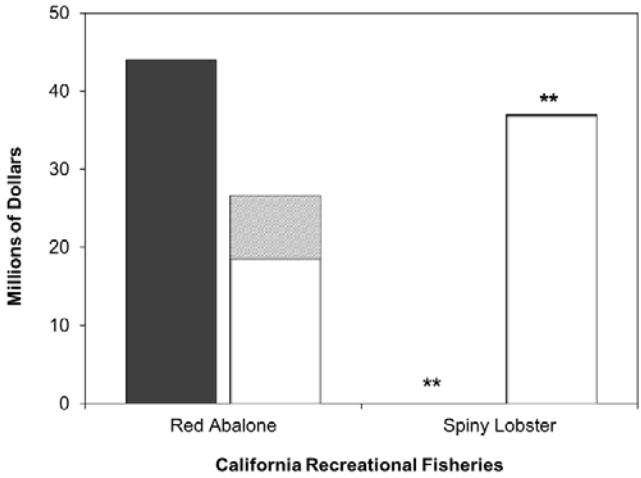

FIGURE 4.—Economic Value And Economic Impact Of Northern California Red Abalone Recreational Fishery Compared to spiny lobster economic impact. Black bar = Economic value from travel-cost method (this study); white bar = economic impact: direct expenditures (red abalone – (Reid et al. 2016); spiny lobster - (Hackett et al. 2013)); patterned bar = economic impact: indirect + induced costs (Reid et al. 2016). "**" = no comparable analyses available for spiny lobster other than for direct expenditures.

makers and managers an indication of the high priority of investing in science and law enforcement to sustain the resource. Analyses such as this one have yet to be done for many recreational California fisheries and are desperately needed to inform management. Quantifying the economic importance of a fishery reveals that an investment in resource management can enhance the long term economic benefits derived from the fishery.

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A potential predator-prey interaction of an American badger and an Agassiz's desert tortoise with a review of badger predation on turtles

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Keywords: American badger, *Taxidea taxus*, Agassiz's desert tortoise, *Gopherus agassizii*, predator, prey

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The federally threatened Agassiz's desert tortoise (*Gopherus agassizii*) was listed under the U.S. Endangered Species Act in 1990, but thus far, recovery efforts have been unsuccessful (U.S. Fish and Wildlife Service [USFWS] 2015). Predation has been identified as a contributing factor to declining *G. agassizii* populations range-wide (e.g., Esque et al. 2010, Lovich et al. 2014). Understanding and managing for predator-prey dynamics is thus an important part of the recovery and conservation of this threatened species (USFWS 2011). Desert tortoises have a host of predators at all stages of their life cycle. Over 20 species of birds, mammals, and reptiles have been recorded as known or suspected predators (Woodbury and Hardy 1948, Luckenbach 1982, Ernst and Lovich 2009). American badgers (*Taxidea taxus*, family: Mustelidae) are confirmed excavators of desert tortoise nests (Turner and Berry 1984). They are also suspected predators of adult desert tortoises, a possibility which has been presented in some studies but without empirical verification (Luckenbach 1982, Turner and Berry 1984). Active mostly at night, badgers are solitary, secretive predators (Lindzey 1978, 1982; Armitage 2004) that are extremely difficult to observe in predatory encounters. Recently, strong circumstantial evidence presented by Emblidge et al. (2015) suggests that badgers do prey on adult Agassiz's desert tortoises based on observations of more than two dozen dead tortoises in the Western Mojave Desert of California. In this note, we present another case of potential badger predation on a large adult desert tortoise in the Sonoran Desert of California. Collectively, these recent two cases potentially indicate that badger predation may be more common and widespread than previously thought. In

addition, we review the worldwide literature of badger predation on turtles in general and summarize reported badger observations in Joshua Tree National Park, where our observation occurred, over a period of 55 years.

We initiated research on tortoise demography and reproduction in the vicinity of the southern Cottonwood Mountains of Joshua Tree National Park in March 2015. This area is characterized by gently sloping bajadas to the south and steep hills and boulder piles to the north. Vegetation at the study site is typical of Sonoran Desert plant communities with creosote scrub (*Larrea tridentata*) on the bajadas interspersed with ironwood (*Olneya tesota*), blue palo verde trees (*Parkinsonia florida*), and ocotillos (*Fouquieria splendens*). During our surveys on 1 April 2015, we found one of the two largest tortoises in our marked population—a large (29.6 cm carapace length, 5,000 g) and outwardly healthy male *G. agassizii*—and marked it for future identification. No signs of physical distress or upper respiratory tract disease (URTD) were observed in this tortoise or any other tortoise in the population. The tortoise was in the mouth of a burrow under the caliche layer in a large wash that curves around a southern toe of the Cottonwood Mountains at an elevation of 639 m. The wash contained scat from coyotes (*Canis latrans*) and bobcats (*Lynx rufus*) and had many mammal tracks. On 13 April 2015, we found the same tortoise dead (Figure 1) approximately 80 m from the point of first capture and 4-5 m away from another burrow that it had presumably used at some time, which we concluded from the burrow's well-maintained appearance, lack of cobwebs or debris, presence of tortoise tracks, and a large size and shape consistent with the dead tortoise. No animal tracks or sign were otherwise observed near the dead tortoise. The carcass was overturned onto its carapace with the limbs intact and the head nearly severed from the neck. There was a small hole in the left inguinal pocket through which the tortoise had been eviscerated as intestines were pulled

FIGURE 1.—Adult male *Gopherus agassizii* as found dead in southern Joshua Tree National Park on 13 April 2015. The tortoise was eviscerated through the left rear limb pocket and the neck was nearly severed, implicating the American badger *(Taxidea taxus)* as the predator. Intestines can be seen in the foreground.

away from the body. No tooth marks, scratches, or punctures were observed on the shell or limbs. We concluded that death occurred recently, possibly within 24 hours of discovery, based upon the presence of tacky blood on the carcass and lack of a strong decomposition odor or insects. The recent death of an overtly healthy tortoise would suggest injuries were due to predation as opposed to scavenging. Additionally, scavengers are likely to break or crack bones and scutes as they dry following the death of a tortoise (Berry et al. 2013), which we did not observe.

We were puzzled by the fact that the limbs of the carcass were intact because common predators like coyotes, foxes, and bobcats would be expected to injure or consume these muscle-rich parts of adult tortoises (Woodbury and Hardy 1948, Peterson 1994). This was the first time since we started conducting research in the park in 1997 (Lovich et al. 1999) that we observed a tortoise killed in this manner. Common canid and felid predators are known to chew and scratch tortoise shells, even breaking parts of the shell bones and scutes in the process (Coombs 1977, Peterson 1994, Lovich et al. 2014), but none of those characteristics were noted. Esque et al. (2010) identified coyote predation in the Mojave Desert by looking at two common features among carcasses—bite or chew marks on the shell and limbs of desert tortoises and tracks surrounding the carcasses. Additionally, Esque et al. observed higher mortality rates of smaller adult tortoises within their population that they hypothesized were due to the limited gape of a coyote in relation to tortoise body size. The mountain lion (*Puma concolor*), a less common carnivore in Joshua Tree National Park, is also known to leave teeth marks and puncture wounds on tortoise shells, or even remove large portions of the shell (Medica and Greger 2009, Riedle et al. 2010, Medica et al. 2012). Another potential predator of desert tortoises, the feral dog (*Canis lupus familiaris*) (Boyer and Boyer 2006; Berry et al. 2013, 2014), is not known to inhabit our study area. Common ravens (*Corvus corax*), a highly visible predator of juvenile and immature desert tortoises (Berry et al. 2016), are observed infrequently at our study site. Ravens are less known as predators of adult desert tortoises, but they have been observed attacking adult desert tortoises in the Mojave Desert on a few occasions (Woodman et al. 2013). The attacks resulted in injuries that differ from what we observed. Woodman et al. found tortoises overturned onto their carapaces (the mechanism by which this occurred was not seen), and injuries were observed only in the cloacal region above the tail.

Following the consideration of potential predators but finding little in common with our observations, it was suggested that the tortoise may have been killed by an American badger (R. Averill-Murray, Desert Tortoise Recovery Office, USFWS, personal communication). As a result, we were directed to the research of Emblidge et al. (2015) who recently documented a case of suspected badger predation on a desert tortoise population in the Mojave Desert. Their observations included 27 tortoise carcasses over a period of two years which shared many similarities with ours. Emblidge et al. found tortoise carcasses overturned onto their carapace. They describe their suspected badger kills as characteristically eviscerated through a prefemoral socket with limbs remaining intact. Heads were often removed completely, but occasionally were left incompletely severed. None of their carcasses displayed scratch or chew marks on the shell, and were often found inside or nearby tortoise burrows.

Badgers were not directly observed killing or eating tortoises in the Emblidge et al. (2015) study. However, the use of camera trapping strongly implicated badgers in the deaths of the tortoises. Photo sequences in the Emblidge et al. study showed badgers investigating or digging at tortoise burrow entrances where tortoises were recently observed alive, or

following in the direction of tortoises. The tortoises were later found dead. The similarity of conditions for carcasses observed by Emblidge et al. and our observation suggested that a badger was also the predator at Joshua Tree National Park.

In an effort to determine if a badger was the potential predator of the tortoise in our study, two trail cameras were placed in pinch points of the wash on 29 April 2015, just above the dead tortoise's burrow at first capture. After two months with no carnivore activity captured on camera, the camera angles were adjusted on 15 June 2015 to provide a wider view of the ground. On 27 June 2015, 75 days after the tortoise carcass was found, a badger was photographed by the trail camera (Figure 2). It was walking down the wash,

FIGURE 2.—An American badger *(Taxidea taxus)* photographed by a trail camera on 27 June 2015 in southern Joshua Tree National Park near the site where the tortoise carcass displayed in Figure 1 was discovered.

toward the former burrow of the dead tortoise. At the time the badger was captured on camera, the tortoise carcass remained at the location where it was discovered. The carcass was completely mummified and was missing additional anatomical parts since its initial discovery: three appendages, the head, and the eviscerated intestines. It is unlikely that the mummified carcass would have attracted the badger to the area. No other potential predators, mammalian or avian, were captured on camera during the time it was deployed. The camera trap was located approximately 80 m linear distance up the wash from the known tortoise burrow, and the carcass was discovered another 86 m linear distance down the wash from the burrow (a total linear distance of 160 m from camera trap to carcass).

Miller and Stebbins (1964) noted that, while the climate and environment in Joshua Tree National Park were suitable for badgers, they are not found in great numbers in the

park. However, Luckenbach (1982) stated that badgers are more common in the California desert than is generally recognized. This latter statement is supported by records of reported wildlife sightings maintained by staff at Joshua Tree National Park. Between 1960 and 2015 there were at least 90 reported badger sightings (both young and adults) throughout the park, including in the Cottonwood area, and occasionally sightings were of two to three badgers at once (Figure 3). It is not known how many sightings in contiguous years were of the same individual badger, but given the numbers reported from throughout the park, badgers are not necessarily rare or localized in the park.

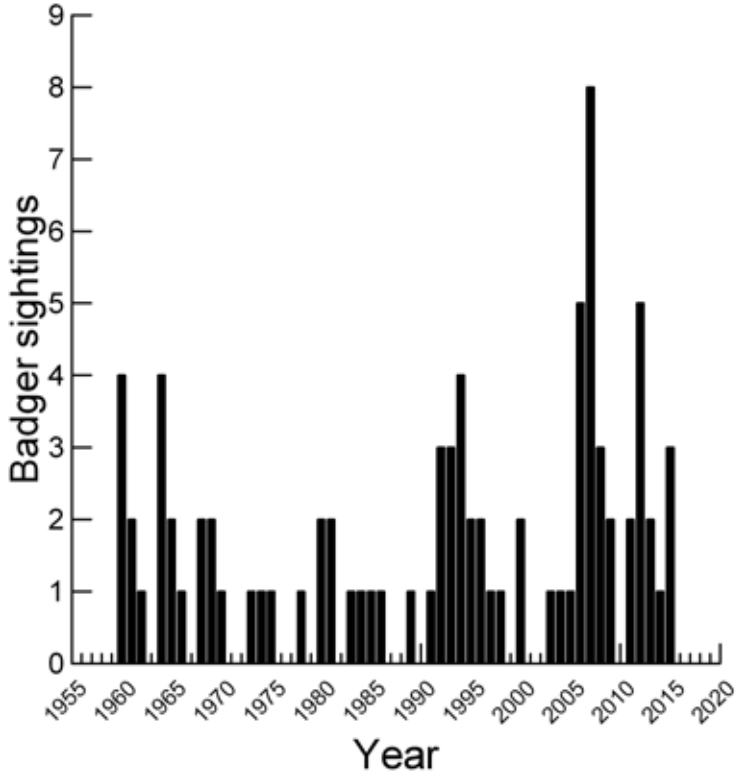

FIGURE 3.—The number of badger sightings reported per year in Joshua Tree National Park for the years indicated. Data are from a database maintained by the Park, and encompass sightings from all areas of the park which includes sightings in the Cottonwood area. A very small number of sightings involved 2-3 badgers at once, possibly of females with young.

The American badger diet in non-desert ecosystems is mostly fossorial or semi-fossorial mammalian species (biomass 95.4%), with the second highest portion comprised of reptilian prey (biomass 3.9%) (Lampe 1982, Marti et al. 1993). However, some studies indicate that reptiles are a larger part of predator diet in desert ecosystems than in prairie or other regions (Delibes and Hiraldo 1987, Hernández et al. 1994). The preferred prey of badgers includes ground squirrels (previously *Spermophilus* spp., but see Helgen et al. 2009) and prairie dogs (*Cynomys* spp.) (Snead and Hendrickson 1942, Messick and Hornocker 1981, Goodrich and Buskirk 1998), only the former of which (including three species: whitetail antelope squirrel [*Ammospermophilus leucurus*], California ground

squirrel [*Otospermophilus beecheyi*], and round-tailed ground squirrel [*Xerospermophilus tereticaudus*]) occurs in Joshua Tree National Park.

In non-desert ecosystems, badgers forage opportunistically, and can have a widely varied diet depending on the availability and abundance of local prey (Lampe 1982). If ground squirrels are not available, badgers will switch to alternative sources, including birds, eggs, reptiles, amphibians, or even plant material (Verts and Carraway 1998). This prey-switching in carnivores can occur during periods of persistent drought when low rainfall causes a reduction in prey populations (Woodbury and Hardy 1948, Chew and Butterworth 1964, Kenagy and Bartholomew 1985, Prugh 2005). Desert rodent populations are influenced by winter precipitation rates and show declines following 10-12 months of low rainfall (Beatley 1976, Whitford 1976, Dickman et al. 1999). Prey-switching response in coyotes from rodents and lagomorphs to tortoises has previously been documented in Joshua Tree National Park and other parts of the Mojave Desert (Peterson 1994, Esque et al. 2010, Lovich et al. 2014), and may be occurring in badgers as well. Precipitation rates at Joshua Tree National Park were at historic lows at the time of our observations due to protracted drought in California (Mann and Gleick 2015). Our study area in the Park was categorized as being in a Severe Drought during the entire 12 months preceding the initiation of our study, and remained in this category during the entirety of the tortoise activity season of 2015 (USDM 2016). Switching from favored prey as it becomes less available has been documented in badgers (Messick and Hornocker 1981), but whether or not the drought conditions caused the suspected badger to prey upon the tortoise is unknown.

Badgers capture their prey by cornering them underground and digging at the entrance (Coulombe 1971), although they occasionally catch prey above ground (Sawyer 1925). Badger presence can be confidently inferred from the existence of extensive diggings at burrow sites of prey items, such as with the removal of large rocks and excavation of burrows (Long and Killingley 1983, Desmond et al. 2000, Armitage 2004, Eldridge 2004). Both of these characteristics were exhibited in a maze of tunnels and excavations occurring in the area around the former burrow of the dead tortoise (Figure 4). Searches for these types of badger sign yield a higher likelihood of detecting badger presence than camera trapping, and therefore it is one of the most useful ways to determine badger presence (Harrison 2015). In conjunction with such diggings, another indicator of badger presence is the decapitation or partial decapitation (Emblidge et al. 2015) of their prey. For example, badgers are known to decapitate prairie dogs, and then consume the entirety of the animal except the head and dorsal fur (Lindzey 1994). Armitage (2004) noted a few marmot (*Marmota flaviventris*) mortalities attributed to badgers in which only the head of the marmot was later discovered.

Uniquely, all suspected badger predation events on desert tortoises described by Emblidge et al. (2015) and this study report evisceration while the limbs remained untouched. It is possible that the armoring provided by the antebrachial scales on the front legs of tortoises are an impediment to badgers, but that seems unlikely since other predators such as coyotes are capable of consuming these portions of a tortoise (Peterson 1994, Lovich et al. 2014). The method of predation observed by Emblidge et al. and this study could also be related to available nutrients and water content of differing portions of the tortoise. According to the United States Department of Agriculture Nutrient Data Laboratory (2015), the internal organs of livestock animals contain a higher content of vitamins and most minerals, as well as slightly higher water content than the same amount of animal muscle tissue. It is possible that, due to lack of available water, a badger may consume only the parts of the tortoise

FIGURE 4.—Cut bank in the wash adjacent to where the desert tortoise carcass shown in Figure 1 was discovered. Note extensive excavations under the caliche layer of the soil horizon attributed largely to a badger digging to excavate rodent prey. When first captured alive, the male desert tortoise was using one of the burrows (not shown) in the same bank, and he may have enlarged some of the holes started by the badger. Overburden above excavations ranges from 0.5 - 1.0 m in thickness.

highest in nutrients and water content (internal organs), including the bi-lobed bladder of a tortoise (located just inside the inguinal area where our tortoise was eviscerated) which acts as a reservoir for water (Nagy 1988), and leave less nutritious parts of the tortoise which would require investment of water to digest with a lesser return in nutritional value. It's possible the tortoise was overturned onto its carapace in order for a badger to facilitate access to this area. While bladder contents of tortoises may be distasteful to some predators (e.g., kit fox [*Vulpes macrotis*], Patterson 1971), this may not act as a deterrent during drought conditions. Additionally, badgers might decapitate their tortoise prey in order to easily drink blood, which would provide another means of water intake.

Badgers of various species have been documented as predators of adult turtles for roughly 123,000 years (Kahlke et al. 2015) as detailed in Table 1. Although badgers in the genera *Meles*, *Taxidea*, and *Mellivora* are not closely related, their general ecomorphological convergence allows them all to be occasional predators of turtles (Koepfli et al. 2008). Although the published literature on this poorly documented predator-prey interaction is scarce, there is at least one account of an American badger carrying off an adult ornate box turtle (*Terrapene ornata*) in its mouth (Legler 1960). In a study by Lloyd and Stadler (1998) in South Africa, honey badgers (*Mellivora capensis*) were indicated as predators of

TABLE 1.—Citations of American, European, and honey badgers as predators of turtles and tortoises. Only citations with direct evidence of badger predation or strong evidence of the possibility of badger predation are included. There are many references in which predation by badgers, especially on turtle or tortoise nests, is speculated or listed based only on the fact that the area is within the distribution of a certain species of badger.

Turtle/Tortoise Species	Badger Species	Citation	Comments
Hermann's Tortoise (*Testudo hermanni*)	European badger (*Meles meles*)	Swingland & Stubbs 1985	Documented nest predation in the south of France
		Bertolero et al. 2007	Documented predation on at least one adult in the western Mediterranean
European Pond Turtle (*Emys orbicularis*)	European badger (*Meles meles*)	Kahlke et al. 2015	Dates predation on adults to the Eemian interglacial period
		Mosimann & Cadi 2004	Documented predation upon nests and hatchlings in Switzerland
Pond Slider (*Trachemys scripta*)	European badger (*Meles meles*)	Mosimann & Cadi 2004	Predation upon nests and hatchlings of introduced red-eared sliders in Switzerland
Loggerhead Sea Turtle (*Caretta caretta*)	European badger (*Meles meles*)	Durmus et al. 2013	Badgers and foxes depredated 58.1% of eggs studied over one year on Dalyan Beach, Turkey
		Türkozan & Yilmaz 2008	Depredated 7% of eggs studied over one year on Dalyan Beach, Turkey
		Baskale & Kaska 2005	Five sea turtle nests on Dalyan Beach, Turkey depredated over the period of one year
Green Sea Turtle (*Chelonia mydas*)	European badger (*Meles meles*)	Yilmaz et al. 2015	Depredated 0.6% of green turtle nests over a period of six years on Akyatan Beach, Turkey
	Honey badger (*Mellivora capensis*)	West 2009	Depredated nests on multiple beaches in Tanzania
		West 2010	High levels of nest predation in the Temeke District of Tanzania
Tent Tortoise (*Psammobates tentorius*)	Honey badger (*Mellivora capensis*)	Lloyd & Stadler 1998	Strong evidence of predation on adults based on unique method of killing paired with tracks at fresh tortoise carcasses
Ornate Box Turtle (*Terrapene ornata*)	American badger (*Taxidea taxus*)	Legler 1960	Observed badger carrying off an adult turtle in its mouth
Painted Turtle (*Chrysemys picta*)	American badger (*Taxidea taxus*)	Platt et al. 2009	Observed badger excavating painted turtle nest in southwestern South Dakota
		Lampe 1982	Found evidence of *Chrysemys* eggs in badger scat in east central Minnesota
		Errington 1937	Found evidence of *Chrysemys* eggs in badger scat in northwestern Iowa
Agassiz's Desert Tortoise (*Gopherus agassizii)*	American badger (*Taxidea taxus*)	Emblidge et al. 2015	Strong evidence of predation on adults based on unique method of killing paired with camera trap documentation
		Turner & Berry 1984	Nests: Excavated by badgers in at least four cases Juveniles/ Adults: Deduced predation from indirect signs of badger presence/ abundance (e.g. scats, burrows)
Unidentified	American badger (*Taxidea taxus*)	Sovada et al. 1999	Found evidence of turtle eggs in badger gastrointestinal tract
		Lampe 1982	Found evidence of turtle eggs (*Chelydra* or *Chrysemys*) in badger scats and stomach contents

the tent tortoise (*Psammobates tentorius*) after discovering many tortoise carcasses killed in a unique and similar manner. Anterior plastrons were ripped away from the body, but there was no evidence of tooth marks on the removed portion or any other part of the shell - a characteristic similar to that found in both our and Emblidge et al.'s (2015) studies. This indicated the predator had the capability to pull apart a shell using only the strength and force from its forelimbs and without chewing or biting. Additionally, badger tracks were present near a fresh tortoise carcass killed in this manner. European badgers (*Meles meles*) were identified as predators of adult European pond turtles (*Emys orbicularis*) based on microstratigraphic fossil evidence from Germany. Similar to modern badgers, prehistoric badgers killed the turtle by biting the head and then opening the carcass from the posterior portion of the shell (Kahlke et al. 2015). These accounts, along with additional references detailed in Table 1, establish badgers from three genera as predators of all life stages of turtles. Our observation, coupled with the photographic evidence and report of Emblidge et al. (2015), suggest that modern badgers continue to be periodic predators of large adult desert tortoises. With so many tortoise deaths attributed to badgers in their study, Emblidge et al. (2015) noted that a single American badger has the potential for substantial impacts on *G. agassizii* populations. Similarly, Bertolero et al. (2007) found that a single event of European badger predation on an adult Hermann's tortoise in one year reduced survivorship of a population of reintroduced tortoises significantly in comparison to other years. Lloyd and Stadler (1998) also found substantial predation on tent tortoises (31 shells) attributed to honey badgers in South Africa which totaled 49% of all shells and carcasses found over a period of four years. Ironically, American badgers have also been documented to cohabitate with *G. agassizii* with no predatory behavior (Germano and Perry 2012), suggesting that tortoises are not a preferred prey item but may be killed opportunistically or during times of stress.

We strongly suspect, but cannot confirm, that the tortoise we observed was killed by an American badger based on three lines of supporting evidence: 1) the tortoise exhibited signs of mortality consistent with other reports of potential predation by a badger, 2) signs of badger excavations were abundant in the area, and 3) a badger was the only potential predator subsequently photographed by a trail camera in the vicinity of the dead tortoise. Although the evidence appears to indicate badger predation, we approach this conclusion with caution and note that further research on the enigmatic predator-prey relationship of the desert tortoise and the American badger is needed. If correct, our conclusion suggests that occasional badger predation of tortoises is more widespread than is generally appreciated, and our review of badger predation on turtles worldwide supports that conclusion. Understanding predator-prey relationships as well as the effects of climate change on these relationships is an important component in the conservation of the desert tortoise. Currently, there is a paucity of information on interactions between desert tortoises and badgers, indicating that the ecology of these predators and their potential effects on desert tortoise populations in the desert ecosystems of the southwestern United States is understudied.

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Absence of leopard sharks in catch surveys in Puget Sound, Washington

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The leopard shark (*Triakis semifasciata*) is an inshore species that is a prominent member of the elasmobranch fauna along the Pacific coast from Mazatlan, Mexico including the Gulf of California, to Oregon, USA (Ebert 2003). Within its historic range, this species is particularly common in California bays and estuaries including Elkhorn Slough, San Francisco Bay, Tomales Bay, and Humboldt Bay. Until 2009, this shark had not been officially known to occur north of Oregon. However, Farrer (2009) reported the capture of a single male specimen (133.4 cm TL) in Samish Bay in northeastern Puget Sound, Washington on 6 September 2007 by a commercial fisherman as a range extension. The range extension reported by Farrer (2009), while not confirmed through subsequent captures, has been cited in Nosal et al. (2014) and Barker et al. (2015).

As of 1 January 1994, the California Fish and Game Commission imposed a minimum size limit of 91.4 cm TL on commercial sale of leopard sharks (California Fish and Game Code, 1993, Ch. 2, Article 9, section 8388.5a,b). However, in 2006 and 2013, several individuals were charged with violating the federal Lacey Act (16 U.S.C. 3372a), which incorporates California law, for attempting to sell thousands of undersized leopard sharks (TL range: 21.6–44.4 cm), in some cases for hundreds of dollars each, from San Francisco Bay, California (Flaherty 2007, Haag 2013). Leopard shark pups were quite popular among aquarists as I witnessed throughout from the 1970s to the 1990s in several aquarium shops in the San Francisco Bay Area with prices ranging from $35 to $80 per pup. Given the historic popularity and availability of this species, at least up to 2013, it is possible that some leopard sharks remain in private aquariums (Smith and Horeczko 2008).

Since 2008, the Washington Department of Fish and Wildlife has conducted annual April to June bottom trawl surveys using a 400-mesh eastern bottom trawl net with a 3.2 cm cod-end liner, 10 cm cookie gear on the footrope and a mouth that opens 9.1 m to 13.7 m, depending on depth, in 51 locations spread throughout state waters of Puget Sound. Altogether from 2008 to 2016, 731 tows have been made at depths ranging from 5 to 125 fa. Based on 2014 data, trawls averaged about 11.0 minutes per trawl at a depth range from 6 to 115 fa with an average of 49.0 fa per trawl, and representing hundreds of hours of field time and analysis. More recently, elasmobranchs appeared in 40 (70.2%) out of 57 tows

made in 2014, 42 (76.4%) of the 55 tows made in 2015, and 46 (83.6%) of the 55 tows made in 2016 (J. Blaine, Washington Department Fish and Wildlife, unpublished data). The elasmobranchs captured during these three trawl seasons (*n*=958) included: 295 (30.8%) big skate (*Raja binoculata*), 303 (31.6%) longnose skate (*R. rhina*), 14 (1.5%) sandpaper skate (*Bathyraja kincaidii*), 345 (36.0%) spiny dogfish (*Squalus suckleyi*) (Ebert et al. 2010), and 1 (0.1%) brown catshark (*Apristurus brunneus*). The three species of skates and spiny dogfish have appeared regularly each trawl season. Sixgill sharks (*Hexanchus griseus*) have been caught prior to 2014, but not during this three-year period. During the nine-year period these data represent, no leopard sharks have been caught in Puget Sound by this method. (D. Lowry and J. Blaine, Washington Department Fish and Wildlife, personal communication).

Concurrently, I conducted a separate long-line study during the summer of 2014 to collect gill and heart parasites from elasmobranchs in Bellingham Bay within a 3–5 km radius of the coordinates 48° 41' N, 120° 30' W immediately to the north of Samish Bay, the area for the original, single leopard shark capture. In keeping with permit conditions, I used a 100 m long-line with 14/0 tuna hooks baited with squid set for 1 h to 1.5 h in water ranging from 18.3 m to 27.4 m deep, but with the long-line kept off the bottom by at least 3 m by interline floats. Six long-line events from May through September resulted in the capture of 49 spiny dogfish and no other elasmobranchs. By late September most spiny dogfish had migrated out of the area, as is typical in this region (McMillan 1999).

The extensive trawl data from the WDFW as well as data from the six long-line sets do not support the suggestion that leopard sharks have extended their range as proposed by Farrer (2009). There are possible explanations for the appearance of this lone leopard shark in eastern Puget Sound. While it is beyond the scope of this paper to prove the source, it is possible that a lone individual swam north from Oregon (Bates et al. 2014, Hight and Lowe 2007, Smith 2001) or this individual was the result of an aquarium release and not a range extension constituting a significant demographic unit shift for the species.

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Information for Contributors

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