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SCIENTIFIC EXPERIMENT TO EVALUATE DOLPHIN COW-CALF SEPARATION DURING PURSE SEINE FISHING OPERATIONS IN THE EASTERN TROPICAL PACIFIC OCEAN[1]

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PROJECT AIM

Despite a > 99% reduction in bycatch-related mortality in the Eastern Tropical Pacific (ETP) purse-seine tuna industry over the past three decades, eastern spinner dolphin (*Stenella longirostris orientalis*, hereafter referred to as spinner dolphins) and northeastern offshore pantropical spotted dolphin (*S. attenuata attenuata*, hereafter referred to as pantropical spotted dolphins) populations have not increased as projected and are still considered depleted under the Marine Mammal Protection Act[1,2]. This suggests that cryptic and unobserved sources of mortality may be occurring, preventing population recovery. For the past two decades, it has been postulated that one such source may be mother-calf separation during fishery interactions, leading to calf mortality[2-6]. The aim of this project is to use unmanned aerial vehicles (UAVs) to determine: (i) if mother-calf pairs become separated during chase, encirclement, backdown, and/or post-release "run" from the net; and (ii) if/how mother-calf separation may be affecting population growth. These results will help to inform population models and management and conservation actions for dolphins in the ETP.

BACKGROUND AND JUSTIFICATION

In the ETP, small pelagic dolphins (primarily spinner and pantropical spotted dolphins) and yellowfin tuna (*Thunnus albacares*) associate in large, multi-species groups. This association is likely driven by increased predator protection gained by dolphins and tuna in large groups, in combination with a shallow thermocline overlaying a thick oxygen minimum zone that restricts tuna to surface waters[7]. Since the 1950s, the ETP tuna purse-seine fishery has taken advantage of this association by setting nets on dolphins to catch the tuna schooling underneath them. After the enactment of practices to minimize dolphin bycatch (e.g., backdown procedures, Medina panels, high-intensity floodlights, swimmers to assist

[1] NOTE FROM THE SECRETARIAT: The project of scientific experiment described in this document is currently ongoing, under the supervision of the IATTC staff and with funding from the Mexican Alianza del Pacífico por el Atún Sustentable. The University of Alaska and AIMM were selected after a rigorous process of selection carried out by that staff between the candidates which had responded to the public call for applicants.

dolphins out of the net), dolphin mortality has dramatically decreased, from hundreds of thousands of dolphins killed each year during the 1960s to < 1,000 killed per year since the 1990s[1,2,4].

While direct mortality during net encirclement has decreased, the number of intentional sets on dolphins has not and it is estimated that any given dolphin is chased and encircled multiple times each year[1,2,8-10]. While recorded dolphin mortality is typically based on deaths observed during encirclement, mortality could also occur at other points of the fishing process, including the pre-capture chase and the post-release "run" from the net[2,3]. During any stage of fishery interactions, dolphins may suffer acute or chronic stressors leading to lethal or sublethal effects, impacting physiology, reproduction, and calf-rearing; all of these factors may limit population growth[3,8,11-13]. However, such cryptic factors may be unobserved, undetected, and unaccounted for.

One such cryptic factor is mother-calf separation during fishery interactions. This has been a long-standing hypothesis to explain the lack of recovery in spinner and pantropical spotted dolphins[1-4,6,10,14,15]. Observations that 75-95% of lactating females killed in nets were not accompanied by a calf (i.e., the "calf deficit") suggested that high calf mortality could be occurring as orphaned calves would be unlikely to survive[3,4]. It was estimated that unobserved calf mortality due to mother-calf separation during encirclement could increase overall calculated dolphin mortality rates by up to 15% per year[10]. However, this is likely an underestimate as mother-calf separation could also occur during the chase or during the post-release "run" from the net[3,4,6,10]. Although mother-calf separation during the chase has rarely been documented[5], there are indications it may occur.

As there is no place to "hide" in the ocean, cetacean mothers cannot cache their young for predator protection. Calves thus exhibit a following strategy for predator protection and remain close to their mothers, a strategy also exhibited by their herd-forming ungulate ancestors[6]. Calves travel close to their mothers in echelon (swimming alongside the mother) or infant (swimming underneath the mother) position[14,16-18]. During the first month of life, calves tend to swim primarily in echelon position. Infant position then predominates throughout the remaining period of calf dependency[11,16,18,19]. However, echelon swimming is likely to persist during high-speed travel[5].

These two positions (echelon and infant) are infant carrying behaviors that provide hydrodynamic benefits for calves while simultaneously decreasing the swimming efficiency of mothers[5,15,20-22]. Studies in bottlenose dolphins (*Tursiops truncatus*) have shown that calves swimming in echelon position (vs. independently) exhibit increased swim speeds, increased distance covered per stroke, and reduced stroke amplitude stroke[6]. Given the underdeveloped musculature and reduced swimming performance of calves, these attributes result in physical and energetic benefits for the calf that enable them to swim close to their mothers under natural conditions[6,14,17,20,23].

However, during the prolonged, high-speed travel typical during purse-seine fishery interactions, mother-calf formation swimming may break down. Mothers incur increased energetic expenditure and decreased swimming performance while carrying calves in both echelon and infant positions[20,23]. At very high speeds, the precise swimming needed to maintain the energetic benefits of drafting and the physical laws maintaining the echelon position may not be possible[5]. When calves lose the hydrodynamic benefits of drafting alongside their mothers, they cannot physically keep up with their mothers[5,6,14]. Due to the strong herd-conforming behaviors in pelagic dolphin pods, there are indications that mothers may maintain group conformity at the expense of abandoning their calves[5,6,20,24]. All of these lines of evidence indicate that mother-calf separation may occur during the chase.

Calves < 1 year old may be particularly vulnerable to mother-calf separation due to their underdeveloped physical, morphological, and physiological abilities[6,14,25,26]. Yet, reduced swimming performance may persist up to four years of age, prolonging the period of vulnerability to mother-calf separation[6]. This is potentially significant given that typical calf weaning age is two years in spinner dolphin calves and 3-7 years in pantropical spotted dolphins[11]. However, the chance of mortality diminishes after calves reach one year old[6]. Further, observations that mothers with calves > 1 year old remained together throughout multiple chases and encirclements indicates that separation between mothers and older calves may be unlikely, even during prolonged, very high-speed travel[27,28].

Perhaps the best evidence collected to date that mother-calf separation may occur comes from aerial photographs of spinner dolphins. This showed a calf of probable neonate size leaping alongside its mother during a high-speed chase, subsequently losing position alongside its mother, potentially leading to separation[5]. The underdeveloped myoglobin buffering capacity of spinner dolphin calves may prevent a calf from re-establishing echelon position once it breaks; this risk has been reported for spinner dolphins up to 1.6 years of age[26].

Previously, methodological limitations precluded collection of the fine-scale data required to determine if mother-calf separation occurs, and further, the number of observations required to yield the sample size needed to determine the probability of separation. These limitations may now be overcome by recent advancements in UAV technology which provide the ideal vantage point required to observe and track fine-scale changes in mother-calf spatial positioning without impacting the animals. The goal of this study is to: (i) use UAVs to determine if mother-calf separation occurs during fisheries interactions (including chase, encirclement, backdown, and post-release run); and (ii) determine if/how mother-calf separation may be affecting population growth. Our methods are based on our common dolphin (*Delphinus delphis*) research off Portugal in conjunction with the non-profit, Associação para Investigação do Meio Marinho (AIMM).

PREVIOUS RESEARCH

The south coast of Portugal is an important breeding and nursery ground for common dolphins[29,30]. Over the past eight years, our team has studied common dolphin mother-calf strategies. In particular, we developed a novel method for assessing mother-calf spatial positioning using UAVs[31,32]. This method is directly translatable to assessing mother-calf separation, and will be refined and improved upon during this study.

The first step in our prior study to assess mother-calf strategies using UAVs was to investigate the behavioral impacts of the UAV itself on small delphinids (bottlenose and common dolphins, the most abundant delphinids at our study site)[29] (Fig. 1). We considered an impact to be a change in: (i) direction, (ii) swimming speed, and/or (iii) dive pattern. We conducted random visual surveys aboard a 7-m rigid-hull inflatable boat RHIB to encounter either target species. We used a vertical take-off and landing (VTOL) quadcopter (DJI Phantom 2) paired with a GoPro HERO4 camera linked to a radio antenna system installed on the UAV to live-stream the in-flight video to a tablet.

The UAV was only launched during adequate environmental conditions (Beaufort sea state ≤ 3, < 0.5 m swell, visibility ≥ 5 km and skyline clear of clouds) and after a 15-min habituation period of the dolphin group to the research vessel. We flew the UAV at an initial height of 50 m above the group and lowered it 5 m every 30 s until reaching the minimum height of 5 m above sea level (Fig. 1). Bottlenose dolphins did not show any visible response to the UAV. The only significant change observed for common dolphins was a change in direction when the UAV was at a height of 5 m. With this study, we demonstrated that UAVs have a low impact on delphinid behavior and can therefore be considered an effective tool to study such species.

We then used a similar UAV system (DJI Phantom 2 and 4) to assess maternal strategies in common dolphins. We tested if/how the predation, infant safety, and scramble competition hypotheses influence mother-calf behavior. In brief, we found strong support for the predation and infant safety hypotheses and partial support for the scramble competition hypothesis[31].

We launched the UAV during common dolphin sightings with at least one mother-calf pair present and during adequate environmental conditions (see above). We considered calves to be animals ≤ ½ the length of an adult, traveling alongside an adult[29]. During a deployment, we flew the UAV as centered as possible above a group while maintaining an altitude ≥ 20 m and a distance of 30-100 m from the research vessel.

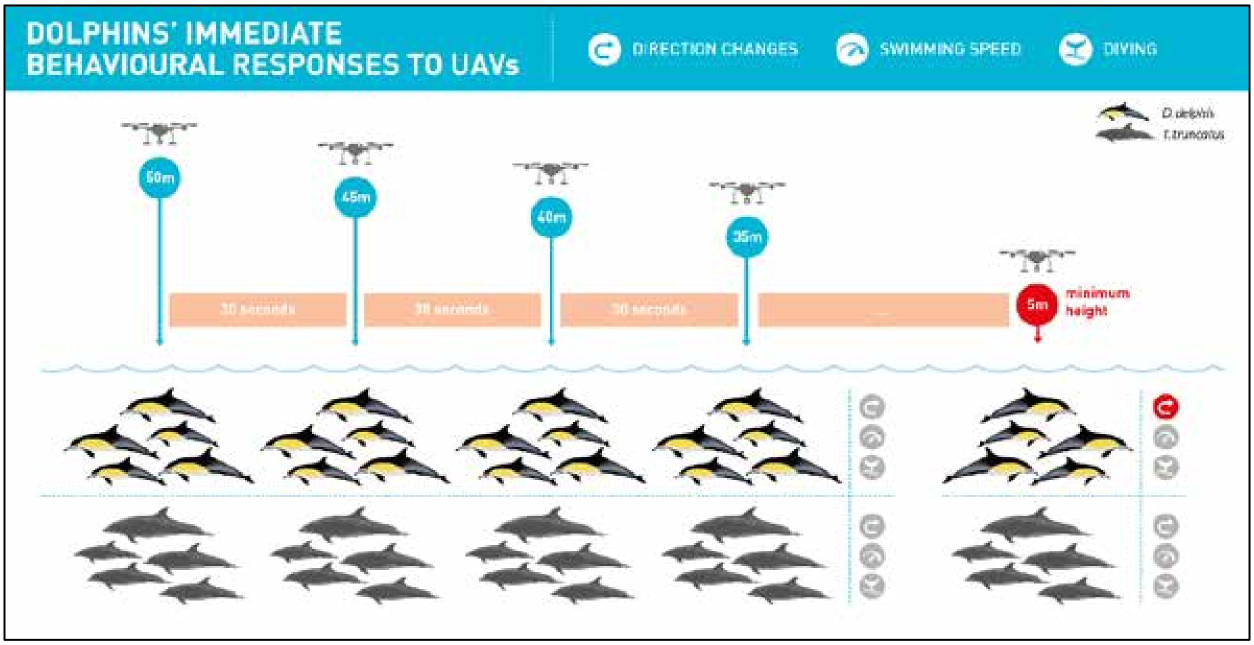

FIGURE 1. Graphical abstract of our study to test the impacts of UAVs on dolphins[32].

We conducted 39 UAV focal follows of common dolphin groups. Average flight time was 12.4 min, yielding an average of 7.8 min of usable video footage analyzed per follow. We analyzed all imagery post-hoc at 30 s intervals (N = 753 intervals totaling ~8 h of footage), recording group size, number of calves, spatial group formation (lone, parallel, linear vertical, pack, scatter, echelon), and group cohesion (very compact, compact, dispersed, very dispersed, lone) for each interval (Fig. 2). For each mother-calf pair, we documented their position within the group (front center, front edge, back center, back edge), and measured the distance between mothers and their calves and the distance between calves and their nearest neighbor that was not their mother. We used the average size of an adult common dolphin (2.0 m)[33] as our reference point for distance measurements. During each sampling interval, we also recorded the number of nursing/suckling events, leaps, belly-up contact, and socio-sexual events. To allow more precise observations of data extracted from the video, playback was slowed down as much as 10× the original speed. We ran Generalized Additive Mixed Models (GAMMs) and Generalized Estimating Equations (GEEs) in R (Vienna, Austria) to analyze, for example, the effect of group size, cohesion, formation, and group position on distance of a calf to its mother, and the effect of group size, cohesion, and nearest neighbor type on distance of a calf to its nearest neighbor that was not its mother.

The novel aerial perspective of using an UAV allowed us to collect these parameters in an effective and accurate manner, successfully overcoming the difficulty of assessing group configuration and inter-individual distances from a low-vantage, horizontal point of view as obtained from vessels.

The challenges we encountered during our data collection, as well as during analysis, included wind and swell (impacting UAV operation and water surface visibility), glare, dolphins' behavior, and limited battery life (< 25 min) that prompted us to re-launch the UAV multiple times during a sighting instead of continuously collecting data.

In the present study, we will strive to overcome the aforementioned challenges by using two cutting-edge UAVs (see below) with longer battery life, higher wind resistance, higher camera resolutions with infrared capabilities, and filters to improve image quality. These UAVs will allow us to image a wide field of view at high altitudes while preserving image quality to detect mother-calf pairs, even in large groups. We will test these UAVs to determine the optimal system(s) for our work.

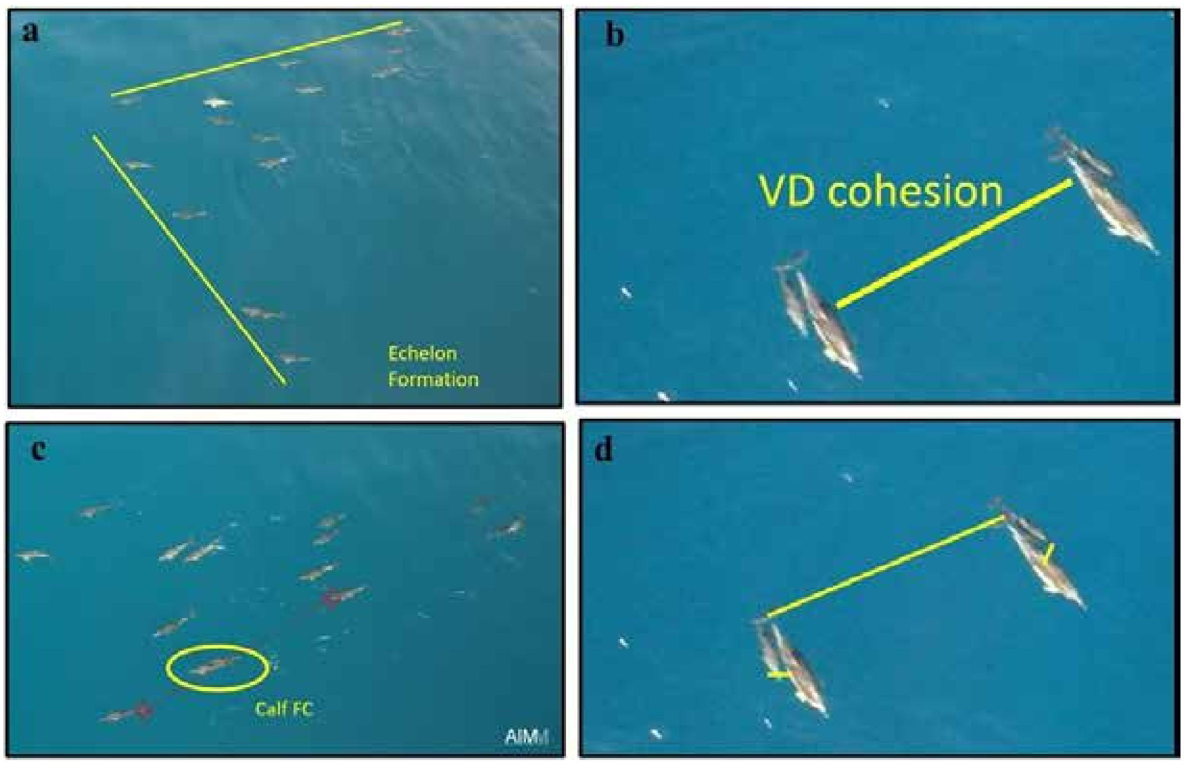

Fig. 2. UAV video stills showing metrics used to examine common dolphin maternal strategies off Portugal. a) group formation, b) cohesion (VD = very dispersed), c) calf position in the group (FC = front center), d) distance between mother and calf, and calf and its nearest non-mother neighbor.

Our experience with the above methodologies is readily transferable to the present study. In addition to having similar body lengths (within 0.5 m)[34], the behavioral ecology of common dolphins off the south of Portugal is broadly similar to the behavioral ecology of small oceanic dolphins in the ETP, which also includes common dolphins[1]. For example, common, pantropical spotted, and spinner dolphins may occur in mixed sex groups, or in groups segregated by sex and/or age class[29,31,35,36]. The societies of these three species of dolphins are characterized by a high degree of fission-fusion dynamics[1], with the fission-fusion rate for common dolphins in the south of Portugal being one of the highest reported in mammals[31]. All three species are known to travel long distances in short time spans[37] and traveling is one of the most common behaviors documented for *D. delphis* off the south of Portugal[31]. Generally, the diet of spinner, pantropical spotted and common dolphins is also comparable, their primary prey being small mesopelagic fishes and squid from the deep scattering layer, as well as epipelagic schooling species[38-40]. This dietary preference has also been documented for common dolphins off Portugal[41]. Finally, the reproductive biology is similar in all three species. Gestation periods range from approximately 10 to 11.7 months[40,42-

[45] and size at birth ranges from 75 to 93 cm[38-40]. To date, there have been no studies to investigate size at birth for common dolphins specifically in the south of Portugal. However, field observations match the size ranges reported in the literature and are thus comparable to newborn sizes of small oceanic delphinids in the ETP. Calving intervals and age at sexual maturity of the three species also fall within similar ranges[46].

However, we note some important differences between the systems. First, we anticipate that most groups targeted by the fishery will be mixed-species groups of spinner and pantropical spotted dolphins which are larger (median size = 200, CI = 112-334)[1] as compared to the single-species common dolphin group sizes (21.59 ± 14.95 individuals, range 1-69) we analyzed off Portugal via the above UAV methods[31]. Second, as larger dolphin groups carry more tuna, they are preferentially set on[9]. Third, the dolphins studied during the present project are expected to exhibit fast travel throughout the duration of the UAV observations. While traveling was also the predominant behavioral state in our study of common dolphin fission-fusion dynamics[30], we anticipate that the dolphins observed during the present study will predominantly be traveling at higher speeds than typically observed at our study site. This could complicate image analysis as fast-traveling dolphins typically produce a lot of white water, which could obscure observations of individual dolphins, particularly mother-calf pairs. Finally, during the present project, we anticipate needing to work under rougher sea conditions given the open ocean system and the need to conduct observations during as many fisheries sets as possible to obtain an adequate sample size. However, based on IATTC data (courtesy Ernesto Altamirano), 88% of searches by the Mexican fleet in Aug to mid-Nov during the years 2017 to 2022 occurred under Beaufort ≤ 3; this is the maximum Beaufort scale under which our previous research in Portugal occurred. Nonetheless, given these key differences, in addition to the inherent differences between working from a research vs. fishing vessel, we will first conduct a pilot study.

METHODS

We will conduct a two-phased pilot study, followed by the main study. The first pilot study phase will occur at our study site off the south of Portugal. The second pilot study phase, and the main study, will occur on purse-seiners in the ETP. We have all necessary research and UAV permits to conduct the pilot study in Portugal. We will work with the IATTC to obtain all necessary permits for the research and UAV operations occurring in the ETP. Additionally, we have already obtained approval from the University of Alaska Fairbanks Institutional Animal Care and Use Committee (IACUC) to conduct fieldwork in Portugal (protocol #1885382) and approval from the University of Alaska Fairbanks to collect fieldwork in the ETP is pending.

Pilot study

Phase 1: Portugal

The objectives of the first phase of our pilot study are to: i) test and become proficient with two new UAVs (Table 1), (ii) test UAV performance (e.g., with respect to battery life, wind), (iii) assess video quality under varying environmental conditions (e.g., wind, sun glare, sea state, water visibility), (iv) test the resolution of the visible light and infrared cameras at various heights to determine the maximum height at which we can fly the UAVs and still extract metrics for assessing mother-calf separation (Table 2), and (v) refine image analysis techniques.

Two UAVs models with complementary flight features (Table 1) will be used to facilitate successful implementation of our experiment. This will allow us flexibility in adjusting our methodology as needed according to field conditions and fishery phase while also ensuring continuity in sampling in case of loss/failure of one of the systems.

TABLE 1. Specifications of the two UAVs to be used.

Attribute	DJI Matrice 30T quadcopter	Autel Dragonfish Standard fixed-wing aircraft with L20T camera
Flight time (max)	41 min	93 min
Flight speed (max)	23 m/s	30 m/s
Wind resistance (max)	15 m/s	15 m/s
Transmission range (max)	15 km	50 km
Wide camera (visible light)	12 MP, 4K, 24 mm	12 MP, 4K
Zoom camera (visible light)	48 MP, 4K, 5-16x optical, 200x max. hybrid zoom	4K, 20x optical zoom, 240 max. hybrid zoom
Thermal camera	640x512, 40 mm	640x512, 25 mm, 16x zoom
Laser rangefinder	3m – 1200m ± 0.2 m	10m – 1200m ± 1m

TABLE 2. Metrics recorded from UAV video footage to assess mother-calf separation.

General Group Metrics	Specific Mother-Calf Metrics
Group size	Formation (mother-calf vs. non-mother-calf)
Composition (proportion of each species, proportion of mother-calf pairs)	Calf position relative to mother (echelon, infant, trailing behind, not closely associated with another non-calf)
Predominant behavioral state (forage, rest, socialize, travel)	Mother-calf distance, latitudinally and longitudinally ("slippage")
Group formation (lone, parallel, linear vertical, pack, scatter, echelon)	Distance between calf and its nearest non-mother neighbor
Cohesion (very compact, compact, dispersed, very dispersed, lone)	Calf position in the group (front center, front edge, back center, back edge)
Travel speed	Calf size (based on proportion of mother's body length)

The primary advantage of the Matrice is increased maneuverability via the multi-rotor system; however, this comes at the expense of battery life. The Dragonfish is capable of higher speeds, increased transmission range, and has more than double the battery life of the Matrice, but this comes at the expense of ease of maneuverability as it is a fixed-wing aircraft. However, the Dragonfish can operate as a multi-rotor (but this will decrease the battery life) with an easy and quick transition from fixed-wing to multi-rotor in flight. Both UAV models have VTOL capability that we consider critical features for this project. The Dragonfish also has an ADS-B receiver with an excellent obstacle avoidance feature that can easily inform other aircraft of its position. Importantly, both UAVs are equipped with wide-angle, high-resolution, 4K cameras with infrared sensors and laser rangefinders (Table 1) that will be ideal for observing fine-scale dynamics of mother-calf spatial positioning. On the Dragonfish, all three cameras (visible wide angle and zoom, infrared) record simultaneously, so even if we are zoomed into a particular part of the dolphin pod, the wide-angle camera will still be recording, thus increasing our ability to continuously monitor the entire pod.

Autel personnel will conduct an on-site multi-day training session on the Dragonfish at the start of the pilot study in Portugal. We do not anticipate needing formal training on the Matrice as our team has been flying similar DJI UAVs for > 6 years and we are quite proficient in their use.

The pilot study will coincide with our normal field operations where we conduct surveys from our 7-m RHIB 3-5 d/wk Apr-Nov, weather-permitting. For this pilot study, we will dedicate 30 d of field time during May-Jul. The two delphinids regularly occurring at our study site off the south of Portugal are common dolphins and bottlenose dolphins (*Tursiops truncatus*)[29,47]. To best mimic the groups we expect to observe in the ETP, we will seek out large groups of common dolphins. While our previous research focused on smaller (≤ 50) groups of common dolphins, groups up to 1,000 individuals do occur[29]. The predominant behavior of these large groups is fast-traveling (J. Castro, unpubl. data), which will help to simulate the behavior of ETP dolphin groups.

During both pilot study phases and the main study, the UAV crew will consist of three people: pilot, co-pilot, and visual observer. The co-pilot will assist the pilot with UAV take-off and landing. Once the UAV is airborne, the co-pilot and visual observer will help the pilot to navigate to the dolphin group. Once the group is visible on the UAV ground control station screen, the pilot will keep eyes on the screen to stay with the group. Using a second screen, the co-pilot will help the pilot to stay with the group and make decisions on which portion of the group to focus as needed (e.g., if the group splits or is larger than the desired field of view for sufficient image resolution). The visual observer will keep eyes on the UAV as much as possible, guiding the pilot as necessary.

Following previous methods[11,31], we will conduct a post-hoc examination of UAV video footage to record our metrics (Fig. 1, Table 2). We will note differences in our ability to assess these metrics according to: (i) UAV type (Matrice vs Dragonfish), (ii) camera (visible vs. infrared), (iii) UAV height, (iv) group size, (v) group behavior, (vi) group composition, and (vii) environmental conditions. Image analysis for both pilot study phases and the main study will be conducted using Image-Pro v. 11 (Media Cybernetics, Inc., Rockville, MD). We will explore the potential to customize this program with automatic/semi-automatic processing functions that will expedite analysis (Fig. 3).

Phase 2: ETP

The objectives of the second phase of the pilot study are to: (i) apply the UAV methods used during pilot study phase 1 to a purse seiner in the ETP; (ii) become familiar with fishery operations, the behavior of the dolphin species of interest, and working under various fishery and environmental conditions (e.g., Beaufort sea state, swell) in the ETP; (iii) determine the optimal UAV(s) and camera(s) for use during the main study (e.g., according to image resolution, battery duration); (iv) define mother-calf separation; (v) determine if mother-calf separation can be observed in real time; and (vi) obtain a preliminary estimate of mother-calf separation.

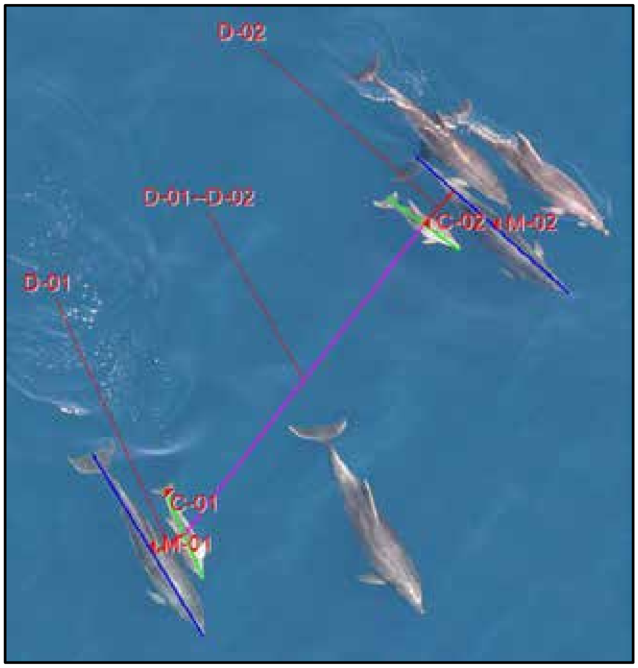

Fig. 3. Sample image showing an example of how Image-Pro v. 11 may be customized for UAV image analysis. After using the "Mother Line" (blue, M-01 & M-02) and "Calf Line" (green, C-01 & C-02) tools to measure and calculate calf size relative to its mother, distances between individuals can be calculated using "Distance between Centers" (red, mother-calf distance D-01 & D-02) and "Set to Set Distance" (magenta, calf-calf distance D01-D02) tools. Figure courtesy Matthew Batchelor, Alces Imaging and Automation, LLC. Image from https://www.onegreenplanet.org/wp content/ uploads/2014/08/bottlenosedolphin5_ fullsize1.jpg

Fieldwork for pilot study phase 2 will occur during a two-week period in Jul-Sep when the fishery reopens after the June closure. We will work closely with the IATTC to determine the optimal time for conducting the pilot study and to coordinate the logistics of getting our team to/from the purse seiner. We are aware of working around crew schedules and the difficulties associated with getting to/from shore during an active fishing season.

We will fly a UAV from the purse seiner whenever possible throughout all fishery interactions occurring during adequate environmental conditions. Based on IATTC data (courtesy Ernesto Altamirano) from the Mexican fleet from Oct to mid-Nov during the years 2017 to 2022, the modal number of chases per fishing trip was 41-50 and the average fishing trip duration was 51 d. We anticipate that during the 14-d pilot study, we will be able to image ≥ 10 chases, also considering limitations due to environmental conditions.

We will keep the UAV in visual line of sight as much as possible, understanding that the chase may start up to 2 nm from the vessel. We recognize that the UAV could interfere with fishery helicopter operations and will minimize the impact as much as possible. We will work closely with the helicopter and fishery crew to develop a detailed safety plan and a clear line of communication.

We will follow the UAV methods described above for pilot study phase 1. The live video feed from the UAV will be displayed on a second screen that the co-pilot will continuously monitor to help guide the pilot. One objective of pilot study phase 2 will be to determine if mother-calf separation events can be identified in real-time. This is important because it will help us to follow a separated calf for as long as possible to determine its fate, especially if it leaves the field of view of the main group being imaged. The aim will be to follow the calf until the group is encircled and beyond. This approach could help to determine if separated calves rejoin the pod if/when the chase circles back around, and if separated calves stay near the net after encirclement. If mother-calf separation is observed several times, a balance will be sought during subsequent chases between following the separated calf and resuming the follow over the main group to increase the possibility of observing more separation events, thus increasing sample size.

We will also determine if calf "slippage" events can be identified in real-time. An increase in the latitudinal and/or longitudinal distance between a mother and her calf can break down the hydrodynamic benefits afforded by echelon-position swimming, potentially leading to separation[5]. If a slippage event is observed, we will attempt to stay with that mother-calf pair for as long as possible to determine if slippage events lead to separation events.

We will aim to keep the entire group in the UAV's field of view whenever possible, particularly at the start of the chase to obtain an initial estimate of total group size including the total number of mother-calf pairs. However, if the UAV height necessary for the entire group to be in the field of view diminishes image resolution so that we cannot reliably record our metrics of interest (Table 2), then we will focus on mother-calf pairs. While pantropical spotted dolphins (the best-studied ETP dolphin species with respect to social structure) live in fission-fusion societies with most groups composed of all age-sex classes, there are indications that mother-calf pairs may preferentially associate with one another in subgroups[1]. If the entire pod cannot be kept in the field of view, we will also explore the utility of focusing on the back part of the pod to observe animals (potentially calves) falling behind. If a pod splits, the decision rule will be to remain with the pod or part of the pod that is being chased, all the way until net encirclement. Understanding the optimal balance between viewing the entire vs. part of the group according to UAV height and image resolution is a primary objective of the ETP pilot study.

Another primary objective of the ETP pilot study is to understand fishery phase duration with respect to UAV battery life. Throughout each fishery phase, we will develop a method for quickly and efficiently bringing the UAV back to the purse-seiner as needed to change batteries and fly back to the dolphin group. We will have multiple sets of fully charged Dragonfish and Matrice batteries ready to go on deck, in addition to having another fully charged Matrice ready to go. As another back-up, we will bring our currently-owned DJI Mavic quadcopter that will also be on deck charged and ready to go at all times.

According to IATTC data (courtesy Ernesto Altamirano), 98% of chases last ≤ 60 min with 55% lasting ≤ 15 min. Given the maximum Dragonfish battery life of 93 min (under ideal conditions using fixed-wing only and not multi-rotor) and considering flight time from the vessel to the start of the chase, we anticipate that for most chases, the entire chase may be imaged during one UAV flight.

A UAV will then be flown above the net during encirclement to monitor dolphins swimming outside the net; this may reveal if a calf stays near the net if its mother is encircled or vice versa. As this will occur in close range to the vessel and the UAV will likely need to hover above the net, we will use the Matrice during this period. Considering that modal encirclement duration is 31-40 min, with 96% of encirclements lasting ≤ 60 min (IATTC data, courtesy Ernesto Altamirano), and a maximum Matrice battery life of 41 min, we anticipate needing to return the UAV to the vessel to change batteries (or deploy a fresh UAV) one time during encirclement.

Finally, we will fly a UAV over dolphins during backdown to determine if mother-calf separation occurs during the post-release run from the net. Due to the high speeds and long distances dolphins are expected to travel during this time period, we will fly the Dragonfish over the group for as long as possible, considering the battery life needed to return the UAV to the vessel.

During post-hoc image analysis, a mother-calf pair will be identified as two dolphins swimming in closer proximity to each other than any other dolphin, with the smaller member of the pair being ≤ 0.75 the length of the larger member (the presumed mother)[11]. Calves at this body length are expected to be ≤ 1 year old (Table 3) and thus vulnerable to separation and subsequent mortality[6]. We will further classify calves as neonates, considered to be < 0.5 the length of the presumed mother, oftentimes with fetal folds or lines visible[16]. Neonates are likely the most vulnerable to separation and subsequent mortality due to their underdeveloped swimming abilities and heavy reliance on echelon position swimming which may not be possible during prolonged, high-speed travel[5,6]. Based on established methods[31,48], measurements of inter-individual distance will be made in terms of mother body lengths (Table 3). To account for changes in UAV altitude and angle throughout a sampling period, at each 30 s interval, we will standardize our measurements by measuring mother body length and using that as the reference point for identifying calves in that interval (Fig. 3). Whenever possible, we will also use known lengths of chase vessels in the field of view for determining dolphin body length; this will be particularly important for determining lengths (and therefore ages) of single individuals separated from the pod.

Table 3. Body length measurements used to calculate approximate calf size relative to mothers.

	Spinner dolphin	Pantropical spotted dolphin	Reference(s)
Adult female	171 cm	188 cm	[3,49]
Neonate *(proportion of mother body length)*	77 cm *(0.45)*	83 cm *(0.44)*	[49-51]
1-year old *(proportion of mother body length)*	128 cm *(0.75)*	130 cm *(0.69)*	[50]

The first step in post-hoc image analysis will be to watch all footage in its entirety (reducing playback speed as necessary) to detect mother-calf separation and/or slippage events. While we will attempt to view these events in real-time as described above, we recognize this may be challenging and that post-hoc identification of separation/slippage events may be more practical. Each sampling period (i.e., one fishery interaction) will be scored as Yes/No for mother-calf separation and/or slippage; if Yes, the timestamp for each separation/slippage event will be recorded. Next, we will re-watch the video footage to record our metrics of interest (Table 2) at 30 s intervals. If separation/slippage occurred during that sampling period, we may refine our sampling scheme to record our metrics of interest at shorter intervals to capture the fine-scale details preceding and following separation/slippage. We will note differences in our ability to reliably assess our metrics according to UAV, dolphin group, and environmental parameters.

We are particularly interested in determining the latitudinal and longitudinal distance between each mother and her calf to identify slippage (Fig. 4). If a calf is separated from its mother too far latitudinally, the calf no longer benefits from Bernoulli suction which pulls the calf towards its mother for a "free ride". If a calf separates from its mother too far longitudinally (ahead or behind her), the calf may get caught in its mother's wake zone and no longer benefit from her drafting force[5,52]. In each case, the calf may be unable to keep up with the group, potentially leading to separation. During the pilot study, we will work to identify possible mother-calf distance thresholds leading to observed and potential separation.

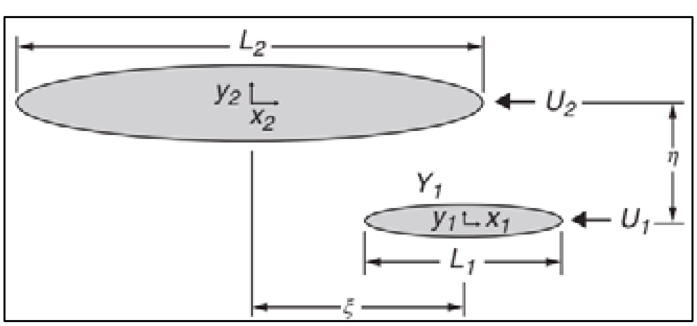

Fig. 4. We will measure the latitudinal distance between the midline of each mother to the midline of her calf (η), and the longitudinal distance between the midpoint of the mother to the midpoint of her calf (ξ). Figure from Weihs[5].

At the conclusion of pilot study phase 2, we aim to obtain a preliminary estimate of the probability of mother-calf separation. This will be based on the actual number of separation events observed, in addition to potential separation events based on observations of calf slippage.

To assess the probability of mother-calf separation, we will consider the: i) total no. mother-calf pairs in the initial group being chased; ii) total no. mother-calf pairs in the UAV's field of view during chase, encirclement, backdown, and post-release run; iii) no. calves observed separated during each phase or whenever the field of view changes; and iv) no. calves, and the relative age of each, that slip out of echelon position, thus representing potential separation.

For each fishery interaction (i.e., chase through post-release run or however long we are able to image the group), we will calculate the probability of mother-calf separation (S_{prob}) as:

$$S_{prob} = \frac{S_{obsv}}{MC_{obsv}}$$

where S_{obsv} = no. separation events observed and MC_{obsv} = no. mother-calf pairs observed in the field of view at the time of separation.

To account for changes in group size, and/or if it is not possible to keep the entire group in the UAV's field of view at all times, we will extrapolate as needed to an estimated total number of separation events ($\hat{S}$) using the following equation:

$$\hat{S} = S_{prob} * MC_{init}$$

where MC_{init} = total no. initial mother-calf pairs observed at the start of the chase. We will use the same formulae to calculate potential mother-calf separation based on observed and estimated calf slippage events. We will conduct exploratory analyses to identify which metrics predict separation or potential separation.

Main Study

Fieldwork for the main study will occur from Oct to mid-Nov. Based on the aforementioned IATTC data regarding average fishing trip duration (51 d) and modal number of chases per trip (41-50), we anticipate approximately one chase occurring per day. We will conduct UAV flights during as many chases as possible, considering the potential environmental limitations identified during pilot study phase 2.

The main study will follow directly from the results and lessons learned during the pilot study. We expect the field and image analysis methods used in the main study will closely follow those used in pilot study phase 2. However, we will follow an iterative, adaptive, and collaborative process to continually develop and refine our methods throughout the project, based on results of the pilot study, consultation with the IATTC, and feedback from the advisory board. It is possible the main study will include methods not described in this proposal. Our aim is to consider and incorporate all possible feedback so that we are fully prepared for successfully conducting the main study.

Data Analysis

Using the above formulae, we will calculate the average observed and potential separation rates based on all mother-calf pairs imaged during the main study. Following the analytic methods of Castro[31], we will run GAMMs and GEEs in R (Vienna, Austria) to identify which (if any) metrics (Table 2) predict mother-calf separation. Fishery interaction phase (i.e., chase, encirclement, backdown, post-release run), duration, and configuration (e.g., number of chase boats used) will also be considered as explanatory variables in the models. The probability of mother-calf separation will then be incorporated into population dynamics models to determine if/how mother-calf separation may be affecting population growth. As there have been no new population abundance estimates since 2006 (to our knowledge), we will likely rely on data from Gerrodette et al.[53] for our population models. We welcome the opportunity to collaborate with the IATTC to obtain more recent population data (if available) and to conduct population modeling in support of Obj. 2.

It is estimated that only 2-5 additional dolphin deaths per set, or 2-5 additional dolphin deaths per thousand chased, are needed to prevent the observed lack of recovery[10]. As discussed above, calves < 1 year of age are unlikely to survive if separated from their mothers, and the probability of mother-calf separation obtained during this study will likely be restricted to calves < 1 year old. Thus, even if very few mother-calf separations are observed during this project, it is possible the implications to the greater population could be large. This study aims to uncover if this potentially cryptic source of dolphin mortality is occurring, thus impacting the population growth and recovery of spinner and pantropical spotted dolphins in the ETP.

PERSONNEL

Our international, multi-lingual research team has strong expertise in dolphin behavioral ecology, UAV operation, and statistical analysis. The team will be composed of the following scientists, listed below according to their project role. In addition, project funds will support dedicated time for two additional AIMM researchers to assist with fieldwork and image/data analysis.

Heidi Pearson, PhD (PI), University of Alaska Southeast (UAS): Overall project supervision, coordination, and management, including budgeting; participation in pilot study fieldwork; interpretation, writing, and presentation of results

Joana Castro, PhD (Co-PI), AIMM: AIMM lead scientist; UAV co-pilot; participation in all fieldwork; UAV imagery analysis lead; data analysis; interpretation, writing, and presentation of results

André Cid, MSc (Co-I), AIMM: AIMM financial manager; fieldwork lead and UAV pilot; participation in all fieldwork; interpretation, writing, and presentation of results

Fábio Matos, PhD (Co-I), AIMM: Data analysis lead; interpretation, writing, and presentation of results

TIMELINE

This project will occur from 4/1/2023 to 8/31/2024. The timeline and milestones may be revised in consultation with IATTC. Dedicated personnel for each milestone are indicated in parentheses.

- Apr 1, 2023: Contract begins. Continued literature review, ordering of equipment and supplies, booking travel, team coordination and organization (Pearson, Castro, Cid, Matos)
- May 1, 2023: Submit updated research proposal to IATTC for internal review

- May 15-19, 2023: Presentation of research proposal and study design to the IATTC Scientific Advisory Committee (SAC), La Jolla, CA (Pearson, Castro)
- May-Jul, 2023: Pilot study – Phase 1, Portugal (Pearson, Castro, Cid, AIMM Research Assistant (RA) #1)
- Jun 30, 2023: Deadline to incorporate feedback from SAC on the initial proposal and re-submit to advisory committee
- Aug 2023: Pilot study – Phase 2, ETP (Pearson, Castro, Cid)
- Sep 2023: Analysis of pilot study data, revision of methods based on pilot studies (Pearson, Castro, Cid, Matos, AIMM RAs #1 and #2
- Sep 29, 2023: Submit report of pilot study (phases 1 and 2) and mid-term financial report (Pearson, Castro, Cid)
- Oct-Nov 2023: Main field study, ETP (Castro, Cid,AIMM RA #1)
- Jan-Apr 2024: Data analysis and report writing (Pearson, Castro, Cid, Matos, AIMM RAs #1 and #2)
- Mar 15, 2024: Submit progress report to IATTC for internal review
- May 2024: Presentation of final report to the IATTC SAC meeting, La Jolla, CA (Pearson, Castro, Cid)
- Aug 31, 2024: Submit final project and financial reports to IATTC (Pearson, Castro, Cid)

BUDGET

The estimated total cost of this project is $498,000 (Table 4). The costs below include a reduced UAS facilities and administration (F&A) rate of 20% applied to all UAS costs. There is no F&A for AIMM.

TABLE 4. Estimated budget by institution and budget category.

Institution	Category	Cost (USD)	Items
UAS	Personnel	62,614	Salary + benefits for 3.4 months of PI Pearson's time
	Travel	10,203	Round-trip travel (incl. airfare, lodging, per diem, and local transport) for PI Pearson from Juneau to: San Diego (x2), Lisbon, and Mexico (city TBD)
	Supplies	1,000	Camera and computer supplies
	Contractual	1,000	Medevac insurance and Merchant Mariner Credential fees
	F&A	14,963	
	Total UAS	**89,780**	
AIMM	Personnel	152,633	Salary for: Co-PI Castro (6 mo.), Co-I Matos (5 mo.), Co-I Cid (4 mo.), AIMM RAs (2 x 6 mo/ea)
	Travel	25,838	Round-trip travel (incl. airfare, lodging, per diem, local transport) for Co-PI Castro and Co-I Cid from Lisbon to: San Diego (x2) and Mexico (x2, city TBD), and AIMM RA Lisbon to Mexico (city TBD)
	Equipment	171,035	UAVs: 1x Autel Dragonfish Standard and 2x DJI Matrice 30T and associated supplies; DSLR cameras with telephoto lenses; image analysis software

	Supplies	23,714	Field laptops, vessel fuel, data storage, binoculars, action cameras
	Contractual	35,000	Software engineer, journal publication fees
	Total AIMM	**408,220**	
UAS +AIMM	**Grand total**	**498,000**	

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