

Research Article

General health status of a stranded *Tursiops truncatus* of the oceanic ecotype in southern Gulf of Mexico, Campeche coasts: a multidisciplinary analysis

Karem Leonela Naranjo-Ruiz¹ , Alberto Delgado-Estrella² , Yassir Edén Torres-Rojas³ 
Isabel Silva⁴ , Mayra D. Manrique-Ortega⁴ , Edgar F. Mendoza-Franco³ 
Jaime Rendón-von Osten³ , Ricardo Dzul-Caamal³  & Rodolfo E. del Río-Rodríguez³ 

¹Posgrado en Ciencias del Mar y Limnología, Instituto de Ciencias del Mar y Limnología
Universidad Nacional Autónoma de México, Ciudad de México, México

²Facultad de Ciencias Naturales, Universidad Autónoma del Carmen (FCN-UNACAR)
Ciudad del Carmen, Campeche, México

³Instituto de Ecología, Pesquería y Oceanografía del Golfo de México
Universidad Autónoma de Campeche (EPOMEX, UACAM), Campeche, México

⁴Centro de Investigación en Corrosión
Universidad Autónoma de Campeche (CICORR, UACAM), Campeche, México

Corresponding author: Yassir Edén Torres-Rojas (yettorres@uacam.mx)

ABSTRACT. Strandings provide valuable information about rare marine organisms and their relationship with their environment. In the southern Gulf of México, specifically on the central coast of Campeche, Mexico, strandings of oceanic cetaceans have rarely been recorded, therefore, biological information has also been scarce. One of such rare stranding cases occurred on September 7, 2022, on the coast of Seybaplaya, Campeche. The specimen was identified as a female of *Tursiops truncatus* with the morphology features of the oceanic ecotype. The carcass was examined shortly after death, and samples for different analyses were obtained fresh. Tissue samples were collected, and some particular observations were made to define the individual's general health status. Results of the analyses revealed damage in the liver, lungs, and kidneys, probably related to 1) the advanced age of the organism (>35 years), and 2) the presence of persistent organic pollutants. Most importantly, this study contributed two new findings, the first record of the parasite *Anisakis* and the presence of renal calculi in the oceanic ecotype of *T. truncatus* in the southern Gulf of Mexico.

Keywords: *Tursiops truncatus*; bottlenose dolphin; *Anisakis*; renal calculi; POPs; health; Gulf of Mexico

INTRODUCTION

Stranding is when a dead or living marine organism is found out of the water or floating near the coast (Gómez-Hernández et al. 2020). These events allow access to animals listed under special protection that are difficult to study, such as cetaceans, and provide invaluable information on their anatomy, life history, genetics, diseases, parasites, predation, pollution, and

trophic ecology (Perrin & Geraci 2009), depending on the condition of the carcass. For these reasons, each stranding should be considered a unique opportunity to learn something that would not have been possible otherwise (Perrin & Geraci 2009).

The causes of stranding can be biological (old age, diseases, predation) or ecological (environmental conditions, temperature rise, anthropogenic interaction, pollution), and determining them is challenging because

the signs and pathologies frequently cannot be determined from a visual examination. In the Gulf of Mexico, 28 species of cetaceans have been recorded (Würsig et al. 2000) from sightings and strandings. However, in the southern Gulf of Mexico (specifically on the Campeche coasts), cetacean strandings are rare events.

One of the best-represented cetaceans in the southern Gulf of Mexico is the bottlenose dolphin, *Tursiops truncatus*, which has a wide distribution. Although two *T. truncatus* ecotypes have been recognized, oceanic and coastal (Wells & Scott 2009), the latter is better represented in the Campeche coast, with the largest population (particularly in the Términos Lagoon) recognized as residents. Most of the local historical strandings of this species have been residents (13 dolphins) and four of the oceanic ecotype (Naranjo-Ruiz 2020).

Two new stranding events have been recorded recently near the Campeche central region: a Mysticeti (*Balaenoptera acutorostrata*) and an Odontoceti (*Peponocephala electra*) (Delgado-Estrella & Naranjo-Ruiz 2018), both considered pelagic or oceanic. On September 7, 2022, a female *T. truncatus* of the oceanic ecotype was stranded alive in the southeastern Gulf of Mexico (coast of the Seybaplaya municipality, Campeche) and died a few hours after the stranding event. The cetacean carcass was fresh at the time of the examination.

This study aimed to perform a multidisciplinary analysis to determine the stranded individual's general health status based on samples and data collected just before his death. The study's output will contribute to the knowledge of this oceanic ecotype of *T. truncatus* in the Gulf of Mexico.

MATERIALS AND METHODS

The geographic location of the stranding

The stranding and subsequent death of the cetacean occurred at 19°37'23.93"N, 90°41'8.52"W (Fig. 1). The Seybaplaya municipality is found in the central-western area of the state of Campeche, corresponding to the Yucatan Peninsula northwestern coast (De la Lanza-Espino et al. 2013) in southeast Mexico, at 17°49'-20°51'N, 89°06'-92°27'W.

Fieldwork

The carcass was examined following the Mexican protocol for "Attention of Strandings of Aquatic Mammals" (DOF 2014) with permission granted by the

SEMARNAT (Board of the Environment and Natural Resources of Mexico). Photographic records, sex identification, morphometric measurements, and necropsy were carried out and recorded *in situ*. Samples were obtained from internal organs (brain, heart, lung, kidney, liver, and spleen). Teeth, skin, blubber, muscle, and gastric cavities were also obtained. Samples were frozen and transported to the Trophic Ecology Laboratory at the Institute of Ecology, Fisheries, and Oceanography of the Gulf of Mexico of Universidad Autónoma de Campeche (EPOMEX-UACAM, by its Spanish acronym) and the College of Natural Sciences of Universidad Autónoma del Carmen (FCN-UNACAR, by its Spanish acronym).

Laboratory work

Age estimation

We analyzed the teeth' dentin and cementum growth layers following Gallo-Reynoso et al. (2014). A tooth from the middle region of the right jaw was removed, and two longitudinal cuts were made, leaving the middle portion of the tooth. The cut was made with a diamond saw blade with a cut bit measuring 152 mm long and 0.5 mm thick. Subsequently, the methodology of Romero-Tenorio & Delgado-Estrella (2015) was followed to wear away and polish the piece until the growth layers could be observed under the stereomicroscope. Dentine layers were counted to determine the specimen age.

Composition of renal calculi

The largest renal calculi (Fig. 2a) was analyzed by X-ray diffraction, scanning electron microscopy, and energy dispersive spectroscopy. The microscopic analyses were performed with a FlexSEM-SU1000™ (Hitachi) scanning electron microscope coupled to a Quantax 75/80™ EDS detector (Bruker). The observation conditions comprised a secondary electron (SE) imaging mode at 15 kV, 5.0 mm working distance, and low vacuum conditions (30 Pa). The conditions of the elemental composition analysis were backscattered electrons (BSE), 15 kV, and 10.0 working distance.

X-ray diffraction was done using a D8 ADVANCE diffractometer (Bruker) with a DAVINCI design (theta-theta geometry) and a LINXEYE silicon detector. The X-ray source is a monochromatic radiation from the Cu tube.

The analytical conditions were NiO 0.02 monochromator, 0.1 mm collimator, 5.0 s pass-through, 0.01° increment, and 6-90° diffraction field. The crystalline phases were identified based on a literature review.

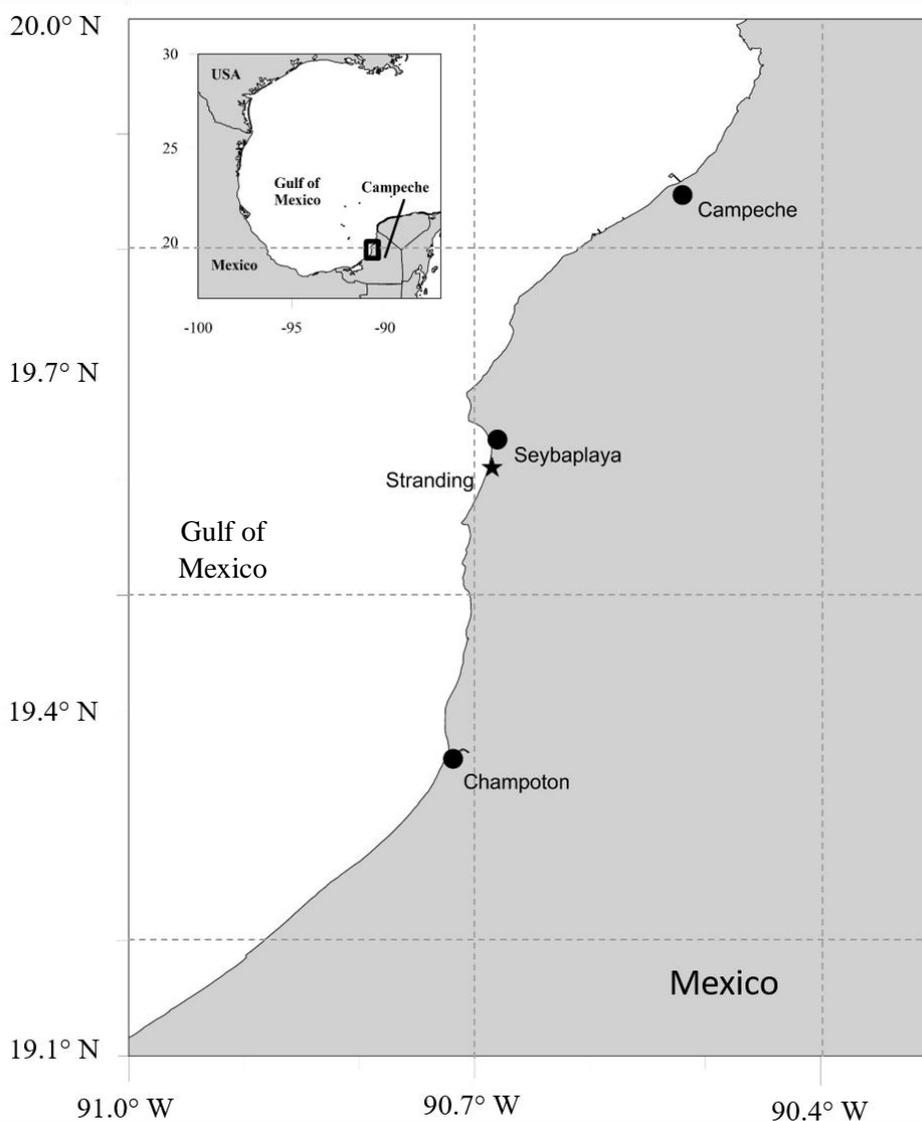


Figure 1. The location of the Seybaplaya municipality, Campeche, and the site where the carcass of a stranded female bottlenose dolphin (*Tursiops truncatus*) was inspected on September 7, 2022.

Identification of parasites in the gastric cavities

The gastric cavities (pre-stomach, main stomach, pyloric stomach) were inspected *in situ*. The parasites in the gastric walls were collected, fixed, and preserved in 70% ethanol. Afterward, they were cleared in alcohol-glycerin (1:20) to observe internal structures in the laboratory.

Histopathology

Brain, heart, lung, kidney, liver, and spleen tissues were fixed in 10% buffered formalin. After 48 h of fixation, portions of all tissues were processed by standard histology, including dehydration, paraffin inclusion, microtome slicing (5 μ m), and stained with Harris

hematoxylin and eosin. Histological sections were permanently mounted on PERTEX and observed under a light microscope at 5, 10, and 40x magnifications.

Biomarker analysis

Portions of sampled tissues (muscle, heart, lung, kidneys, liver, and spleen) were homogenized in 1 mL cold 100 mM phosphate saline (PBS; pH 7.4) using a PRO250 homogenizer (Pro Scientific). The tissue homogenates were divided into two portions; one sample was centrifuged at 12,000 g for 20 min at 4°C using a Centrifuge 5417 R (Eppendorf) to produce the post-mitochondrial fraction (S9). The S9 aliquots were stored at -70°C until tested for glutathione S-transferase

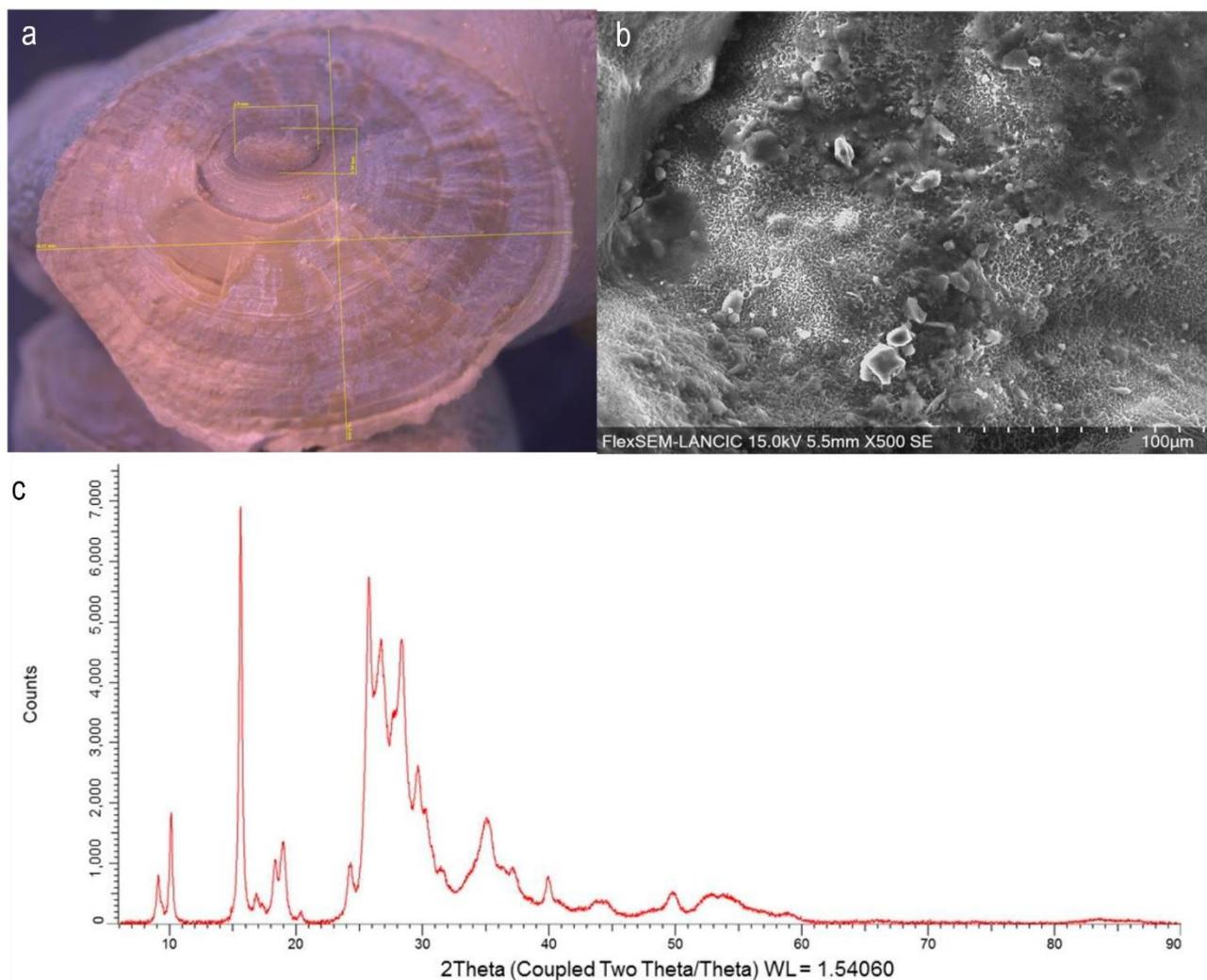


Figure 2. Physical and chemical characteristics of renal calculi found in the stranded female bottlenose dolphin (*Tursiops truncatus*). a) 0.065x stereo microscope photograph of the inner part, b) electron micrograph highlighting the structure of the crystals that make up the stone analyzed, and c) diffractogram of the inner area.

(GST, EC 2.5.1.18), acetylcholinesterase (AChE), and catalase (CAT). The non-centrifuged portion was stored at -70°C until tested for lipid peroxidation (LPO) contents. Total protein concentration was determined using an assay based on the microplate-adapted Bradford (1976) protein-dye binding protocol.

GST activity was evaluated in the S9 fractions of tissues using the method of Habig et al. (1974) adapted to a microplate using 1-chloro-2,4-dinitrobenzene (CDNB; $\epsilon = 9.6 \text{ mM}^{-1} \text{ cm}^{-1}$). Absorbance was recorded at 340 nm (25°C) for 3 min and expressed as $\text{nmol min}^{-1} \text{ mg protein}^{-1}$.

AChE activity was measured using acetylthiocholine (ATC) as substrate according to the colorimetric method of Ellman et al. (1961) with some microplate

modifications (Rendón-von Osten et al. 2005). Enzyme activity was determined kinetically at 414 nm for 5 min. AChE activities were expressed as $\text{nmol min}^{-1} \text{ mg protein}^{-1}$ using a molar extinction coefficient ($13.6 \text{ mM}^{-1} \text{ cm}^{-1}$).

Radi et al. (1991) described a method for estimating CAT activity by measuring hydrogen peroxide (H_2O_2) dismutation at 240 nm for 1 min. CAT activity was calculated as $\text{mmol H}_2\text{O}_2$ consumed per minute per mg of protein using a molar extinction coefficient of $0.043 \text{ mM}^{-1} \text{ cm}^{-1}$.

The LPO breakdown product was determined by the formation of thiobarbituric acid-reactive substances (TBARS), according to Buege & Aust (1978), with modifications to the microplate. LPO results were

expressed in nmol TBARS mg⁻¹ protein using a molar extinction coefficient (MCC) of 1.56×10^5 M cm⁻¹. All spectrophotometric tests were performed using a multimode reader (Multiskan Spectrum, Thermo Scientific).

Contaminant analysis

Blubber, muscle, and liver tissues were dehydrated at 45°C for 24 h, and 1 g of tissue sample was homogenized and placed in vials with 25 mL hexane:dichloromethane (1:1). Then, samples were cleaned in a 30 min ultrasonic pro. After the vials were removed, two drops of toluene were added to each; then, the vials were left to stand for 24 h to allow for toluene evaporation. Subsequently, 500 µL of hexane was added per sample. Samples were purified in glass columns with 5 cm of silica, 4 cm of alumina, and 1 cm of sodium sulfate.

A GC-MS/MS analysis was performed using a Thermo Scientific TRACE 1310 gas chromatograph coupled to a TSQ 8000 Evo triple quadrupole mass spectrometer equipped with SRM type Scan Electronic Ionization methods, a Thermo Scientific AI/AS 1310 automatic sampler, and Trace Finder software. The column used was a TR-PCB 8MS 50 m × 0.25 mm ID × 0.25 µm.

RESULTS

The stranded specimen was visually inspected *in situ* by UNACAR stranding network members and EPOMEX personnel under the permit of PROFEPA and supported by its staff. The carcass corresponded to a female bottlenose dolphin (*T. truncatus*) measuring 260 cm in total length. The specimen was identified as the oceanic ecotype for its darker coloration, presence of two wider stripes at the throat region, a larger body size, and a longer and more falcate dorsal fin in comparison with the coastal ecotype (Simões-Lopes et al. 2019). Scars were observed throughout the whole body, and a 5 cm-diameter healing likely produced by interaction with a pelagic cookie-cutter shark of the genus *Isistius* was also observed.

Age estimation

The tooth dentin and cement growth layers analyses determined that the stranded female was at least 35 years old. This estimated age was also consistent with tooth wear and tear.

Renal calculi composition

Scanning electron micrographs revealed a microscopically rough surface consisting of small needle-shaped crystals (Fig. 2b). The EDS elemental analysis

showed a C, N, and O composition mostly corresponding to ammonium acid urate (NH₄C₅N₃N₄O₃), which has been identified as the primary mineral in kidney stones analyzed by X-ray diffraction (Fig. 2c) (Klohn et al. 1986, Tettenhorst & Gerkin 1999).

Identification of parasites in the gastric cavities

The gastric cavities were empty. We found 98 nematode parasites of the family Anisakidae (*Anisakis* spp.) located in an ulcer lesion of the pre-stomach wall (Pugliares et al. 2007). Of these, 18 adult females were selected for morphometric description. The body length was moderately elongated and averaged 21 mm (range: 16.4-38.9 mm). The body width was 0.63 mm (0.5-0.7 mm). The cuticle was transversely striated, except on the mouth lips. There were three lips: one dorsal (Fig. 3a) and two ventrolateral (Fig. 3b), which were well-developed with a width of 0.17 mm (range: 0.15-0.22 mm). Papillae were not observed. The esophagus was muscular and cylindrical with a width of 0.11 mm (range: 0.07-0.15 mm) wide. The ventricle was sigmoid and inconspicuous, 0.17-0.2 mm long. The tail was conical (Fig. 3c).

Histological diagnosis

Pathological changes were observed in the lungs and liver. The lung tissue showed a large amount of suppurative material in the lower and middle airways, with a variable number of inflammatory response cells, mainly neutrophils. Apart from the cellular components, the suppurative material was probably mucin (Figs. 4a-b). Liver tissue showed infiltration of fibrous tissue (Fig. 4c), probably cirrhotic, with some necrotic foci (Fig. 4d).

Biomarker

The results (Fig. 5) indicated a higher GST activity in the liver, followed by the lung and kidneys. On the other hand, the AChE activity decreased in the following order: liver, kidney, lung, and muscle tissues. Regarding oxidative stress biomarkers, the highest CAT activity was recorded in the liver. Higher lipoperoxidation contents accompanied this antioxidant defense enzymatic activity.

Pollutant analysis

The concentrations of 42 different persistent organic pollutants (POPs) were determined, including polycyclic aromatic hydrocarbons (PAHs), organochlorine pesticides (OCPs), and polychlorinated biphenyls (PCBs) (Table 1). In the blubber, the highest concentration corresponded to Endrin ketone, followed

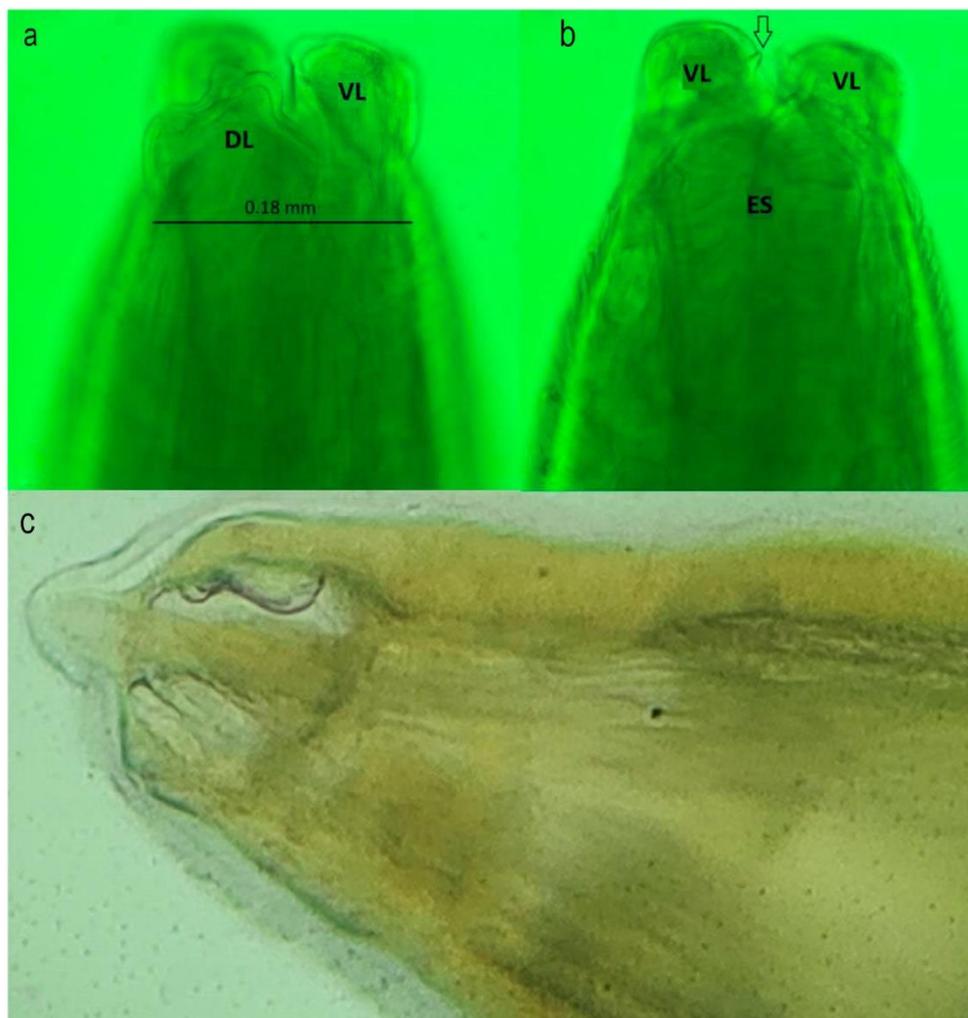


Figure 3. Lips of the anterior end. a) DL: dorsal lip, VL: ventral lip; b) ventral lips showing a papilla (see arrow); ES: esophagus; c) tapered tail of *Anisakis* spp. Specimens found in the stranded female bottlenose dolphin (*Tursiops truncatus*).

by Dieldrin (OCPs); in the liver, the highest concentration was beta-endosulfan (also an OCP). In muscle, the highest compounds were naphthalene and benzo [a]pyrene (PAHs).

DISCUSSION

Cetacean strandings are observed more frequently on the southern coast of Campeche, including Carmen Island, than on the northern and central coasts. Given the few strandings in this region, keeping records and collecting samples of the stranded specimens of cetacean species is important, particularly for the oceanic ecotype of *T. truncatus*, for which limited information is available. The rare stranding of this ecotype allowed us to collect samples and data for analyses of different biological and ecological traits.

The causes of the stranding examined in this study can be diverse. The elderly female examined showed signs of aging, such as tooth wear and a general emaciated condition. It had a wound caused by a shark of the genus *Isistius*, a predator of various cetacean species in the Gulf of Mexico, mainly pelagic species found at depths between 823 and 3038 m (Grace et al. 2018). In the study region, these conditions occur only in offshore areas beyond the continental shelf.

Composition of renal calculi

Pathological ammonium acid urate crystals are most frequent in renal calculi of *T. truncatus* under human care (Geng et al. 2019) and are concurrent with other health disorders such as anemia, high blood urea nitrogen, high creatinine, and a low glomerular filtration rate, in addition to old age (Venn-Watson et

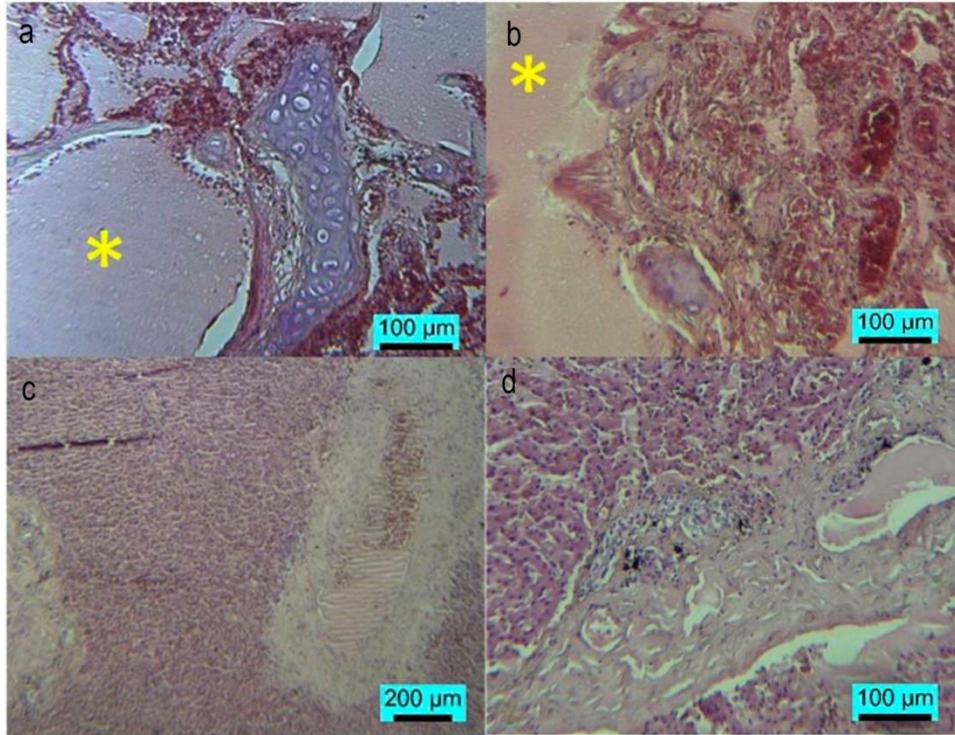


Figure 4. Photomicrographs of lung and liver tissue sections from a female bottlenose dolphin (*Tursiops truncatus*). a) Presence of mucin (yellow asterisk) in the upper pulmonary airways, b) vascular congestion and airway occlusion by mucoid substance (yellow asterisk), c) fibrotic tissue in the liver (right of the photograph), d) necrotic tissue in the liver (center and bottom of the photograph).

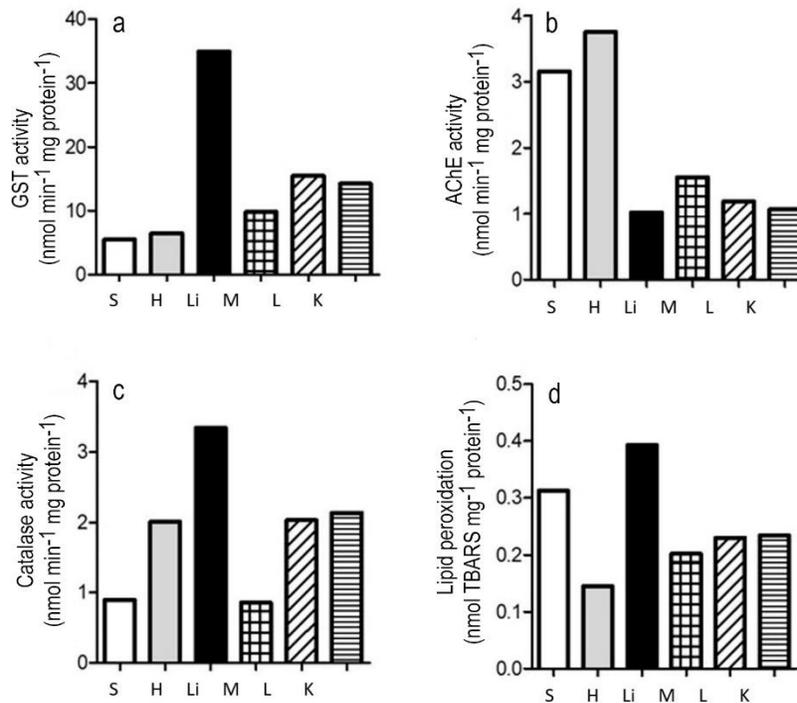


Figure 5. Biomarker response in tissues from a female bottlenose dolphin (*Tursiops truncatus*). a) Glutathione S-transferase (GST), b) acetylcholinesterase (AChE), c) catalase, and d) lipid peroxidation. S: spleen, H: heart, Li: liver, M: muscle, L: lung, K: kidney tissues.

Table 1. Concentrations of persistent organic pollutants in tissues of an oceanic-ecotype female bottlenose dolphin (*Tursiops truncatus*). (Bold values indicate highest concentration).

Compound	Blubber	Muscle	Liver
	ng g ⁻¹		
PAHs			
Acenaphthene	0.08353691	0	0.0252108
Benz[a]anthracene	0	0	0.00241594
Benzo[a]pyrene	0	0.02192163	0
Chrysene	0	0	0.00153644
Dibenz[a,h]anthracene	0	0	0.04916601
Fluoranthene	0	0	0.00331966
Naphthalene	0	0.05216003	0.06898499
Phenanthrene	0	0	0.01289087
OCPs			
Beta HCH	0	0	0.00325965
Gamma HCH	0	0.00442974	0.00144628
pp-DDD	0	0	0.00506563
Aldrin	0.02056907	0	0.00216782
Dieldrin	0.56072211	0	0
Endrin ketone	3.23536072	0	0
Endosulfan sulfate	0.00803047	0.00496553	0.00238679
beta-endosulfan	0	0	0.1837301
Cis-chlordane	0	0.00250302	0.00047597
Trans-chlordane	0	0.01001119	0.00483061
Heptachlor	0.00552813	0	0
Heptachlor epoxide	0	0	0.01014725
Indeno [1,2,3-cd] pyrene	0	0	0.01830591
Methoxychlor	0	0	0.00267089
PCBs			
PCB 149	0.00220687	0.00742944	0
PCB 18	0.00171929	0.000719	0.0007178
PCB 28	0.00366534	0	0
PCB 31	0.0035995	0	0
PCB 44	0.00227751	0	0
PCB 52	0	0.00168912	0.0012491

al. 2010, Colegrove 2018). However, few cases of nephrolithiasis have been reported in wild dolphins from the southern region of the Gulf of Mexico and the state of Campeche. Therefore, this study represents the first renal calculi of ammonium acid urate recorded in a wild oceanic bottlenose dolphin in the study area.

Parasites in gastric cavities

Anisakis nematodes have been described and reported as endoparasites of oceanic dolphins of the families Delphiniidae, Kogidae, Monodontidae, and Phocoenidae (Di-Azevedo et al. 2015, Shamsi et al. 2019). *T. truncatus* has been reported as a host of adult stages of *Anisakis pegreffii*, *Anisakis* spp., and *A. simplex* (s.l.) in the Atlantic Ocean (Mattiucci et al. 2005, Romero et al. 2014) or Mediterranean Sea (Quiñones et al. 2013,

Hrabar et al. 2017, Cipriani et al. 2022), sometimes forming gastric granulomatous ulcers (Hrabar et al. 2017, Ryeng et al. 2021), as observed in the present study. The two geographically nearest records of *Anisakis* spp. infection has been previously reported on *Kogia breviceps* stranded on the Yucatan north coasts (Hernández-Olascoaga et al. 2023) and *Stenella clymene* on the Mexican Caribbean coast (Aguilar-Aguilar et al. 2010). Therefore, the present study is the first to report *Anisakis* spp. parasitizing *T. truncatus* stranded on the Campeche coast (southern Gulf of Mexico).

The adult nematode specimens analyzed in the present study were opportunistically sampled. Specific identification was impossible because male nematodes are required to assess other morphological characte-

ristics, such as ventricle length and shape and papillae morphology in lips and tail.

Histopathology analysis, presence of persistent organic pollutants and biomarkers

POPs can suppress the immune system of organisms (Reckendorf et al. 2010). Besides, dolphins are susceptible to respiratory diseases due to the large amount of air they exchange during breathing (Venn-Watson et al. 2012), and large amounts of mucin are linked to lung disease (Denneny et al. 2020). In this case, these signs, observed in the histopathology analysis, are due to a likely bacterial or viral lung disease causing occlusion of the airways, as observed in the upper and lower respiratory tract. Micrographs also showed diffuse bleeding between the alveoli, which suggests a chronic or acute disease in the airways. Hence, the stranded dolphin might have experienced respiratory issues throughout the disease until it died.

The main causes of liver disorders in wild organisms have been infections and exposure to petroleum products such as oil spills, PCBs, and other POPs (Nollens et al. 2018). In the present study, fibrous tissue, cirrhotic tissue, and necrotic foci were observed (Fig. 4), indicating a nonspecific chronic liver pathology.

Given the lipophilic characteristics of the compounds analyzed, most POPs were detected in the liver, one of the main organs related to metabolism. However, blubber showed the highest POP concentrations, as reported in other studies with cetaceans (García-Álvarez et al. 2014, Lourenço et al. 2021).

In the present study, PAHs were the most outstanding compounds for showing the highest concentrations in the analyzed tissues. Naphthalene has been one of the predominant PAHs in cetacean tissues (Lourenço et al. 2021), and this study recorded the highest concentrations in muscle tissue. The presence of a large number of these compounds is not surprising, considering that one of the main anthropic activities in the Campeche Sound is offshore oil extraction and that this type of compounds can be metabolized by organisms included in the dolphin diet, such as fish and cephalopods. It should be noted that these types of compounds are known carcinogenic and mutagenic chemicals (Lourenço et al. 2021).

In the case of OCPs, when comparing the results of the present study with data from Flores-Sánchez et al. (2018) in blubber from dolphins inhabiting the Terminos Lagoon, no correlation was found in the compounds analyzed between both studies, thus

indicating the differences between the coastal and oceanic habitats of the bottlenose dolphin.

Therefore, the results of the concentrations of different POPs, the high activity of GST, CAT, and LPO, and the low activity of AChE suggest that the organ damage found may be related to exposure to POPs due to the activity of biomarkers signaling the interaction of organisms with environmental agents such as pollutants (World Health Organization & International Programme on Chemical Safety 1993, Hellou et al. 2012).

Despite oil-extraction activities in the study area, the tissue concentrations of chemicals analyzed in this study were low but detectable compared to other studies conducted in Florida and the Canary Islands (Lourenço et al. 2021). Several studies, such as García-Álvarez et al. (2014), mention that OCP and PCB concentrations in *T. truncatus* specimens of the eastern Atlantic Ocean are usually higher in adult males than in females due to the transfer from females to offspring during pregnancy and lactation, which may explain the low concentrations of these compounds in the tissues of the stranded female in this study. However, studies on male specimens of similar ages are needed to confirm this hypothesis in the region.

This study investigated the health status of a stranded female bottlenose dolphin of the oceanic ecotype. Its relevance lies in being a multidisciplinary study that reported two key findings for the first time: one regarding renal calculi and the other reporting *Anisakis* nematodes in a wild dolphin of the oceanic ecotype. These findings raise questions about the health status of cetaceans in the Gulf of Mexico. It should be emphasized that the multidisciplinary analyses on the oceanic ecotype of *T. truncatus* in this study were based on a single individual. Therefore, it is necessary to conduct future research on additional individuals of this species/ecotype for monitoring, management, and conservation purposes.

ACKNOWLEDGEMENTS

NRKL acknowledges the Consejo Nacional de Humanidades, Ciencia y Tecnología (CONAHCYT), for the Ph.D. student grants awarded. DEA acknowledges that the handling of strandings and sample collection from carcasses were covered by the permits issued by the Dirección General de Vida Silvestre number SGPA/DGVS/01726/22. The authors acknowledge PROFEPA staff and the biologist Teresa Cobos for their support during the carcass inspection. Thanks also to the Testing Laboratory of the

Laboratorio Nacional de Ciencias para la Investigación y Conservación del Patrimonio Cultural at Centro de Investigaciones en Corrosión (LANCIC-CICORR) and to the biologist Ana Delia Cu and Francisco Criollo for their logistical and technical support. María Elena Sánchez-Salazar edited the English manuscript.

REFERENCES

- Aguilar-Aguilar, R., Delgado-Estrella, A. & Moreno-Navarrete, G. 2010. New host report for nematodes in a stranded short-snouted spinner dolphin *Stenella clymene* (Cetacea: Delphinidae) from the Mexican Caribbean coast. *Helminthologia*, 47: 136-138. doi: 10.2478/s11687-010-0020-0
- Bradford, M.M. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry*, 7: 248-254. doi: 10.1016/0003-2697(76)90527-3
- Buege, J.A. & Aust, S.D. 1978. Microsomal lipid peroxidation. *Methods in Enzymology*, 52: 302-310. doi: 10.1016/S0076-6879(78)52032-6
- Cipriani, P., Palomba, M., Giulietti, L., Marcer, F., Mazzariol, S., Santoro, M., et al. 2022. Distribution and genetic diversity of *Anisakis* spp. in cetaceans from the Northeast Atlantic Ocean and the Mediterranean Sea. *Scientific Reports*, 12: 13664. doi: 10.1038/s41598-022-17710-12
- Colegrove, K. 2018. Noninfectious diseases. In: Gulland, F.M.D., Dierauf, L.A. & Whitman, K.L. (Eds.). *CRC handbook of marine mammal medicine*. CRC Press, Boca Raton, pp. 267-284.
- De la Lanza-Espino, G., Ortiz-Pérez, M.A. & Carbajal-Pérez, J.L. 2013. Diferenciación hidrogeomorfológica de los ambientes costeros del Pacífico, del Golfo de México y del Mar Caribe. *Investigaciones Geográficas, Boletín del Instituto de Geografía, UNAM*, 81: 33-50.
- Delgado-Estrella, A. & Naranjo-Ruiz, K.L. 2018. Primeros registros de varamientos de la ballena de minke (Balaenopteridae: *Balaenoptera acutorostrata*) y del delfín cabeza de melón (Delphinidae: *Peponocephala electra*), en la costa de Campeche, México. *Revista Mexicana de Mastozoología, Nueva Época*, 8: 18-22. doi: 10.22201/ie.20074484e.2018.1.1.249
- Denneny, E., Sahota, J., Beastson, R., Thornton, D., Burchell, J. & Porter, J. 2020. Mucins and their receptor in chronic lung disease. *Clinical & Translational Immunology*, 9: e1120. doi: 10.1002/cti2.1120
- Di-Azevedo, I.N., Knoff, M., Carvalho, V.L., Mello, W.N., Lopes, T.E.J., Gomez, D.C., et al. 2015. Morphological and genetic identification of *Anisakis paggie* (Nematoda: Anisakidae) in dwarf sperm whale *Kogia sima* from Brazilian waters. *Diseases of Aquatic Organisms*, 113: 103-111. doi: 10.3354/dao02831
- Diario Oficial de la Federación (DOF). 2014. Acuerdo mediante el cual se expide el protocolo de atención para varamiento de mamíferos marinos. [https://www.dof.gob.mx/nota_detalle.php?codigo=5348898&fecha=17/06/2014#gsc.tab=0]. Reviewed: March 10, 2023.
- Ellman, G.L., Courtney, K.D., Andres Jr., V. & Featherstone, R.M. 1961. A new and rapid colorimetric determination of acetylcholinesterase activity. *Biochemical Pharmacology*, 7: 88-95. doi: 10.1016/0006-2952(61)90145-9
- Flores-Sánchez, E.J., García-Salinas, M.A., Delgado-Estrella, A., Calderón-Garcidueñas, A.L., Waliszewski, S.M., Infanzón-Ruiz, R., et al. 2018. Presencia de plaguicidas organoclorados en muestras biológicas de toninas (*Tursiops truncatus*) y manatíes (*Trichechus manatus manatus*) colectadas en el Sur del Golfo de México. *Revista Internacional de Contaminación Ambiental*, 34: 17-28. doi: 10.20937/RICA.2018.34.esp01.02
- Gallo-Reynoso, J.P., Francisco, E.O. & Leo-Ortiz, C. 2014. Age estimation on long-beaked common dolphins, *Delphinus capensis*, from the Gulf of California. *Therya*, 5: 449-460. doi: 10.12933/therya-14-198
- García-Álvarez, N., Boada, L.D., Fernández, A., Zumbado, M., Arbelo, M., Sierra, E., et al. 2014. Assessment of the levels of polycyclic aromatic hydrocarbons and organochlorine contaminants in bottlenose dolphins (*Tursiops truncatus*) from the Eastern Atlantic Ocean. *Marine Environmental Research*, 100: 48-56. doi: 10.1016/j.marenvres.2014.03.010
- Geng, X., Meegan, J., Smith, C., Sakhaee, K. & Rimer, J.D. 2019. Crystallization of hierarchical ammonium urate: insight into the formation of cetacean renal stones. *Crystal Growth and Design*, 19: 6727-6735. doi: 10.1021/acs.cgd.9b01077
- Gómez-Hernández, G., Seingier, G., Elorriaga-Verplancken, F. & Heckel, G. 2020. Status and scope of marine mammal stranding research in Mexico. *Journal of Coastal Conservation*, 24: 3. doi: 10.1007/s11852-019-00725-8
- Grace, M.A., Aichinger-Dias, L., Maze-Foley, K., Sinclair, C., Mullin, K.D., Garrison, L., et al. 2018.

- Cookie-cutter shark bite wounds on cetaceans of the Gulf of Mexico. *Aquatic Mammals*, 44: 491-499. doi: 10.1578/AM.44.5.2018.491
- Habig, W.H., Pabst, M.J. & Jakoby, W.B. 1974. Glutathione S-transferases. The first enzymatic step in mercapturic acid formation. *Journal of Biological Chemistry*, 249: 7130-7139.
- Hellou, J., Ross, N.W. & Moon, T.W. 2012. Glutathione, glutathione S-transferase, and glutathione conjugates, complementary markers of oxidative stress in aquatic biota. *Environmental Science and Pollution Research*, 19: 2007-2023. doi: 10.1007/s11356-012-0909-x
- Hernández-Olascoaga, A., Guillén-Hernández, S. & Díaz-Gamboa, R.E. 2023. Parasites of pygmy sperm whales (*Kogia breviceps*) stranded in the Southern Gulf of Mexico. *Aquatic Mammals*, 49: 177-183. doi: 10.1578/AM.49.2.2023.177
- Hrabar, J., Bocina, I., Kurilj, A.G., Duras, M. & Mladineo, I. 2017. Gastric lesions in dolphins stranded along the Eastern Adriatic coast. *Diseases of Aquatic Organisms*, 125: 125-139. doi: 10.3354/dao03137
- Klohn, M., Bolle, J.F., Reverdin, N.P., Susini, A., Baud, C.A. & Graber, P. 1986. Ammonium urate urinary stones. *Urological Research*, 14: 315-318. doi: 10.1007/BF00262382
- Lourenço, R.A., Taniguchi, S., Da Silva, J., Costa-Gallotta, F.D. & Caruso-Bícego, M. 2021. Polycyclic aromatic hydrocarbons in marine mammals: a review and synthesis. *Marine Pollution Bulletin*, 171: 112699. doi: 10.1016/j.marpolbul.2021.112699
- Mattiucci, S., Nascetti, G., Dailey, M., Webb, S.C., Barros, N.B., Cianchi, R., et al. 2005. Evidence for a new species of *Anisakis* Dujardin, 1845: morphological description and genetic relationships between congeners (Nematoda: Anisakidae). *Systematic Parasitology*, 61: 157-171. doi: 10.1007/s11230-005-3158-2
- Naranjo-Ruiz, K.L. 2020. Poblaciones simpátricas de *Tursiops truncatus*: Delphinidae, presentes en el APFFLT: evaluación del uso de recursos ($\delta^{15}\text{N}$) y hábitat ($\delta^{13}\text{C}$). M.Sc. Thesis, Universidad Autónoma de Campeche, Campeche.
- Nollens, H.H., Venn-Watson, S., Gili, C. & McBain, J. 2018. Cetacean medicine. In: Gulland, F.M.D., Dierauf, L.A. & Whitman, K.L. (Eds.). *CRC handbook of marine mammal medicine*. CRC Press, Boca Raton, pp. 887-907.
- Perrin, W.F. & Geraci, J.R. 2009. Stranding. In: Perrin, W.F., Würsig, B. & Thewissen, J.G.M. (Eds.). *Encyclopedia of marine mammals*. Academic Press, London.
- Pugliares, K.R., Bogomolni, A., Touhey, K.M., Herzig, S.M., Harry, C.T. & Moore, M.J. 2007. *Marine mammal necropsy: an introductory guide for stranding responders and field biologists*. Woods Hole Oceanographic Institution. Technical Report, WHOI-2007-06.
- Quiñones, R., Giovannini, A., Raga, J.A. & Fernández, M. 2013. Intestinal helminth fauna of bottlenose dolphin *Tursiops truncatus* and common dolphin *Delphinus delphis* from the western Mediterranean. *Journal of Parasitology*, 99: 576-579. doi: 10.1645/GE-3165.1
- Radi, R., Turrens, J.F., Chang, L.Y., Bush, K.M., Crapo, J.D. & Freeman, B.A. 1991. Detection of catalase in rat heart mitochondria. *Journal of Biological and Chemical*, 266: 22028-22034. doi: 10.1016/S0021-9258(18)54740-2
- Reckendorf, A., Sierbert, U., Parmentier, E. & Das, K. 2010. Chemical pollution and diseases of marine mammals. In: Brennecke, D., Knickmeier, K., Pawliczka, I., Sierbert, U. & Wahlberg, M. (Eds.). *Marine mammals: a deep dive into the world of science*. Springer, Berlin.
- Rendón-von Osten, J., Ortiz-Arana, A., Guilhermino, L. & Soares, A.M.V.M. 2005. *In vivo* evaluation of three biomarkers in the mosquitofish (*Gambusia yucatana*) exposed to pesticides. *Chemosphere*, 58: 627-636. doi: 10.1016/j.chemosphere.2004.08.065
- Romero, M.A., Fernández, M., Dans, S.L., García, N.A., González, R. & Crespo, E.A. 2014. Gastrointestinal parasites of bottlenose dolphins *Tursiops truncatus* from the extreme southwestern Atlantic, with notes on diet composition. *Diseases of Aquatic Organisms*, 108: 61-70. doi: 10.3354/dao02700
- Romero-Tenorio, A. & Delgado-Estrella, A. 2015. Comparación del ritmo de crecimiento de las toninas (*Tursiops truncatus*) pertenecientes a tres localidades distintas. *Therya*, 6: 389-400. doi: 10.12933/therya-15-263
- Ryeng, K.A., Lakemeyer, J., Roller, M., Wohlsein, P. & Sieber, U. 2021. Pathological findings in bycaught harbor porpoises (*Phocoena phocoena*) from the coast of northern Norway. *Polar Biology*, 45: 45-57. doi: 10.1007/s00300-021-02970-w
- Shamsi, S., Spröhnle-Barrera, C. & Hossen, M.S. 2019. Occurrence of *Anisakis* spp. (Nematoda: Anisakidae) in a pygmy sperm whale *Kogia breviceps* (Cetacea: Kogiidae) in Australian waters. *Diseases of Aquatic Organisms*, 134: 65-74. doi: 10.3354/dao03360
- Simões-Lopes, P.C., Daura-Jorge, F.G., Lodi, L., Bezamat, C., Costa, A.P.B. & Wedekin, L.L. 2019. Bottlenose dolphin ecotypes of the western South Atlantic: the puzzle of habitats, coloration patterns and

- dorsal fin shapes. *Aquatic Biology*, 28: 101-111. doi: 10.3354/ab00712
- Tettenhorst, R.T. & Gherkin, R.E. 1999. X-ray powder diffraction data for ammonium hydrogen (acid) urate, $\text{NH}_4\text{C}_5\text{H}_3\text{N}_4\text{O}_3$. *Powder Diffraction*, 14: 305-307. doi: 10.31349/revmexfis.67.305
- Venn-Watson, S., Daniels, R. & Smith, C. 2012. Thirty-year retrospective evaluation of pneumonia in a bottlenose dolphin *Tursiops truncatus* population. *Diseases of Aquatic Organisms*, 99: 237-242. doi: 10.3354/dao02471
- Venn-Watson, S., Smith, C.R., Johnson, S., Daniels, R. & Townsend, F. 2010. Clinical relevance of urate nephrolithiasis in bottlenose dolphins *Tursiops truncatus*. *Diseases of Aquatic Organisms*, 89: 167-177. doi: 10.3354/dao02187
- Wells, R.S. & Scott, M.D. 2009. Common bottlenose dolphin. In: Perrin, W.F., Würsig, B. & Thewissen, J.G.M. (Eds.). *Encyclopedia of marine mammals*. Academic Press, London.
- World Health Organization & International Programme on Chemical Safety. 1993. *Biomarkers and risk assessment: concepts and principles*. United Nations Environment Programme/International Labour Organization/World Health Organization. World Health Organization, Ginebra.
- Würsig, B., Jefferson, T.A. & Schmidly, D. 2000. *Marine mammals of the Gulf of Mexico*. Texas A&M University Press, Texas.

Received: April 28, 2023; Accepted: September 21, 2023