



Persistent organic pollutants in the olive ridley turtle (*Lepidochelys olivacea*) during the nesting stage in the “La Escobilla” Sanctuary, Oaxaca, Mexico

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Abstract

Persistent organic pollutants (POPs) are chemical substances widely distributed in the environment by the runoff from anthropic activities and can be distributed and bioaccumulated or biomagnified in the environment, affecting the health of organisms. The sea turtle, *Lepidochelys olivacea*, is a long-lived organism, with migratory habits and feeding behaviors that allow exposure to various pollutants. This work aimed to determine long-term exposure to POPs in adult olive ridley turtles (*L. olivacea*), sampled during the nesting season, in “La Escobilla” Sanctuary. Blood samples were collected and processed to obtain plasma. The quantification of POPs in blood was carried out with an extraction technique with a focused ultrasound probe. Twenty-seven POP analytes were determined. The concentrations of hexachlorocyclohexane, endosulfan isomers, dichlorodiphenyltrichloroethane, total polychlorinated biphenyls, and the total sum of POPs found in plasma are higher than those reported in other studies, which reported effects such as hematological and biochemical changes in blood, changes in immune system cells and enzymatic activity related to oxidative stress. These results are important to demonstrate the chronic exposure to POPs in olive ridley turtles in marine ecosystems and to highlight the importance of assessing the associated health risks, considering that these contaminants could be transferred to the offspring and affect future generations of this reptile. It is important to carry out studies that develop conservation strategies for the olive ridley turtle. Also, it is necessary to control the emissions of pollutants into the atmosphere, as well as reduce urban, agricultural, and industrial waste in the environment and marine ecosystems.

Keywords Reptiles · Biomarkers · Sea turtle · Biomonitoring · Ecotoxicology · Pollution

Introduction

Pollution is a historical problem derived from the increase in anthropogenic activities in response to the growth of human populations, resulting in an imbalance in ecosystems (del

Puerto-Rodríguez et al. 2014). Anthropogenic activities such as agriculture and livestock have increased and, in turn, the use of chemical substances meant to control pests and disease-transmitting vectors (El-Shahawi et al. 2010; García-Hernández et al. 2018). Furthermore, industrial activities and the growth of urban areas have led to an increase in waste, contaminated with a wide variety of chemical substances, including POPs (Secretaría del Convenio de Estocolmo 2020).

POPs, also known as first-generation pesticides, are highly persistent due to their physicochemical properties (Secretaría del Convenio de Estocolmo 2020; D’Illo et al. 2011), which include organochlorine pesticides (OP), polychlorinated biphenyls (PCBs), and polycyclic aromatic hydrocarbons (PAHs) (Clukey et al. 2018). POPs are volatile substances, which allows their distribution in the air to other areas of the planet, as well as wet deposition and further distribution through ocean currents (Amdany et al. 2014; Hu

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et al. 2021), to finally be deposited in the sediments (Hong et al. 2016; Ge et al. 2021) and soils (Ortiz-Hernández et al. 2014; Ukalska-Jaruga et al. 2020). They are lipophilic substances that tend to accumulate in fatty tissues and, in some cases, can be biomagnified and distributed through food chains (D’lilio et al. 2011; Perrault et al. 2017; Clukey et al. 2018; Cocci et al. 2018).

According to the literature, POPs may cause some health problems in organisms, like damage to some organs, due to their neurotoxicity and hepatotoxicity; alterations in reproduction and anomalies in the development of embryos caused by endocrine disruption; effects at the cellular level such as genetic damage, induction of apoptosis, immunosuppression and the generation of cancer cells as a consequence of oxidative stress; changes in biochemical and hematological parameters of organisms, among others (Keller et al. 2004; D’lilio et al. 2011; Camacho et al. 2013; Perrault et al. 2017; Tremblay et al. 2017; Cocci et al. 2018; Casini et al. 2018; García-Hernández et al. 2018; Nava Montes et al. 2020). The presence of POPs in tissues and fluids of various organisms has been evaluated in some studies around the world, including amphibians (Valdespino et al. 2015; González-Mille et al. 2010), reptiles (Barraza et al. 2020; Nava Montes et al. 2020; Filippos et al. 2021), birds (Mo et al. 2019; Bouwman et al. 2021; Hao et al. 2021), and marine mammals (Espinoza et al. 2019; Megson et al. 2022).

The sea turtle *L. olivacea* is a pantropical marine species, distributed in tropical and subtropical oceanic regions worldwide. This turtle has a nomadic behavior with flexible strategies that allow it to survive. They can migrate long distances through the sea feeding on surface fauna, but generally, this turtle remains in estuarine ecosystems and coastal waters, where they can dive to a depth of 500 ft to find food. The turtle is omnivorous so that it can feed on a wide variety of organisms, including algae, lobsters, crabs, mollusks, and benthic invertebrates (Behera et al. 2014; Cáceres-Farias et al. 2022; Colman et al. 2014; NOAA Fisheries 2022; Pritchard and Trebbau 1984).

The olive ridley turtle has two types of reproductive behaviors in nesting: solitary nesting and mass nesting in synchrony. The latter is a phenomenon that is colloquially called “arribada”, which refers to the arrival of many turtles to the coast in a short period of time (Cáceres-Farias et al. 2022; Dromgool-Regan and Crowley, n.d.; NOAA Fisheries 2022; Pritchard and Trebbau 1984). “La Escobilla” Sanctuary in Oaxaca, México, is recognized worldwide as a site with high arrival activity for massive olive ridley turtle nesting. Currently, the nesting season spans approximately 11 months of the year, running from May to March. The “arribadas” can last for a day or up to a month (Cervantes-Hernández et al. 2017; Ocana et al. 2012; SEMARNAT 2009; Sosa-Cornejo et al. 2021).

They are considered vulnerable by the IUCN and endangered by the Official Mexican Standard NOM-059-SEMARNAT-2010. The main threats faced by the olive ridley turtle in the world are overexploitation of turtle meat and eggs, incidental capture in fishing activities, and exposure to contaminants during their long life. Some characteristics of these turtles, such as their eating habits, wide distribution, and migratory habits, aggravate the degree of exposure to pollutants (Muñoz and Vermeiren 2023; Cortés-Gómez et al. 2017; Dias et al. 2023; Dromgool-Regan and Crowley, n.d.; Filippos et al. 2021; IUCN 2022; Keller et al. 2004; Nava Montes et al. 2020; Salvarani et al. 2023; SEMARNAT 2010). Therefore, it is necessary to carry out studies that allow us to know the health status of the olive ridley sea turtle populations. It is also essential to implement strategies that promote the preservation of the species and the conservation of the habitats in which it develops. Then, this work aimed to determine long-term exposure to POPs in adult female olive ridley turtles (*L. olivacea*), sampled during the nesting season, in “La Escobilla” Sanctuary, Oaxaca.

Material and methods

Study area

The study was carried out at the “La Escobilla” Sanctuary, located in Santa María Tonameca, Oaxaca, Mexico, on the Mexican Pacific coast (15.433756° N, 96.444923° W) (Fig. 1).

Field sampling

During the June 2014 nesting season, and after female nesting, blood samples were obtained from the dorsal cervical sinus of adult female turtles (having previously sanitized the puncture area). A 5-ml syringe was used, and blood was drawn into Vacutainers® tubes with the anticoagulant lithium heparin (Owens and Ruiz 1980). In situ, after sample collection, 2 ml of plasma was obtained by centrifuging the samples at 3000 rpm for 5 min. The obtained plasma was collected in 3-ml Eppendorf tubes and stored. Samples were stored at 4 °C until processing in the ecotoxicology laboratory of the Faculty of Medicine of the Autonomous University of San Luis Potosí, Mexico. The group obtained a scientific collection permit for the study from the Mexican Ministry of Environment and Natural Resources with the following code: SGPA/DGVS/11741/13.

Pollutant analysis

For the POPs quantification in plasma samples, we used the focused ultrasound-assisted extraction technique described

"La Escobilla" Sea turtle Sactuary

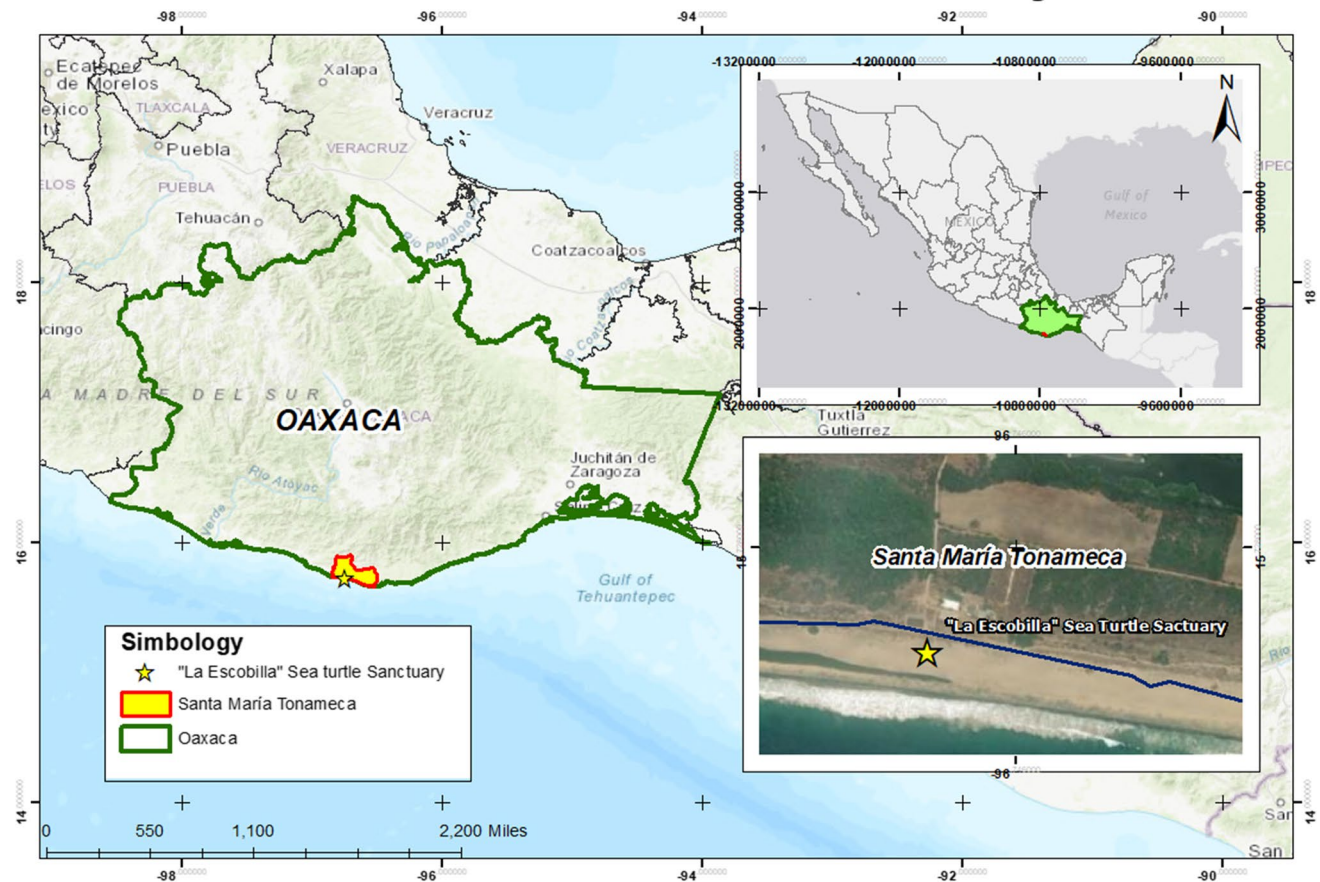


Fig. 1 Localization of the “La Escobilla” sea turtles sanctuary, in Oaxaca, Mexico. (Source: Portal de Geoinformación—CONABIO; Geographic coordinate system, WGS84; elaborated: Sanjuan-Meza, Eleno Uriel and Castillo-Ipiña, Jesús Alfredo)

by Flores-Ramírez et al. (2015) using an Ultrasonic Processor GEX130 (115 V 50/60 Hz) with a 3-mm titanium point. Twenty-seven POP analytes were determined, including the OP: alpha hexachlorocyclohexane (α HCH), beta hexachlorocyclohexane (β HCH), gamma hexachlorocyclohexane (δ HCH), hexachlorobenzene, alpha endosulfan (α endosulfan), beta endosulfan (β endosulfan), endosulfan sulfate, dichloro diphenyl trichloroethane (DDT), its major metabolite, dichloro diphenyl dichloroethylene (DDE), as well as the polychlorinated biphenyls (PCB): PCB 52, PCB 101, PCB 99, PCB 118, PCB 153, PCB 105, PCB 138, PCB 187, PCB 183, PCB 128, PCB 156, and PCB 180. As a quality control, we use an internal standard of α HCB_{C13} and PCB 141_{C13}, prepared to a 1-ppm stock. The internal control and validation of the method were performed by evaluating the limit of detection, the limit of quantification, linearity, sensitivity, and the percentage of recovery. All these parameters were obtained using seven calibration curves in a matrix of five points each (10, 25, 50, 100, and 200 ppb) to validate the precision of the method by evaluating different concentrations (AOAC, Fao, IAEA, Iupac., 2000).

Recovery percentages range between 67.8 and 148.9% at the lowest concentration (10 ppb) and between 90.9 and 119.9% at the highest concentration (200 ppb). For sample fortification, standards of OP compounds and PCB as well as internal standards PCB 141 C₁₃ and α HCH C₁₃ of the Chemservice brand were used at a concentration of 1000 ppb in solvent (Hexane).

Statistical analysis

The STATISTICA version 10 software was used to perform the analysis (StatSoft Inc 2010). Normality tests were performed and according to the results, non-parametric descriptive statistics were executed, including the median, the 25th and 75th percentile, and the minimum and maximum values per compound. Additionally, the sums of the isomers of lindane, endosulfan, DDT metabolites, PCBs, and POPs in general were calculated to compare our results with those published in other studies.

Results and discussion

Among the quantified POPs, the exposure pattern from highest to lowest concentration was DDT > DDE > PCB 118 > α endosulfan > γ HCH > PCB 52 > endosulfan sulfate. OPs such as lindane (δ HCH) were detected in all turtle samples ($n=40$), while DDT and DDE were found in 97.5% of the samples, as well as PCB 52 (97.5%). PCB 118 was found in 62.5% of the samples. The results of plasma POPs of the present study (ng/g lipid) are as follows (median, range): \sum HCH = 16.67, 5.04–46.57; \sum DDT = 45.01, 42.19–70.42; \sum endosulfan = 7.75, 0.0–922.21; \sum PCB = 36.58, 8.30–103.33; \sum POP = 124.26, 79.48–1000.8 (Table 1).

In ecotoxicological studies, reptiles are the least evaluated taxonomic group, so there are few POP studies on sea turtles. Table 2 shows the results compared to the reports in some studies around the world. Camacho et al. (2012) determined the concentrations of OPs and PCBs of *Caretta caretta* in Islas Canarias. In their results, PCB 52 was found in 47% of the samples (median, 0.01 ng/mL; rank, 0–0.27 ng/mL), while DDE was found in 89.6% of the samples (median, 0.28 ng/mL; rank, 0.0–8.97 ng/mL). One year later, Camacho et al. (2013) determined the POP and PCB concentrations in blood samples of nesting females of *C. caretta*. They report the following values: p,p'-DDE, 0.057 (<LOD–0.377) ng/mL; PCB-52, 0.01 (<LOD–0.06) ng/mL; \sum PCBs, 0.11 (0.02–1.25) ng/mL. Later, in Boa Vista, Africa, Camacho et al. (2014) determined POP and PCB concentrations in plasma samples from *Chelonia mydas* ($n=21$) and *Eretmochelys imbricata* ($n=13$). Total PCBs concentrations were 0.23 (0.01–4.04) ng/mL in *C. mydas* and 0.04 (<LOQ–1.41) ng/mL in *E. imbricata*. Recently, Filippou et al. (2021) reported in a study carried out in the Biological Reserve “Atol das Rocas” in Brazil

the concentrations of various POPs in organisms of *L. olivacea* [\sum HCH = 0.010 (<LC–0.069), \sum DDTs = 0.031 (<LC–0.560), \sum PCBs = 0.441 (0.088–3.04)].

When comparing our results with those reported in other studies, it was observed that the concentrations of HCH, isomers of endosulfan, DDT, total PCBs, and the total sum of POPs in the present study for *L. olivacea* are higher than those reported in studies of other species of sea turtles in the world. These results are relevant because those studies have related physiological effects with the concentrations of POPs they reported (Table 2). For example, some studies have related high levels of PCB with effects on hematological and biochemical parameters in blood, and possible anemia in the organism (Keller et al. 2004; Cocci et al. 2018). Camacho et al. (2013) found that the volume of blood-packed cells (BPC) inversely correlated with the higher concentrations of POP (Table 2), and their results were several times lower than those found in the present study. In addition, they found that PCB 52 and HCB had a negative correlation with white cell counting (WCC) and thrombocyte counting, important cells in the immune system defense processes. Moreover, they also found that turtles with higher concentrations of PCB 52 had elevated blood phosphorus levels and the lowest glucose level. Besides, high concentrations of \sum PCB were associated with low levels of uric acid and total cholesterol, as well as a negative correlation with the Na/K relationship in blood, corroborating what was mentioned by McConnell 1985, which claimed that kidneys are sensitive to the presence of POP.

In addition, some other studies have reported various effects on antioxidant activity enzymes (Labrada-Martagón et al. 2011). For example, Camacho et al. (2013) found that the correlation between the activity of the γ -glutamyl transferase (GGT) and high concentrations of p-p'-DDE and \sum POP was negative (POP concentrations in Table 2), and their results are several times lower than those found in the

Table 1 Concentrations analyzed in plasma of nesting olive ridley sea turtle (*Lepidochelys olivacea*) in La Escobilla Sanctuary, Oaxaca, Mexico ($n=40$)

| Compound | % > DL | Median | | P25 | | P75 | | Minimum | | Maximum | |
|---------------------|--------|--------|------|------|------|------|------|---------|------|---------|------|
| | | a | b | a | b | a | b | a | b | a | b |
| γ HCH | 100.0 | 5.2 | 11.2 | 3.8 | 8.3 | 6.1 | 13.3 | 2.3 | 5.0 | 14.3 | 30.9 |
| DDE | 100.0 | 8.6 | 18.7 | 8.2 | 17.7 | 8.9 | 19.3 | 8.2 | 17.7 | 14.4 | 31.1 |
| DDT | 100.0 | 11.5 | 25.0 | 11.3 | 24.4 | 11.7 | 25.3 | 11.3 | 24.4 | 13.6 | 29.4 |
| α Endosulfan | 25.0 | 8.5 | 13.9 | 3.5 | 7.6 | 14.1 | 27.9 | 3.5 | 7.6 | 19.6 | 42.3 |
| Endosulfan sulfate | 20.0 | 1.1 | 4.0 | 1.1 | 2.4 | 3.4 | 7.4 | 1.1 | 2.4 | 4.5 | 9.7 |
| PCB 52 | 97.5 | 2.6 | 9.0 | 1.6 | 6.4 | 4.1 | 13.2 | 0.8 | 1.7 | 6.1 | 13.2 |
| PCB 118 | 62.5 | 4.6 | 18.1 | 4.6 | 15.3 | 4.6 | 32.0 | 4.6 | 9.9 | 32.5 | 70.2 |

Column a: ng/ml of plasma

Column b: ng/g of lipids in plasma

Column % > DL: percentage values above the detection limit

Column P25: 25th percentile

Column P75: 75th percentile

Table 2 Comparison of plasma POPs concentrations (ng/mL) in studies from various parts of the world with various sea turtle species

| Species | N | Sample | Site | \sum HCH | \sum Endosulfan | \sum DDTs | \sum PCBs | \sum POPs | Reference |
|-------------------------------|----|--------------------|---|-------------------|-------------------|---------------------------|----------------------|---------------------|---------------------------|
| <i>Lepidochelys olivacea</i> | 40 | Plasma (Me; rank) | Protected natural area "La Escobilla" Sanctuary, Oaxaca, Mexico | 7.72 (2.34–21.57) | 3.6 (0–427.17) | 20.85 (19.6–32.6) | 20.67 (4.45–70.18) | 60.1 (39.2–467.3) | This study |
| <i>Eretmochelys imbricata</i> | 29 | Plasma (mean; SEM) | Punta Xen Turtle Camp Yucatan, Mexico | 1.948 ± 0.93 | 1.166 ± 0.545 | 1.593 ± 0.631 | NA | 7.395 ± 3.378 | Salvarani et al. (2023) |
| <i>Lepidochelys olivacea</i> | 19 | Plasma (Me; rank) | Biological Reserve "Atol das Rocas", Brazil | 0.010 (<LQ–0.069) | NA | 0.031 (<LQ–0.560) | 0.441 (0.088–3.04) | NA | Filippos et al. (2021) |
| <i>Caretta caretta</i> | 28 | Plasma (Me; rank) | | 0.038 (<LQ–0.195) | NA | 0.102 (0.0007–0.396) | 1.02 (0.018–2.4) | NA | |
| <i>Chelonia mydas</i> | 31 | Plasma (Me; rank) | | 0.018 (<LQ–0.107) | NA | 0.003 (<LQ–0.022) | 0.171 (0.039–0.951) | NA | |
| <i>Lepidochelys kempii</i> | 79 | Plasma (Me; rank) | "Santuario Playa de Rancho Nuevo" ANP Tamaulipas, Mexico | <DL | NA | 3.45 (3.45–213.67) | 7.61 (2.45–50.55) | 49.53 (9.85–233.77) | Nava Montes et al. (2020) |
| <i>Chelonia mydas</i> | 23 | Plasma (Me; rank) | San Diego Bay, California, EUA | NA | NA | NA | 5.07 (0.36–30.11) | 5.275 (0.36–30.79) | Barraza et al. (2020) |
| <i>Chelonia mydas</i> | 16 | Plasma (Me; rank) | Seal Beach National Wildlife Refuge, California, USA | NA | NA | NA | 0.32 (0.12–5.46) | 0.44 (0.25–5.61) | |
| <i>Caretta caretta</i> | 20 | Plasma (Me; rank) | Adriatic sea coast, Italia | NA | NA | NA | 0.18 | NA | Cocci et al. (2018) |
| <i>Eretmochelys imbricata</i> | 54 | Plasma (Me; rank) | Punta Xen, México | 0.05 (0.0–21.69) | 0.05 (0.0–12.11) | NA | NA | NA | Salvarani et al. (2018) |
| <i>Caretta caretta</i> | 50 | Plasma (Me; rank) | Cabo Verde, África | NA | NA | p,p'DDE 0.057 (<LQ–0.377) | 0.11 (0.02–1.25) | NA | Camacho et al. (2013) |
| <i>Chelonia mydas</i> | 13 | Plasma (Me, m; SD) | Kiholo Bay, Hawaii | NA | NA | NA | 2.84, 25.1 (51.9) | NA | Keller et al. (2004) |
| <i>Chelonia mydas</i> | 14 | Plasma (Me, m; SD) | Hawaiian Islands | NA | NA | NA | 0.170, 1.260 (2.890) | NA | |

Table 2 (continued)

| Species | <i>N</i> | Sample | Site | Σ HCH | Σ Endosulfan | Σ DDTs | Σ PCBs | Σ POPs | Reference |
|------------------------|----------|-------------|---|--------------|---------------------|---------------|---------------|---------------|-----------------------|
| <i>Caretta caretta</i> | 10 | Plasma (Me) | Migratory organisms. Cabo Cañaveral, Florida, EUA | NA | NA | 0.701 | 7.267 | 9.523 | Ragland et al. (2011) |
| <i>Caretta caretta</i> | 10 | Plasma (Me) | Resident organisms. Cabo Cañaveral, Florida, EUA | NA | NA | 0.106 | 5.033 | 5.492 | |

Me median, *m* mean, *SD* standard deviation, *LQ* limit of quantification, *DL* detection limit, *NA* not available

present study. Later, Salvarani et al. (2018) found a negative correlation between the concentrations of OPs, mainly endosulfan, chlordane, methoxychlor, and HCH metabolites, with the activities of catalase and glutathione peroxidase, concluding that these contaminants induce oxidative stress in hawksbill turtles (*Eretmochelys imbricata*). Among other effects, Cocci et al. (2018) reported a positive correlation between global DNA hypermethylation and PCB 52 exposure in sea turtles of the Adriatic Sea, Italy. Notably, our results were higher in POP concentrations than the reported effects (Table 2).

The POP levels found in the sea turtles of the “La Escobilla” Sanctuary may be the result of the conjunction of several exposure routes with different sources of contaminant emissions. For example, the extensive agriculture and livestock farming that occurs in human localities located in the highest parts of the coastal plain in the region (INEGI 2022) promotes the runoff of these chemical substances that are carried away by water currents, such as the Cozaltepec River, which flows to the side of the nesting beach (CONANP 2018). On the other hand, this entire region was considered an important distribution site for diseases such as dengue; hence, large quantities of DDT were applied in the past in response to the health problem, thus generating historical contamination (Secretaría de la Convención de Estocolmo, 2020). Regarding PCBs, some studies have found these contaminants in different environmental matrices with proximity to areas with industrial activity, including petrochemicals (Gedik et al. 2010; Schuhmacher et al. 2004; Wang et al. 2012), and in this study, a refinery in the municipality of Salina Cruz is located close to the study site; on the coast of Oaxaca.

However, reviewing the literature, exposure to POPs for aquatic and semi-aquatic reptiles is also determined by some characteristics of the chemical compounds, as well as by the physiological, feeding, and migratory habits of the organisms and by the stage of the life cycle in which they live.

Other determining factors are the longevity of the turtles, in addition to the characteristics of the habitats in which they live. On the other hand, this family of contaminants is lipophilic, which allows them to accumulate in adipose tissue and to be distributed throughout the bloodstream. However, in some stages of the life cycle, such as reproduction, migration, and nesting, where fatty tissue is used as an energy reserve, it is diluted and distributed throughout the body, which can produce variations in the concentrations of POPs in the blood of organisms and increase exposure to these chemicals (Camacho et al. 2014; Bucchia et al. 2015; García-Besné et al. 2015; Nava Montes et al. 2020). Additionally, POPs can be transferred from the mother to the neonate turtles, and accumulated POPs can persist beyond the lifetime of an organism, affecting population dynamics in the following generations after the organism was exposed (Muñoz and Vermeiren 2020, 2023).

Therefore, the main routes of exposure of sea turtles to POPs, in order of importance, are ingestion, direct contact, and inhalation, and in hatchlings, transfer to offspring (Bucchia et al. 2015; Camacho et al. 2014; Dias et al. 2023; Filippos et al. 2021; Salvarani et al. 2023).

Conclusions

The levels of POPs found in the sea turtles of the “La Escobilla” Sanctuary, specifically the concentrations of HCH, isomers of endosulfan, DDT, total PCBs, and the total sum of POPs in the present study for *L. olivacea* are higher than those reported in studies of other species of sea turtles in the world, being important evidence of exposure to xenobiotics in the marine environment.

The presence of POPs in olive ridley sea turtles represents a potential risk to the health of the population of this marine reptile; however, it is important to evaluate the effects of exposure on organisms with ecotoxicological tools, aiming

to establish safety values for the protection of the species. On the other hand, this study highlights the importance of blood as a biomarker for exposure in sea turtles; it is a non-destructive method and is an appropriate substitute for pollutant analysis in different tissues (liver, kidneys, brain, etc.), which possess a greater toxicological risk and are considered destructive.

These results contribute to the generation of studies that allow us to know the current situation of the populations of this species in the world. It establishes important values on exposure to xenobiotics to encourage actions and strategies that promote the conservation of the olive ridley sea turtle, as well as the generation of evidence that allows inferring public policies on the emission and disposal of chemical substances that pollute the environment. It also contributes to promoting the formulation of strategies that allow the remediation of contaminated sites and the mitigation of the effects they cause.

Author contribution All authors contributed to the study's conception and design. Material preparation, data collection, and analysis were performed by Sagrario Paola Mendoza Rivera, Rogelio Flores Martínez, Jesús García Grajales, Alejandra Buenrostro Silva, Eleno Uriel Sanjuan Meza, and Guillermo Espinosa Reyes. The first draft of the manuscript was written by Sagrario Paola Mendoza Rivera and Eleno Uriel Sanjuan Meza, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Data availability Not applicable.

Declarations

Ethical approval This study was carried out with a wildlife scientific collection permit (SGPA/DGVS/11741/13) granted by the Ministry of Environmental and Natural Resources (Secretaría de Medio Ambiente y Recursos Naturales -SEMARNAT-, in Spanish). During the study, we never worked with human populations.

Consent to participate Not applicable.

Consent to publish Not applicable.

Competing interests The authors declare no competing interests.

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