

***EXAMINING LONG-RANGE TRANSPORT OF MONTROSE DDE VIA MARINE  
MAMMALS: EVALUATING RISKS TO CALIFORNIA CONDORS***

Final Report to the Montrose Settlements Restoration Program Trustee Council  
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**SUMMARY.**

**Background.** Dead marine mammals, particularly California sea lions (*Zalophus californianus*) beach-cast on the central California coast, are a potential source of food for reintroduced California condors (*Gymnogyps californianus*). California sea lions contain high concentrations of DDTs (i.e., sum of DDT compounds measured) and PCBs (i.e., sum of PCB congeners measured) (Kajiwara et al. 2001, Kannan et al. 2004, Blasius and Goodmanlowe 2009), thought to be due in part to their breeding on the Channel Islands, where the marine environment has been contaminated by the Montrose Chemical Corporation and other companies' industrial discharge of DDTs and PCBs between the late 1940's and early 1970's. A recent study by Burnett et al. (2013) suggested California condors are experiencing egg shell thinning, possibly due to exposure to DDE<sup>2</sup>, a major metabolite of the agricultural pesticide DDT, from feeding on beach-cast California sea lions. Yet, the extent to which Montrose's discharge of DDT is a source of DDE for condors is not known. For this study, published data were used in addition to new analyses of samples collected from marine mammals beach-cast on the central California coast and California condors to: 1) evaluate the ability to trace DDE in California sea lions to Montrose DDE using a DDTs/PCBs ratio and 2) evaluate the current risk from Montrose DDE to California condors that scavenge along the California coast.

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<sup>2</sup> DDE refers to *p,p'*-DDE

**Study Limitations.** Due to limited resources, the study's findings were based on organochlorine analysis of samples from a small number of California sea lions (n=12) and other marine mammals (n=5) stranded along the central California coast as well as published and unpublished marine mammal organochlorine data. A moderately small sample of California condors (n=33) from California were analyzed in this study for evaluation of toxicological risk from DDE exposure. Condor plasma DDE concentrations were used as an indicator of toxicological risk; assessing relationships between condor egg shell thickness and condor DDE concentrations (either plasma or egg) was beyond the scope of this study. The extent to which the samples analyzed for this study are representative of marine mammals that strand along the central California coast or condors in California is unknown but is an important factor to consider alongside this study's conclusions.

**Objective 1** *Evaluate the ability to trace DDE in California sea lions to Montrose DDE using a DDTs/PCBs ratio.* Based on the data evaluated for this study, both newly generated and published, the evidence does not support the use of the ratio of sum DDTs (or DDE)<sup>3</sup> to sum PCBs as a reliable indicator of Montrose-related contamination. This conclusion was based on four lines of evidence: 1) California sea lions sampled in Southern California (close to the point source for Montrose's discharge of contaminants) had similar DDTs/PCBs compared to California sea lions sampled in central or northern California, indicating that the DDTs/PCBs ratio in sea lions is not influenced by sampling location or distance from the point source of

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<sup>3</sup> As most papers reviewed reported DDTs (i.e., sum of the DDT compounds measured), DDTs were used when feasible but DDE (*p,p'*-DDE) was used if this was the only compound reported. DDE comprised the vast majority (~95%) of DDTs in marine mammal and condor samples analyzed as part of this report, thus exchanging DDTs for DDE did not have a significant impact on our reported findings; we report DDE/PCBs alongside DDTs/PCBs for data generated as part of this study. All tables and figures clearly state if DDTs/PCBs or DDE/PCBs were used for ratio comparisons.

Montrose contamination. 2) California sea lions have similar DDTs/PCBs to other marine mammals (e.g., harbor seal, Risso's dolphin) that have no evident link to Montrose contamination. 3) Ratios of DDTs/PCBs in sediment cores are highly variable within the Palos Verdes Shelf where Montrose contamination occurred. 4) Ratios of DDTs/PCBs are highly variable in species from California and Oregon, including species thought to be exposed to Montrose contamination, such as bald eagles (*Haliaeetus leucocephalus*) nesting on Catalina Island, and species not suspected of exposure to Montrose contamination, such as Orcas (*Orcinus orca*) in Oregon and prairie falcons (*Falco mexicanus*) in Central California.

Highly significant correlations ( $> 0.75$ ,  $p < 0.01$ , Spearman's correlation,  $n=12$ ) between multiple chlorinated pesticides, including some not associated with Montrose, suggest that California sea lions are exposed to well-mixed sources of organochlorine contamination that are enriched in DDTs, likely from multiple sources related to the widespread and historic use of DDT as an agricultural pesticide along the California coast. A potential source of DDTs has been documented near Elkhorn Slough, located within the Monterey Bay region. In the mid-1990's Caspian Terns (*Hydroprogne caspia*) breeding in Elkhorn Slough experienced DDE-associated reproductive failure with fledgling success dropping from ~80% to zero two years following a major flood event considered responsible for re-suspension of DDE from historic DDT use (Parkin 1998). Thus, although DDE concentrations in California sea lions almost surely reflect some exposure from Montrose-related contamination in the Southern California Bight, the proportion of Montrose DDE compared to other DDE sources (e.g., historic use of DDT in the Elkhorn Slough region) for sea lions stranded in central California is unknown.

***Objective 2) Evaluate the risk from Montrose DDE to California condors that scavenge along the California coast.*** Plasma DDE levels generated for this study indicate some condors in central California are exposed to potentially harmful levels of DDE via ingestion of marine mammals such as California sea lions. However, an extremely high degree of variation in DDE concentrations in California sea lion blubber samples was observed (0.1 – 67 µg/g, wet wt.) that introduces substantial uncertainty in correlating risk of exposure to DDE with feeding on California sea lions. This study also found elevated DDE concentrations in other marine mammals stranded along the central California coast, and thus multiple marine species are a potential source of DDE to California condors. As such, ~50% of California condors from central California evaluated (n=22) have plasma DDE concentrations that may be of toxicological concern, yet the degree to which a condor’s DDE exposure is from Montrose DDE is unknown. Further, to our knowledge, the population-level effects of DDE exposure on condors has not been investigated, yet a recent publication concluded that if lead poisoning was eliminated as a mortality source, the state-wide condor population in California would have positive growth based on current reproductive rates (Finkelstein et al. 2012).

***Data gaps and recommendations.***

***Objective 1) Evaluate the ability to trace DDE in California sea lions to Montrose DDE using a DDTs/PCBs ratio.*** Given the lack of evidence to support tracing Montrose DDE through the coastal California ecosystem based on DDTs/PCBs ratios, the ubiquitous historic use of DDT as an agricultural pesticide on the California coast (Kimbrough et al. 2008), and the wide-ranging behavior of individual California sea lions [e.g.; south to Baja and north to Alaska (Maniscalco et al. 2004, Weise et al. 2006)], estimating the proportion of Montrose-specific DDE in

California sea lions will require pursuit of additional research questions such as: i) What are the variables (e.g., foraging and migratory behavior, health status, age) that influence an individual's DDE exposure risk; ii) What proportion of DDE within the waters of the Southern California Bight can be attributed to Montrose discharges; iii) What proportion of DDE within a California sea lion comes from exposure while foraging within waters of the Southern California Bight.

*Objective 2) Evaluate the risk from Montrose DDE to California condors that scavenge along the California coast.* Preliminary data from this study suggest that between 4 and 50% of condors in central California may be exposed to DDE concentrations that could induce egg shell thinning; yet, estimating the proportion of Montrose DDE within a California condor is impeded by our inability to estimate the proportion of Montrose DDE within beach-cast mammals in condor habitat. To better understand the risk of DDE to condor reproductive success regardless of DDE source, future research questions could include: i) What is the relationship between a female's DDE plasma concentration and her chick's hatching success? ii) What is the maximum amount of DDE-associated reproductive impairment that condors can withstand and still maintain a self-sustaining population? iii) Does a condor's lead exposure history affect their reproductive success? California condors are lead poisoned on a regular basis and lead-related mortalities are preventing their recovery in the wild (Finkelstein et al. 2012). Yet, the effects of lead poisoning on condor reproductive success is unknown, even though lead is a well-established reproductive toxin (NRC 1993, Goyer 1996, Telisman et al. 2000). Further, the potential effects of lead on egg-shell thickness in condors is not clear; a study by Grandjean (1976) suggests lead exposure might be associated with reduced egg shell thickness in common kestrels (*Falco tinnunculus*) although a second study by Pattee (1984), in American kestrels (*Falco sparverius*), found no evidence of lead exposure on reproductive success.

## **INTRODUCTION.**

The vast majority (>95%) of the United States population (the Pacific Temperate stock) of California sea lions breed on the Channel Islands, located off the southern California coast (Aurioles and Trillmich 2013). Waters near the Channel Islands have been contaminated by PCBs and DDTs from the Montrose Chemical Corporation and other companies' industrial discharge of such magnitude that the area of discharge has been designated a superfund site (Palos Verdes, EPA #: CAD008242711) (Eganhouse and Pontolillo 2000). Thus, the high concentrations of DDTs and PCBs measured in California sea lion tissue compared to similar species such as elephant seals (Kajiwara et al. 2001, Kannan et al. 2004, Blasius and Goodmanlowe 2009) is not surprising. However, the potential to distinguish Montrose DDE from other DDE sources within California sea lions, known to travel large distances (Maniscalco et al. 2004, Weise et al. 2006), had not been established. In order to assess the ability to quantify the proportion of Montrose DDE within California sea lion tissues, this study's first objective was to examine the DDTs/PCBs ratio in California sea lion tissue samples using published and newly generated data to determine if this ratio is diagnostic of Montrose DDE as has been suggested previously (Le Boeuf et al. 2002).

California condors are known to forage on marine mammals, such as California sea lions, stranded along the central California coast (Chamberlain et al. 2005) and a recent study by Burnett et al. (2013) suggested that condors are experiencing DDE-induced egg shell thinning. However, to our knowledge, limited and non-published Ventana Wildlife Society data existed with respect to DDE concentrations in California condors. Further, the proportion of marine mammals in the diet of a condor that foraged along coastal California was also not known. In order to evaluate the risk from Montrose DDE to animals such as California condors that

scavenge along coastal California, the second goal of this study was to: i) evaluate organochlorine concentrations in California condor plasma samples to assess if condors were exposed to potentially harmful concentrations of DDE, ii) use carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) stable isotope analyses of condor and their marine and terrestrial mammal prey to quantify the amount of marine mammals in a California condor's diet, and iii) assess the potential to estimate a condor's exposure to Montrose DDE.

## **METHODS.**

**Study Area.** Samples from stranded marine mammals were collected in Monterey County California while samples from California condors were collected opportunistically by biologists associated with the California Condor Recovery Program in California. California condors released from Pinnacles National Park and Ventana Wildlife Society have been observed feeding on dead marine mammals along coastal central California (Burnett et al. 2013) and their ranges as identified via telemetry data overlap with the marine mammal sampling locations (Figure 1).

**Sample Collection. Organochlorines. *Marine mammals.*** Blubber samples from 12 California sea lions, one harbor seal (*Phoca vitulina*), one unidentified cetacean, one humpback whale (*Megaptera novaeangliae*), one Risso's dolphin (*Grampus griseus*), and one grey whale (*Eschrichtius robustus*) were collected from animals dead stranded along the central California coast between 2008-2012 by personnel affiliated with the Moss Landing Stranding Network (Table 1). Blubber samples (~5 grams) were wrapped in aluminum foil, placed in plastic Ziploc bags and stored at -20 °C until processing and analysis.

**California condors.** Blood samples (~4mLs) were collected opportunistically between 2009 and 2012 from 33 California condors in California (three pre-release, eight from southern California

and 22 from central California<sup>4</sup>) by field biologists<sup>5</sup> during routine trap-up and handling events. Blood was collected into Vacutainers with heparin and placed on ice (or 4°C) until processing. Within ~24 hours of blood collection, whole blood was centrifuged and plasma (~2mLs) was collected with kilned glass pipettes and placed in kilned glass vials. Plasma samples were then stored frozen at -20°C until processing and analysis.

Carbon and nitrogen stable isotopes. *Marine mammals.* Muscle samples collected opportunistically from marine mammals stranded along the central California coast were frozen at -20°C until processed and analyzed (Kurle et al. 2013).

*California condors.* ~100-200 µL subsamples from Vacutainers collected with EDTA were transferred into cryovials and frozen at -20°C until processed for isotope analysis according to previously published methods (Kurle et al. 2013).

***Organochlorine analysis.*** Marine mammal and condor samples were shipped frozen to the California Department of Fish and Wildlife Water Pollution Control Laboratory for organochlorine analysis. Twenty six chlorinated pesticides, including the seven metabolites of DDT and 54 PCB congeners were evaluated in each sample (see Appendix 1 for complete list of compounds measured).

***Carbon and nitrogen stable isotope analysis.*** Prey muscle and condor blood samples were freeze-dried, homogenized, and ~0.7 mg of each tissue was sealed into 5 x 9 mm tin capsules and analyzed for stable C and N isotope ratios using a Carlo Erba CE1108 elemental analyzer interfaced via a CONFLO III device to a Thermo-Electron Delta Plus XP mass spectrometer at the University of California Santa Cruz Department of Earth and Marine Sciences.

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<sup>4</sup> Southern California condors are not known to forage on the coast and thus do not have access to stranded marine mammals. Central California condors are known to forage on the coast and have been observed feeding on marine mammals (Burnett et al. 2013).

<sup>5</sup> Field biologists were affiliated with release sites managed by the U.S. Fish and Wildlife Service, Pinnacles National Park and the Ventana Wildlife Society.



## RESULTS AND DISCUSSION.

### **Objective 1) Evaluate the ability to trace DDE in California sea lions to Montrose DDE using a DDTs/PCBs ratio**

#### *i) Organochlorine concentrations in California sea lions.*

California sea lions stranded in central California had, on average, lower concentrations of DDTs and PCBs than samples collected from other marine mammals also stranded in central California, yet there was high variability among all marine mammal samples evaluated [Table 1, (DDTs (average  $\pm$  SD  $\mu\text{g/g}$  wet wt.): California sea lions  $12.6 \pm 18.9$  (range 0.1 to 67.5), other marine mammals  $20.8 \pm 18.4$  (range non-detectable<sup>6</sup> to 36.7), Figure 2; PCBs (average  $\pm$  SD  $\mu\text{g/g}$  wet wt.): California sea lions  $3.0 \pm 3.6$  (range 0.1 to 13.2); other marine mammals  $5.7 \pm 5.4$  (range non-detectable<sup>7</sup> to 10.9)]. Sum DDTs and sum PCBs were highly correlated ( $r > 0.95$ , Spearman's correlation) within individuals, a pattern we have observed in other high trophic level marine species such as albatross (Finkelstein et al. 2006). Sum DDTs comprised  $>90\%$  of the total chlorinated pesticides measured in all samples with the exception of the humpback whale with sum DDTs comprising 84% of total chlorinated pesticides. DDE was  $\sim 95\%$  of the sum DDTs. Similar to patterns observed in other studies on California sea lions (Bacon et al. 1992, Nino-Torres et al. 2009), PCB 153 was the most abundant congener measured, representing between 16 and 29% of the sum PCBs.

The extremely large ( $> 500$ -fold) variation observed in DDT concentrations (wet wt.) between individual California sea lions stranded along the California coast may in part be due to the sex, body condition, and age of an individual, as these factors are known to be important for predicting an individual's contaminant levels (Connolly and Glaser 2002, Hall et al. 2008,

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<sup>6</sup> grey whale blubber sample had non-detectable concentrations of DDTs

<sup>7</sup> grey whale blubber sample had non-detectable concentrations of PCBs

Blasius and Goodmanlowea 2009). The lipid content between individual California sea lion blubber samples was highly variable (% lipid 0.9 - 76, n=12), indicating that stranded animals reflected a high variation in body condition, as observed in previous studies on stranded California sea lions (Le Boeuf et al. 2002, Blasius and Goodmanlowe 2009). A negative relationship between lipid concentration ( $\mu\text{g/g}$ ) or % lipid and organochlorines ( $\mu\text{g/g}$ , lipid weight) in blubber samples has also been previously reported (Kajiwara et al. 2001, Blasius and Goodmanlowe 2009), yet we did not observe a significant relationship in our California sea lion blubber samples ( $r=0.399$ ,  $p > 0.20$ , Spearman's correlation for % lipid versus log DDTs ( $\mu\text{g/g}$  lipid),  $n=12$ , Figure 3). Large variation in lipid content in blubber samples has not been reported for non-stranded animals (DeLong et al. 1973, Nino-Torres et al. 2009). Thus, the extreme variation in lipid content within stranded animal blubber samples (> 80-fold) combined with the lack of a predicable relationship between % lipid and DDTs reported here impede the ability to accurately extrapolate data from stranded animals to live animals.

Organochlorines are known to accumulate in higher concentrations with increasing trophic level in the California current ecosystem (Jarman et al. 1996), therefore, sea lion foraging ecology and trophic position are potentially important factors driving variation in contaminant levels between individuals. Geographic foraging location has also been shown to be an important factor in contaminant accumulation. Studies on California sea lions in southern California waters demonstrate higher organochlorine concentrations than pinnipeds such as northern elephant seals who are transient in southern California and partition their time between California and Alaska (Kajiwara et al. 2001, Kannan et al. 2004). However, little information exists on how geographic separation within the California sea lion population might influence their organochlorine

exposure, particularly among different age classes and sexes that are known to vary in their yearly migrations away from their southern California breeding area.

***ii) Ratio of sum DDTs (DDTs) to sum PCBs (PCBs) in marine mammals and California condors as an indicator of Montrose-related contamination.***

A high ratio of DDE or DDTs to PCBs in marine animals that inhabit the waters off coastal California has been suggested as a way to trace DDE contamination to the Montrose-associated contaminant discharge of DDTs and PCBs in the Southern California Bight (Le Boeuf et al. 2002). However, this study found no evidence to support the use of the ratio of DDE or DDTs to total PCBs as diagnostic of Montrose-associated contamination and thus caution should be used to classify the proportion of Montrose-related contamination in wide-ranging marine animals.

**Marine mammals.**

Studies of California sea lions between 1991 and 2013 from southern, central, and northern California report a range of blubber tissue levels of DDTs or DDE<sup>8</sup> to PCBs that do not appear to be related to collection region (Figure 4, Table 2). A comparison of adult and juvenile males and females within our samples for which age and sex was known (Table 1) showed that males (n=3) had higher DDTs compared to females (n=3) (DDTs: 14 versus 1.4 µg/g wet weight, respectively), a finding that was previously observed (Kajiwara et al. 2001, Kannan et al. 2004, Blasius and Goodmanlowe 2009), and might be due to the ability of female marine mammals to excrete organic contaminants through their milk (Aguilar and Borrell 1994). Males (n=3) also had a marginally higher ratio of DDTs/PCBs than females (n=3) (p=0.077, Mann-Whitney U;

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<sup>8</sup> In studies that only reported DDE concentrations, DDE/PCBs was used for ratio comparisons (see Table 2).

DDTs/PCBs: 3.5 - 4.3 versus 1.7 – 3.5, respectively). California sea lion females and juvenile males remain closer to the Channel Islands during the non-breeding season whereas adult males can range as far north as Alaska (Maniscalco et al. 2004). Notably, the extremely small sample size of males and females evaluated here precludes drawing broad conclusions from this comparison.

The ratio of DDTs to PCBs for California sea lion blubber samples collected from central California (n=12, DDTs/PCBs: 1.6 to 5.1; DDE/PCBs: 1.5 to 5.1) were not different than the DDTs/PCBs ratio of the four other marine mammals sampled<sup>9</sup> (n=4, DDTs/PCBs: 3.3 to 4.6; DDE/PCBs: 3.0 to 4.4) ( $p > 0.2$ , Mann-Whitney U, Figure 5), suggesting that ratio patterns of DDTs to PCBs are not unique to California sea lions. In support of these findings, published data in orcas stranded along the Oregon coast illustrate that the DDE/PCBs ratio can vary widely across individuals (range = 0.9 to 7.7, n=5) (Hayteas and Duffield 2000). These data underscore the suggestion that a DDTs/PCBs ratio  $>1$  in wide-ranging marine mammals is indicative of exposure to elevated sources of coastal DDTs, such as that found along the California coast from historic use of DDT as an agricultural pesticide (Kimbrough et al. 2008, Parkin 1998), and cannot be attributed exclusively to Montrose contamination in the Southern California Bight. Black-footed (*Phoebastria nigripes*) and Laysan albatross (*Phoebastria immutabilis*), known to be pelagic species (Whittow 1993b, a) and thus not thought to be directly exposed to coastal sources of DDTs, have DDTs/PCBs ratios of  $< 1$  (~0.6) (Finkelstein et al. 2006).

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<sup>9</sup> One harbor seal, one unidentified cetacean, one humpback whale, and one Risso's dolphin (Table 1).

### **California condors.**

Organochlorine concentrations measured in plasma samples collected between 2009 and 2012 from California condors that are known to forage in central coastal California (Figure 1) exhibited a large range in DDTs/PCBs (1.2 to 6.8, DDE/PCBs: 1.1 to 6.7, n=16<sup>10</sup>). A comparison of the ratio of DDTs/PCBs in California condors with other studies that include soil from the Palos Verdes Shelf, marine mammals in California and Oregon, and prairie falcons from California, indicates that this ratio is highly variable with no indication of a south-north regional pattern (Figure 6). Thus, no evidence was found to support the use of a ratio of DDTs to PCBs in condor plasma samples to trace the source of a condor's DDE exposure to Montrose discharges.

The DDE/PCB153 ratio was also assessed for its applicability to be diagnostic of a DDE source. For central California condors with quantifiable concentrations of DDE and PCB153 (n=15), the DDE/PCB153 ratio ranged from 10 to 25 and was similar to the range observed for the DDE/PCB153 ratio in California sea lions in central California (9 to 22, n=12). The DDE/PCB153 ratio for other marine mammals in central California was less variable, yet fell in the middle of the range observed for condors and California sea lions (DDE/PCB153: 15 -18, n=4). The finding that the DDE/PCB153 ratios of condors, California sea lions, and other marine mammals overlap suggest this ratio might not be of diagnostic value to identify a single DDE exposure source. One factor that might be contributing to the large variation in the DDE/PCB153 ratios reported is that PCB153, although the most abundant congener measured, still only accounted for 16 and 29% of the sum PCBs measured in California sea lions. Another factor is the variety of potential sources of DDE to the California coastal marine environment relative to the attenuated sediment concentrations associated with Montrose (Kimbrough et al. 2008).

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<sup>10</sup> 22 central California condor plasma samples were analyzed but detectable concentrations of PCBs and DDTs were measured in 16 of the 22 samples.

*iii) Relationships between DDTs and other chlorinated pesticides that are not tied to Montrose releases indicate California sea lions are exposed to a well-mixed source of contamination.*

Correlations between sum DDTs and other chlorinated pesticide compounds were also evaluated to identify potential patterns of exposure source. Sum DDTs, trans-nonachlor, and  $\beta$ -hexachlorocyclohexane were highly correlated with each other ( $> 0.75$ ,  $p < 0.01$ , Spearman's correlation,  $n=12$ ) (Figure 7), suggesting that California sea lions are exposed to a well-mixed source of contamination. One could predict that if the majority of the DDTs came from Montrose and was thus at least partially uncoupled from other persistent pesticides used in California, there would not be a strong correlation between DDTs and trans-nonachlor and  $\beta$ -hexachlorocyclohexane, pesticides that were not discharged by Montrose Corporation. Rather, the finding that individuals with a high concentration of DDTs are also likely to have high concentrations of other persistent pesticides not of Montrose origin supports the observation from the ratio data that contaminant patterns in these animals are not indicative of a single source, but rather reflect complicated coastal processes most likely representing several well-mixed sources that have biomagnified through the food web.

**Objective 2) Evaluate the risk from Montrose DDE to California condors that scavenge along the California coast**

*i) Organochlorine concentrations and potential risk to condors from DDE exposure.*

Organochlorine concentrations were measured in plasma samples from 33 California condors as follows: three pre-release (background), eight condors from southern California that are not known to forage on the coast, and 22 condors from central California that are known to forage on the coast. Total organochlorine concentrations were non-detectable in pre-release condors;

southern California condors ranged from non-detectable to 87 ng/g (wet wt.), and central California condors ranged from non-detectable to 3200 ng/g (wet wt.). Similar to the findings in marine mammals, DDTs comprised >80% of the total chlorinated pesticides measured in central California condors with DDE accounting for >96% of the sum DDTs and PCB 153 was one of the most predominant congeners present, accounting for 10-48% of sum PCBs.

To evaluate potential risk to condors from DDE exposure, a conversion factor reported in Henny and Meeker (1981) was used to estimate condor egg DDE concentrations from birds with plasma levels >100 ng/g (wet wt.)<sup>11</sup>. A wide range of DDE toxicity thresholds for avian species has been reported with respect to reproductive impairment. For example, the threshold level of DDE for prairie falcon eggs is reported as 2,000 ng/g (wet wt.), while the threshold for peregrine falcons (*Falco peregrinus*) is ~7-fold greater (15000 – 20000 ng/g wet wt.) (Fyfe et al. 1976, 1988). Although the DDE-induced egg shell thinning threshold for condors is unknown, compared to other avian species, between 4 and 50% of coastal condors might be suffering from ≥ 10% egg shell thinning (Figure 8). The extent to which DDE is impairing condor reproductive success is unknown.

***ii) Proportion of marine mammals in the diet of California condors.***

$\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values were determined in condor blood and their prey to estimate the proportion of marine mammals in the diet of California condors. The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values from California condors in central California that are known to forage on the coast were significantly higher than those from southern California condors that are considered to be exclusive terrestrial feeders (Figure 9). The higher  $\delta^{15}\text{N}$  values are indicative of a diet composed of higher trophic level prey

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<sup>11</sup> As Henny and Meeker (1981) only used plasma samples with DDE >100 ng/g wet wt. to calculate the equation: DDE in egg contents = 6.243\*(plasma DDE)<sup>1.033</sup>, we also only used plasma samples with DDE >100 ng/g wet wt. when using this equation.

which would be expected from condors foraging on carnivorous sea lions versus those eating herbivorous terrestrial grazers. The higher  $\delta^{13}\text{C}$  values are indicative of a diet composed of greater amounts of marine derived carbon as the primary production at the base of marine food webs has considerably higher  $\delta^{13}\text{C}$  values than plants at the base of a terrestrial food web.

Bayesian stable isotope mixing models incorporating stable isotope discrimination factors for condors (Kurlle et al. 2013) and stable isotope values from condors, marine mammals, and terrestrial mammals indicated that an average of ~27% of the diet from central California condors could be attributed to a marine-source (e.g., California sea lions, harbor seals). However, when evaluating each condor individually, the mean proportion of marine mammals within a condor's diet was highly variable (~4-50%), which likely contributes to the high degree of variability within the condor plasma DDE concentrations observed here (non-detectable to 2700 ng/g wet wt.). Model constraints precluded the ability to determine the degree to which condors were ingesting individual marine mammal species as their stable isotope values were too similar to be treated as distinct entities within the model. Ventana Wildlife Society biologists recorded observations of California condor non-proffered feeding behavior between 1999 and 2013. Out of ~230 observation days between 1999-2013 where condors were observed feeding on marine mammals, biologists documented California condors feeding on California sea lions ~70% of the time, other marine mammals (e.g., grey whale, harbor seal, etc.) 20% of the time, and unidentified pinnipeds ~10% of the time (Burnett et al. 2013, J. Burnett, pers. comm.). A study in which these authors are collaborators on is underway to evaluate how a condor's behavior (e.g., % time observed on the coast) as well as demographic variables (e.g., age, breeding status) may influence the proportion of marine mammals within an individual condor's diet as well as



hatching success. Preliminary analyses suggest that the proportion of time a female condor spends on the coast may be related to the hatching success of that condor's egg.

***iii) Proportion of Montrose DDE within a California condor.***

Similar to California sea lions, the proportion of Montrose DDE compared to other DDE sources for California condors is unknown. Connolly and Glaser (2002) concluded that some female California sea lions from San Miguel Island had DDE concentrations that could only be obtained from consuming highly contaminated fish associated with the contaminants discharged by Montrose and other corporations to the Palos Verdes Shelf. However, other marine mammals in central California have comparable DDE concentrations to California sea lions stranded in central California (Figure 2) and so the conclusion that condors could only be obtaining their observed DDE concentrations from feeding on California sea lions is not supported by the data presented in this study. Nonetheless, given that all California sea lions along the west coast spend some part of their lives foraging in southern California waters during their breeding season, DDE concentrations in California condors eating California sea lions most likely contain some proportion of Montrose related DDE.

As no evidence was found to support the use of the ratio of DDTs/PCBs as diagnostic of Montrose DDE, and the concentrations of DDE in California sea lions are comparable to other marine mammals (Figure 2), estimating the proportion of Montrose DDE that a condor is exposed to will require quantifying many factors such as: i) the proportion of marine mammals a condor feeds on; ii) the proportion of California sea lions ingested compared to other marine mammals (e.g., grey whales, Risso's dolphins, harbor seals); iii) the variation in DDE concentrations within marine mammal tissue that condors ingest; and iv) the proportion of

Montrose DDE within marine mammal tissues that condors ingest. Given the extreme variation observed for some of these factors evaluated for this study (e.g., proportion of marine mammals a condor feeds upon, DDE concentrations within marine mammals and condors), performing more sophisticated analyses to estimate the proportion of Montrose DDE exposure a condor receives over any specified time frame would not be appropriate with the current available data.

### **ACKNOWLEDGMENTS.**

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**Table 1:** Detailed information for blubber samples collected from 17 marine mammals stranded dead along the central California coast and analyzed for organochlorine concentrations (Appendix 1). CASL = California sea lion, RIDO = Risso's dolphin, HASE= harbor seal, HUWH = humpback whale, GRWH= grey whale, UNK = unknown, JUV = juvenile, ND = values below limit of quantification.

<u>Species</u>	<u>ID</u>	<u>Date collected</u>	<u>Age</u>	<u>Sex</u>	<u>Condors observed feeding?</u>	<u>DDTs (µg/g wet wt.)<sup>12</sup></u>	<u>PCBs (µg/g wet wt.)<sup>13</sup></u>
CASL	P2	6/18/2010	UNK	UNK	NO	11	2.6
CASL	P5	6/22/2009	UNK	UNK	NO	0.1	0.1
CASL	P6	12/29/2008	UNK	UNK	YES	4.6	1.4
CASL	P9	UNK	JUV	UNK	NO	6.2	1.9
CASL	P10	UNK	JUV	UNK	NO	4.4	1.6
CASL	P12	6/2/2009	Adult	F	YES	0.2	0.1
CASL	2507	3/28/2012	Adult	F	NO	6.3	1.7
CASL	2532	6/22/2012	Adult	F	NO	1.7	0.8
CASL	2484	10/19/2011	Adult	M	NO	8.8	2.3
CASL	2460	8/9/2011	Adult	M	NO	12	3.2
CASL	2437	5/21/2011	JUV	M	NO	29	6.5
CASL	2523	5/25/2012	PUP	M	NO	68	13
RIDO	CET1	5/5/2011	UNK	UNK	YES	36	11
HASE	P8	UNK	JUV	UNK	NO	10	2.2
HUWH	CET3	8/14/2011	JUV	UNK	NO	0.4	0.1
DOLPHIN-UNK	CET 2	5/26/2011	UNK	M	NO	37	9.6
GRWH	160	5/23/2012	UNK	F	YES	ND	ND

<sup>12</sup> Sum of seven DDT metabolites measured (Appendix 1).

<sup>13</sup> Sum of 54 PCB congeners measured (Appendix 1).



**Table 2.** Description of studies shown in Figure 4 comparing the ratio of DDE or sum DDTs to PCBs in tissues from California sea lions collected in Sothern California, Central California, and Northern California between 1991 and 2012.

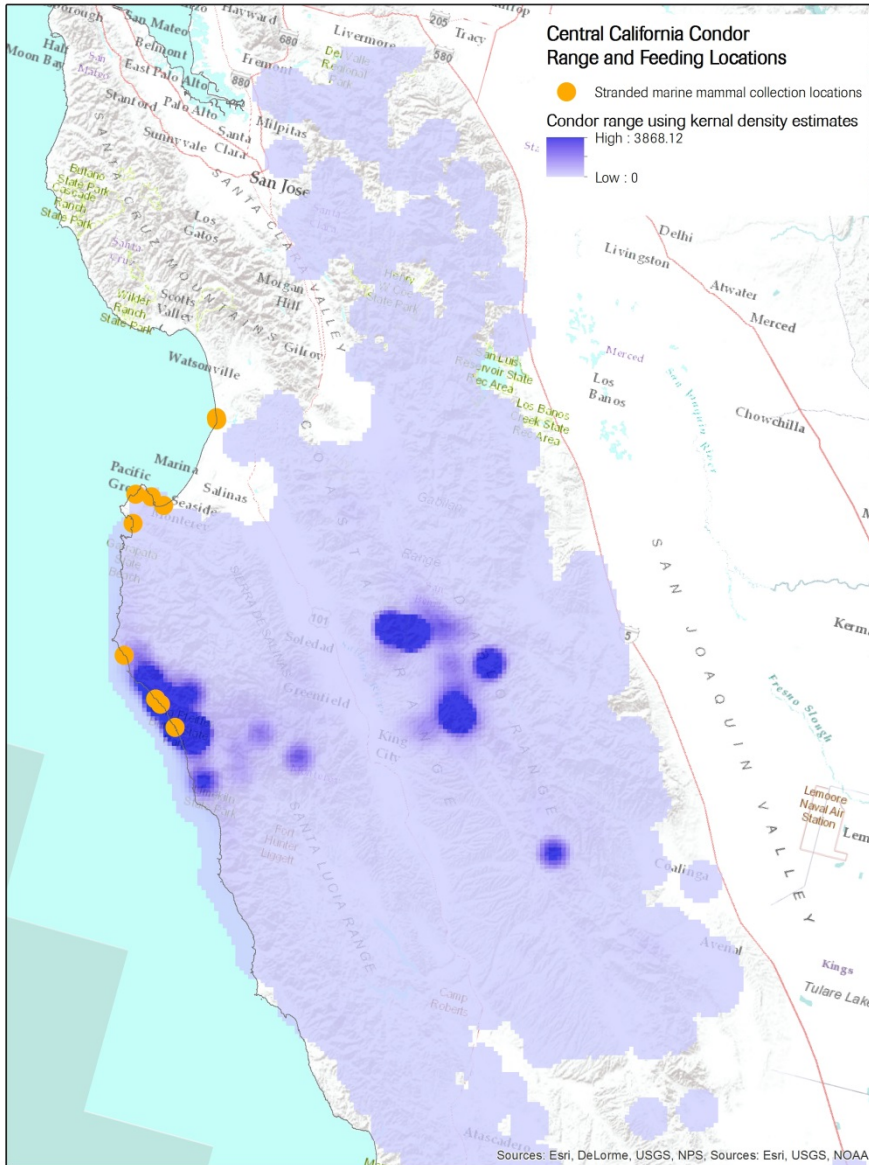
<u>study</u>	<u>region</u>	<u>n</u>	<u>tissue, units</u>	<u>PCBs</u>	<u>DDTs or p,p' DDE</u>	<u>ratio</u>
Le Boeuf et al. 2002	South CA	7	blubber, µg/g, lipid wt	12, 18 ± 16 (geomean, mean ± SD )	33, 61 ± 62 (geomean, mean ± SD )	2.8 [geomean (DDTs/PCBs)]
Blasius and Goodmanlowe 2009	South CA	92	blubber, µg/g, lipid wt	87 ± 263 (mean ± SD )	594 ± 1668 (mean ± SD )	6.9 (mean DDTs/mean PCBs)
Costa et al. 1994 <sup>14</sup>	South CA	10	blubber, ppm, fat	3 (mean)	10 (mean)	3.3 (mean DDE/mean PCBs)
Kajiwara et al. 2001	Central CA	13	blubber, µg/g, lipid wt	117, 428 ± 523 (geomean, mean ± SD )	209, 671 ± 1040 (geomean, mean ± SD )	1.7 [geomean (DDTs/PCBs)]
Debier et al. 2005	Central CA	12	plasma, ng/g, wet wt	26 ± 24 (mean ± SD )	47 ± 31 (mean ± SD )	1.8 (mean DDTs/mean PCBs)
Le Boeuf et al. 2002	Central CA	24	blubber, µg/g, lipid wt	26, 54 ± 93 (geomean, mean ± SD )	80, 170 ± 307 (geomean, mean ± SD )	3.0 [geomean (DDTs/PCBs)]
this study	Central CA	12	blubber, µg/g, wet wt	1.5, 3.0 ± 3.6 (geomean, mean ± SD )	4.7, 12.6 ± 18.9 (geomean, mean ± SD )	3.0 [geomean (DDTs/PCBs)]
Ylitalo 2005 <sup>15</sup>	Central CA	16	blubber, µg/g, wet wt	20 ± 18 (mean ± SD )	76 ± 62 (mean ± SD )	3.8 (mean DDTs/mean PCBs)
Gundersen et al. 2013	North CA	8	blubber, µg/g, lipid wt	4.2, 6.5 ± 7.8 (geomean, mean ± SD)	7, 11.2 ± 12.9 (geomean, mean ± SD)	1.7 [geomean (DDTs/PCBs)]
Yurok Tribe <sup>16</sup>	North CA	24	blubber, µg/g, wet wt	4.0, 5.1 ± 3.7 (geomean, mean ± SD)	14, 18 ± 15 (geomean, mean ± SD)	3.4 [geomean (DDTs/PCBs)]
Le Boeuf et al. 2002	North CA	5	blubber, µg/g, lipid wt	31, 36 ± 23 (geomean, mean ± SD)	99, 127 ± 103 (geomean, mean ± SD)	3.2 [geomean (DDTs/PCBs)]

<sup>14</sup> Costa, H., Wade, T., Bailey, A., 1994. Analytical chemistry data report for the Southern California Natural Resources Damage Assessment as cited in Connolly and Glaser (2002)

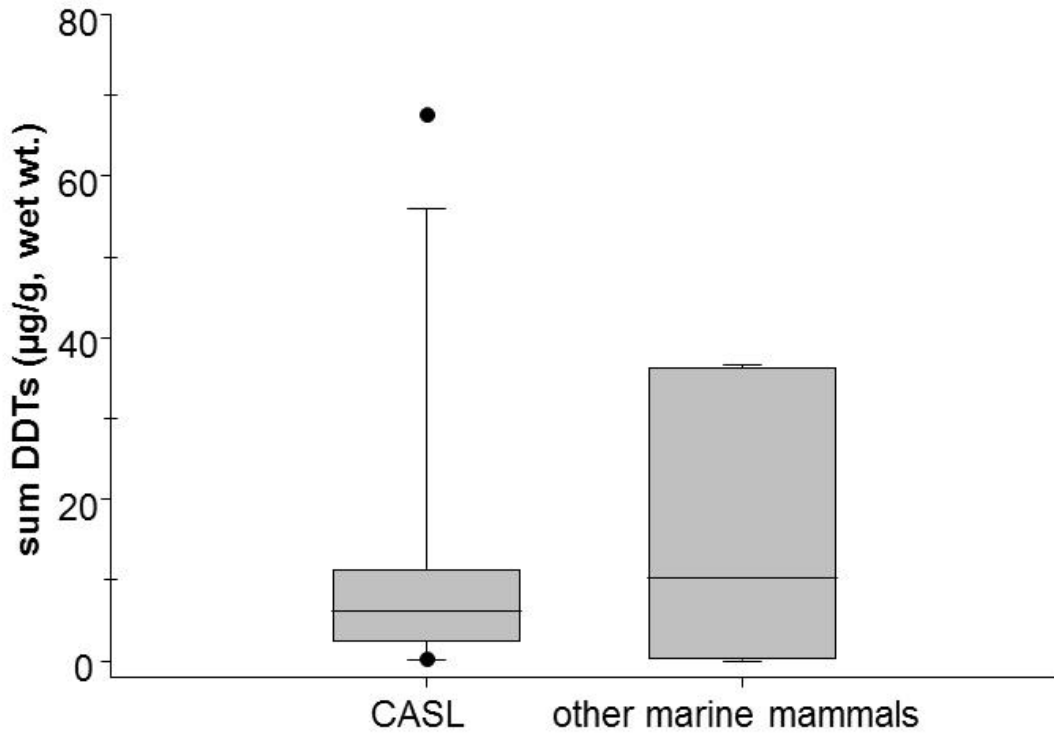
<sup>15</sup> Only show values for males (n=16) as mean DDTs/mean PCBs for females is the same (3.8)

<sup>16</sup> Unpublished data from Chris West, Yurok Tribe

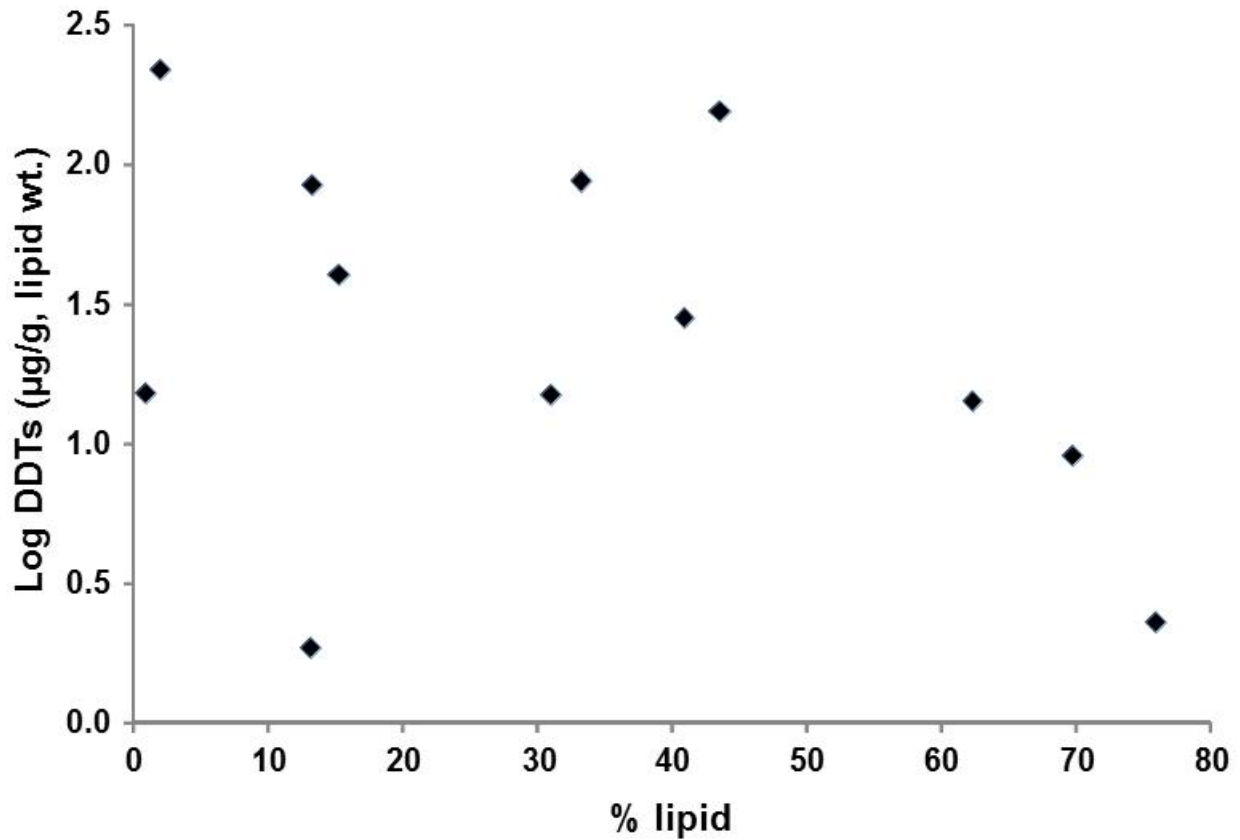
**Figure 1.** Marine mammal samples were collected from dead stranded animals within Monterey County between 2008 and 2012 with orange dots representing known collection locations. Blue shaded area represents California condor range identified via telemetry data. California condor blood samples were collected between 2009 and 2012 and, as illustrated by their telemetry data, overlap in range with the marine mammal sampling locations.



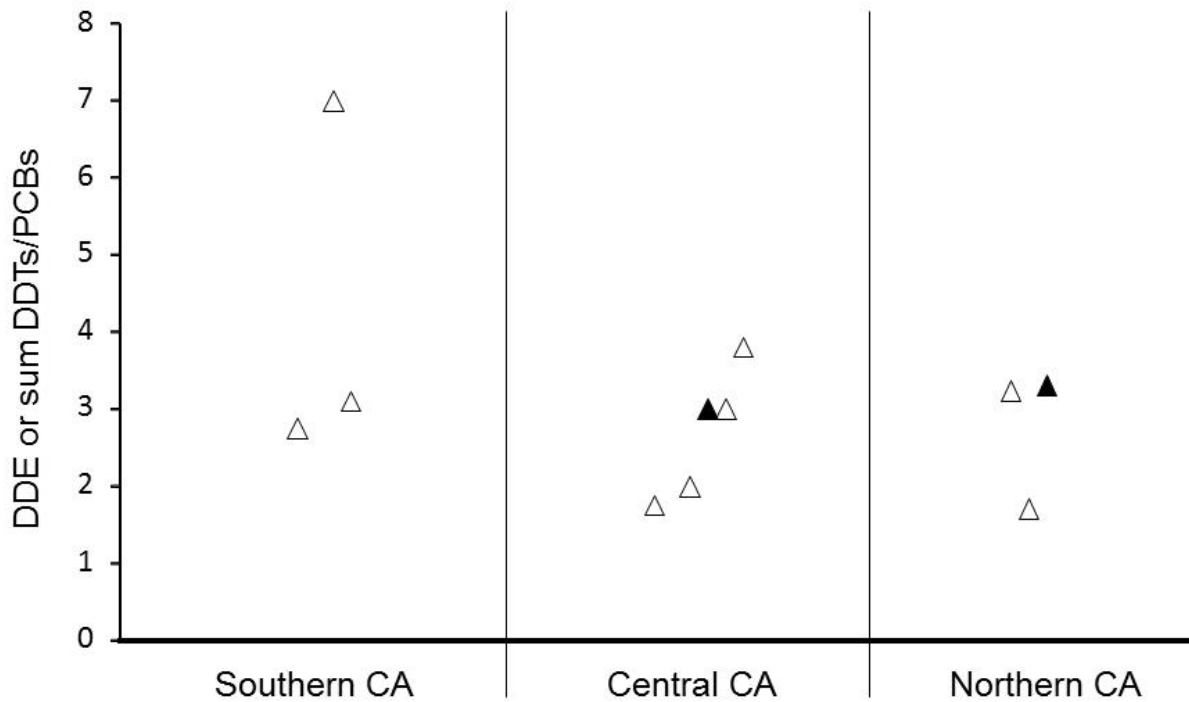
**Figure 2.** Sum DDTs in California sea lion blubber samples collected from dead animals stranded along the central California coast (CASL, n=12) compared to sum DDTs in blubber samples from other marine mammals dead stranded along the central California coast (Risso's dolphin, harbor seal, humpback whale, grey whale, and unidentified dolphin, n=5). Box indicates median, upper and lower 75<sup>th</sup> and 25<sup>th</sup> percentiles, whiskers represent 10<sup>th</sup> and 90<sup>th</sup> percentiles.



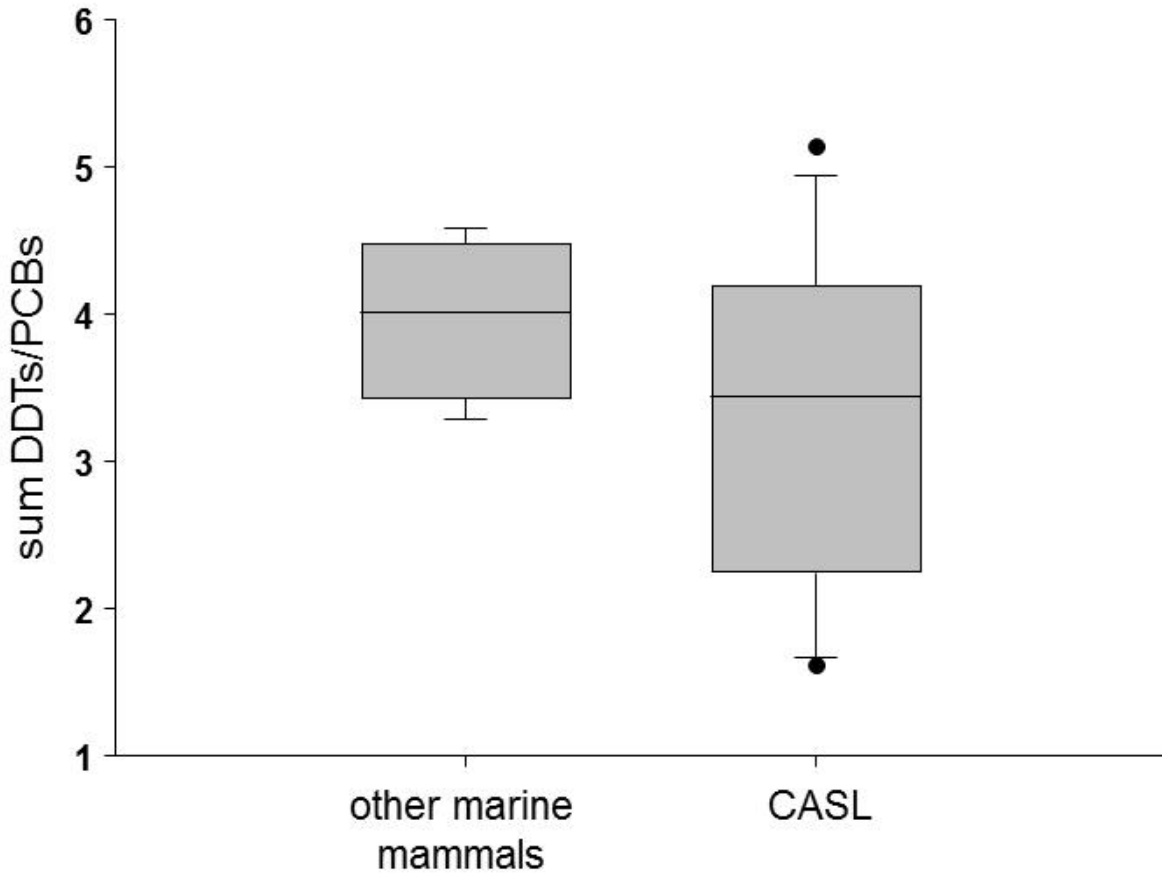
**Figure 3.** No significant relationship was found between sum DDTs (log,  $\mu\text{g/g}$  lipid wt.) and % lipid in blubber samples collected from Californian sea lions dead stranded along the central California coast ( $r=0.399$   $p > 0.20$ , Spearman's correlation,  $n=12$ ). However, a slight negative trend in the correlation suggests a larger sample size might help elucidate the nature of this relationship.



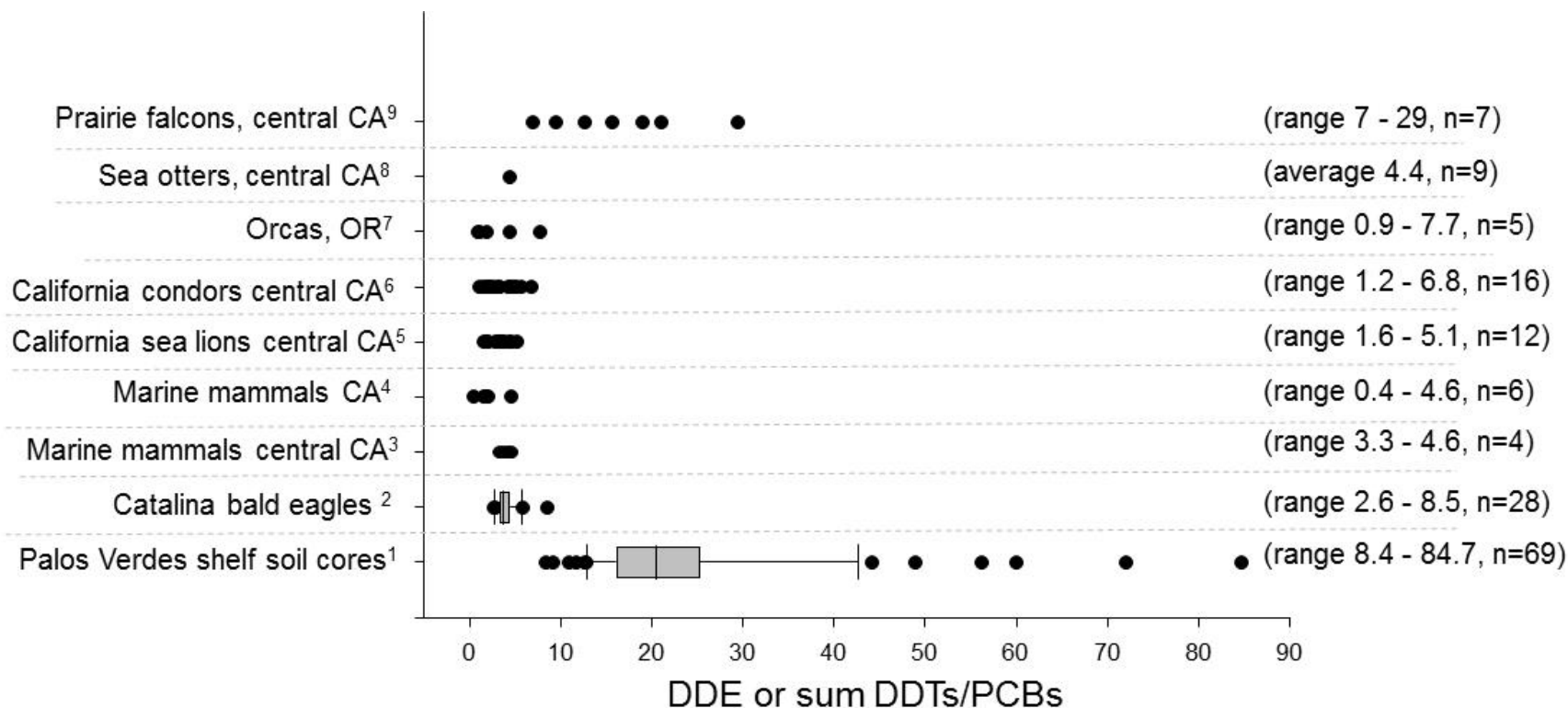
**Figure 4.** Ratio of DDTs (or DDE) to PCBs from studies on California sea lions collected in Southern California, Central California, and Northern California from 1991-2012. Filled squares represent unpublished data. See Table 2 for descriptions of studies representing each symbol. Southern California samples collected between San Diego and San Luis Obispo; Central California samples collected between San Luis Obispo and San Francisco; Northern California samples collected from Marin to Humboldt County.



**Figure 5.** Ratio of DDTs to PCBs in blubber samples collected from California sea lions (CASL, n=12) and other marine mammal (n=4, one harbor seal, one unidentified cetacean, one humpback whale, and one Risso's dolphin) in central California. DDTs/PCBs were not different between groups ( $p > 0.2$ , Mann-Whitney U). Box plot indicates median, upper and lower 75<sup>th</sup> and 25<sup>th</sup> percentiles, whiskers represent 10<sup>th</sup> and 90<sup>th</sup> percentiles.



**Figure 6.** Comparison of DDE or sum DDTs to PCBs from multiple studies do not support the use of the DDE to PCBs ratio as diagnostic of Montrose contamination but rather suggests that an enriched signature of DDTs to PCBs indicates exposure to elevated sources of DDTs, most likely including the widespread, historic use of DDT as an agricultural pesticide.



## Figure 6. Footnotes.

<sup>1</sup> Average total DDTs (normalized to carbon)/average total PCBs (normalized to carbon) from soil cores, EPA report, “Final Data Report for the Fall 2009 Sediment Sampling Program”, November 2013, data shown as box plot

<sup>2</sup> DDE/total PCBs from bald eagles on Catalina Island 1989 to 1998, D. K. Garcelon, unpublished data, data shown as box plot

<sup>3</sup> Total DDTs/total PCBs in blubber samples from four marine mammals (one harbor seal, one unidentified cetacean, one humpback whale, one Risso’s dolphin) collected from animals stranded in central California between 2009 and 2011 for this study

<sup>4</sup> Total DDTs/total PCBs in blubber samples from marine mammals (two northern elephant seals, one sea otter, one humpback whale, one grey whale, and one harbor seal) stranded along the California coast (from Rio Del Mar to Año Nuevo) from Kannan et al.(2004)

<sup>5</sup> Total DDTs/total PCBs in California sea lion blubber samples collected from sea lions stranded along the central California coast between 2008 and 2012 for this study

<sup>6</sup> Total DDTs/total PCBs in California condor plasma samples from central California collected between 2009 and 2012 for this study. Condor samples with detectable levels of DDTs and PCBs included for comparison of ratio data (n=16 out of 22 central California condor plasma samples analyzed)

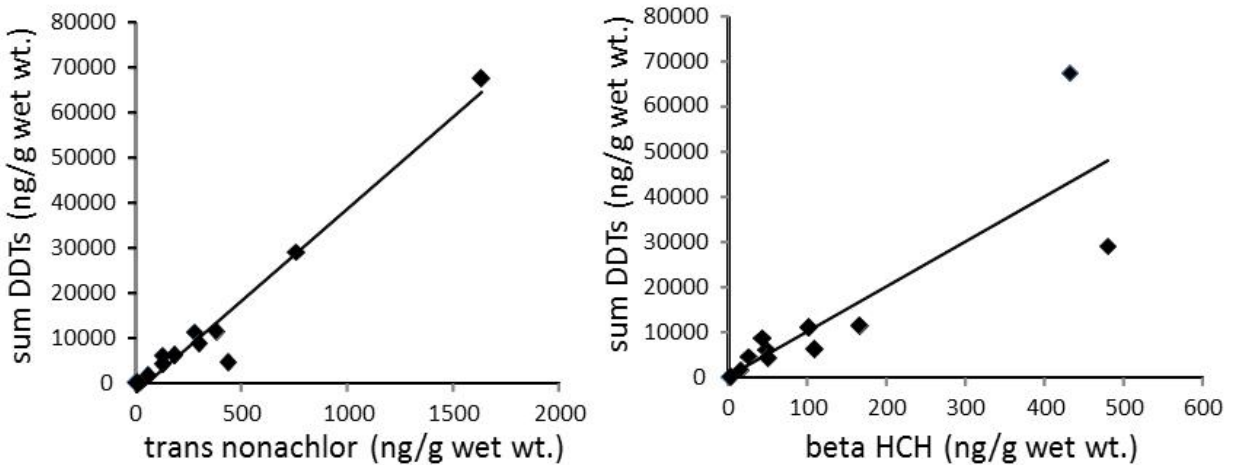
<sup>7</sup> DDE/total PCBs in killer whales stranded along the Oregon coast between 1988 and 1997 from Hayteas et al. (2000)

<sup>8</sup> Average DDE/average total PCBs for California sea otter liver samples collected in central California (e.g., Elkhorn Slough) between 1988 and 1991 from Bacon et al. (1999)

<sup>9</sup> DDE/total PCBs for prairie falcon eggs collected from 1989 and 1991 from Jarman et al (1996). Note that each data point graphed represents seven locations that reflect between one to six eggs per location. If a location represents >1 egg, average DDE/average PCBs is shown

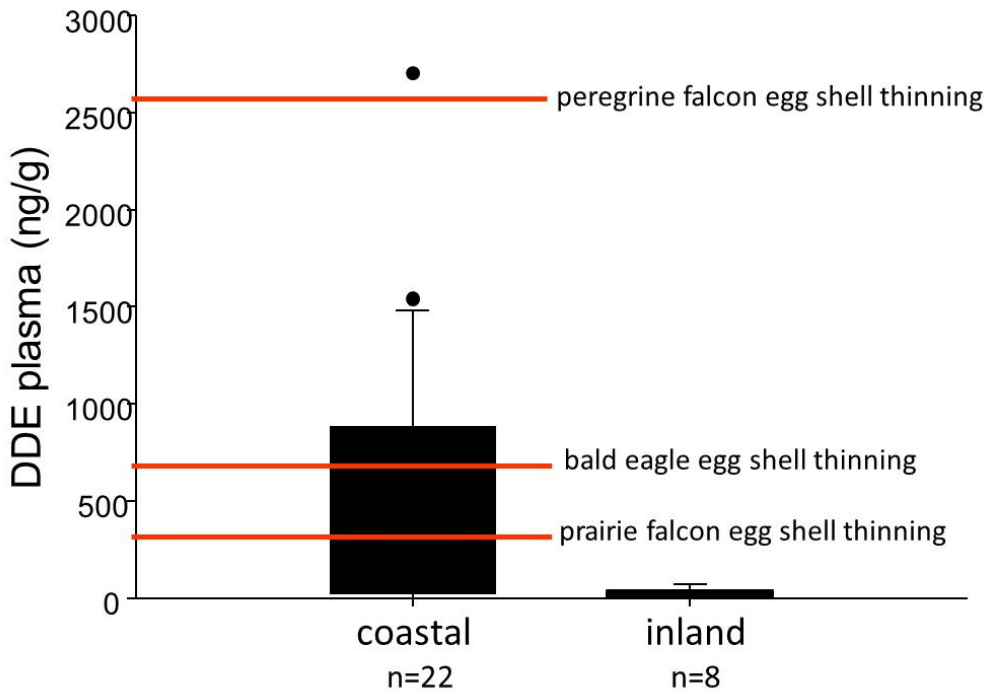


**Figure 7.** Correlations between concentrations of sum DDTs and trans-nonachlor, sum DDTs and  $\beta$ -hexachlorocyclohexane in blubber samples collected from California sea lions (n=12) stranded along the central California coast. Correlations between all three pesticide compounds were highly significant ( $r>0.75$ ,  $p<0.01$ , Spearman's correlation), demonstrating that if a sea lion had high concentrations of DDTs, they were very likely to have high concentrations of trans-nonachlor and  $\beta$ -hexachlorocyclohexane, which are not major components of releases by Montrose, and supports the suggestion that these sea lions were exposed to a well-mixed source of chlorinated pesticides.



Spearman's correlation	beta HCH	sum DDTS	trans nonachlor
beta HCH	1.00		
sum DDTS	0.909	1.00	
trans nonachlor	0.767	0.883	1.00

**Figure 8.** Central California condors (i.e., condors that forage on the coast, coastal) have ~ 20-fold higher plasma DDE concentrations than Southern California condors (i.e., condors that are not known to forage on the coast, inland). Although the toxicity threshold for DDE-induced egg shell thinning in condors is unknown, if comparing to other avian species<sup>17</sup> such as prairie falcons<sup>18</sup>, bald eagles<sup>19</sup>, and peregrine falcons<sup>20</sup>, between 4 and 50% of coastal condors might be suffering from  $\geq 10\%$  egg shell thinning.



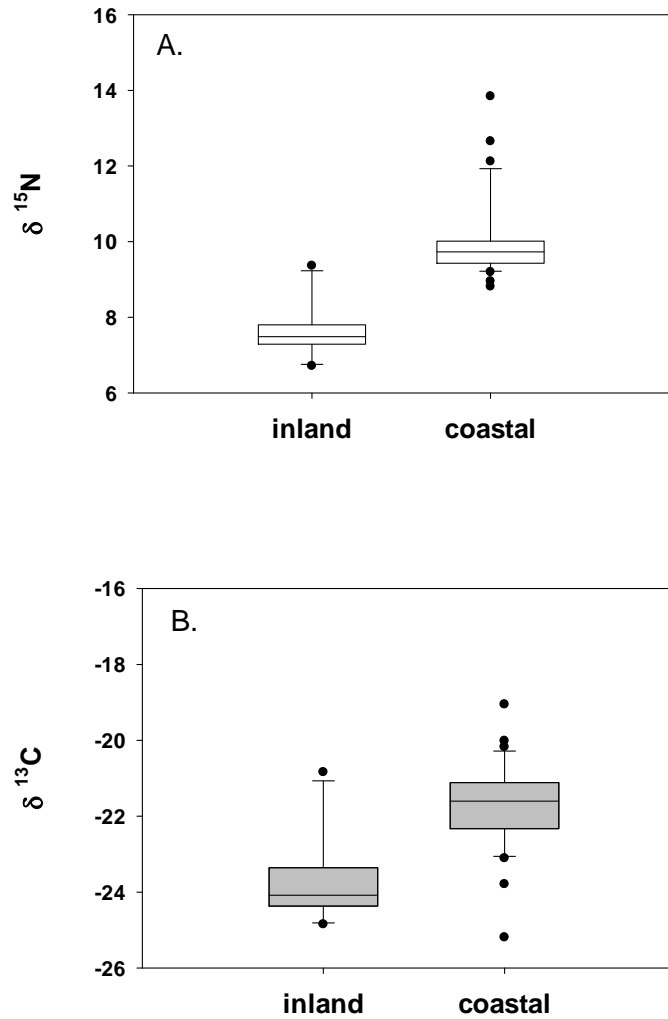
<sup>17</sup> Following Henny and Meeker (1981), estimates to convert DDE in condor plasma to egg DDE values were calculated using the following equation: DDE in egg contents =  $6.243 \times (\text{plasma DDE})^{1.033}$ , only plasma values  $>100$  ng/g were used for estimating egg DDE concentrations.

<sup>18</sup> The threshold level of DDE for prairie falcon eggs is reported as 2,000 ng/g (wet wt.) (Fyfe et al. 1976, 1988).

<sup>19</sup> For bald eagles, 10% egg shell thinning was associated with egg DDE concentrations of 5000 ng/g (Wiemeyer et al. 1984).

<sup>20</sup> The threshold level of DDE for peregrine falcon eggs is reported as 15000 – 20000 ng/g wet wt. (Fyfe et al. 1988).

**Figure 9.** The median A)  $\delta^{15}\text{N}$  and B)  $\delta^{13}\text{C}$  values (reported in ‰) from California condors from central California known to forage on the coast (coastal, n=31) and from southern California known to only forage inland (inland, n=10). Higher  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values in coastal condors indicate they are feeding at higher trophic levels and on more marine-derived prey, respectively, than inland condors. Box plot indicates median, upper and lower 75<sup>th</sup> and 25<sup>th</sup> percentiles, whiskers represent 10<sup>th</sup> and 90<sup>th</sup> percentiles.



**Appendix 1.** Full list of organochlorines measured in marine mammal and California condor samples by the California Department of Fish and Wildlife Water Pollution Control Laboratory.

<u>OCH Pesticides</u>	<u>PCBs</u>	<u>PCBs, continued</u>
aldrin	8	126
chlordane, cis	18	128
chlordane, trans	27	137
dacthal	28	138
DDD, o,p'	29	141
DDD, p,p'	31	146
DDE, o,p'	33	149
DDE, p,p'	44	151
DDMU, p,p'	49	153
DDT, o,p'	52	156
DDT, p,p'	56	157
dieldrin	60	158
endosulfan I	64	169
endrin	66	170
HCH, alpha	70	174
HCH, beta	74	177
HCH, gamma	77	180
heptachlor	87	183 <sup>a</sup>
heptachlor epoxide	95	187
hexachlorobenzene	97	189
methoxychlor	99	194
mirex	101	195
nonachlor, cis	105	198_199
nonachlor, trans	110	200
oxadiazon	114	201
oxychlordane	118	203
		206
		209

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<sup>a</sup> Due to instrument changes, PCB 183 was not analyzed on all samples but accounted for <3% of total PCBs reported.