ΝΟΤΕ

First identification of stereotyped whistle contour types by *Pseudorca crassidens*

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Individual recognition skills of conspecifics living in complex and fluid animal societies, such as that of delphinids, are important survival advantages, since they allow identifying potentially aggressive conspecifics, kin, mates, and allies (Bruck, 2013). Yet, these skills are the key to maintaining both social cohesion and hierarchy, since they allow reciprocal altruism and social threat assessment, in addition to providing reproductive advantages by avoiding inbreeding (Axelrod & Hamilton, 1981; Sayigh et al., 1995; Smuts et al., 1987).

Dolphins use personalized vocalizations, known as signature whistles (SW), which are emitted as repetitive patterns to transmit the senders' identity to their surroundings (Caldwell & Caldwell, 1965). They are individually distinct, frequency-modulated, narrow-band, defined as the predominant whistle contour produced when a dolphin is isolated from conspecifics. Moreover, they are developed by these animals during their first months of life, and they get crystallized overtime (Bruck et al., 2022; Caldwell & Caldwell, 1965; Janik & Sayigh, 2013).

Individual recognition mechanisms have played a key role in odontocete ecology. However, information about SW derives mostly, from the common bottlenose dolphins (*Tursiops truncatus*; Bruck et al., 2022; Caldwell & Caldwell, 1965; Caldwell et al., 1990; Gridley et al., 2014; Janik & Sayigh, 2013; Rio et al., 2022). In addition to bottlenose dolphins, individually distinctive SW have been identified and described for eight other delphinid species (Rio, 2023a), namely: Indo-Pacific bottlenose dolphins (*Tursiops aduncus*; Gridley et al., 2014), spinner dolphins (*Stenella longirostris*; Rio, 2023a), common dolphins (*Delphinus delphis*; Caldwell & Caldwell, 1968; Fearey et al., 2019), Atlantic spotted dolphins (*Stenella frontalis*; Caldwell & Caldwell, 1970), Pacific white-sided dolphins (*Sagmatias obliquidens*; Caldwell et al., 1973), Atlantic white-sided dolphins (*Lagenorhynchus acutus*; Cones et al., 2023), Pacific humpback dolphins (*Sousa chinensis*; van Parijs & Corkeron, 2001), and Guiana dolphins (*Sotalia guianensis*; de Figueiredo & Simão, 2009).

Knowledge about SW remains limited primarily due to the challenges in assessing certain cetacean species. This is the case for false killer whales (*Pseudorca crassidens*), who mostly live in deep waters (Rio, 2023b). False killer

whales are known to mostly emit relatively short-duration, low-frequency, and flat whistles that show a markedly narrow frequency range, sometimes with only a difference of 1.1 kHz (Murray et al., 1998; Oswald et al., 2003; Rio, 2023b; Thode et al., 2016; Weir et al., 2013).

The status of false killer whales according to the International Union for the Conservation of Nature (IUCN) is Near Threatened (Baird et al., 2018). Knowing a given species' individual recognition mechanisms is critical to understanding its cognitive abilities, vocal learning process and social structure, which it is highly relevant to cetacean conservation. Accordingly, the present study is the first to provide acoustic evidence that false killer whales produce stereotyped whistle contour types.

Acoustic recordings were obtained opportunistically from a coastal/inshore false killer whale group, living seasonally in Mexican Pacific waters, close to La Paz Bay (Figure 1) in the southwestern Gulf of California (Blanco-Jarvio et al., 2023). On April 10, 2023, a group of approximately 20 individuals, known to have been previously photographed in the same area (Figure 2) was sighted from a boat (with engines off). The group maintained constant swimming speed and direction (approximately 8 km/hr, heading west), traveling compactly. Although some individuals briefly separated from the group, to pass close the boat and the hydrophone, they immediately regrouped and displayed the same behavioral pattern. Underwater recordings were made with a SQ26-08 hydrophone (frequency range from 20 Hz to 50 kHz, and effective sensitivity of -169 dB, re 1 V/µPa; Cetacean Research Technology, Golden, CO). The device was placed 5 m below the surface; it was connected to a Zoom H1n digital recorder. Recordings were carried out at a 48-kHz sampling rate and 16-bit resolution.

Acoustic analysis was carried out according to the methodology by Rio et al. (2022). All whistles with good signal-to-noise (SNR) ratio, as well as complete and clear spectral contours, were first visually and aurally identified; then, they were manually selected for analysis purposes. Spectrograms were plotted in spectrogram view in Raven



FIGURE 1 Sightings and vocal recordings of false killer whales (*Pseudorca crassidens*) in La Paz Bay (LPB), Gulf of California, Mexico: the green circle highlights the location of the sighting/recording carried out on April 10, 2023. The yellow circle is the location of the sighting/recording performed on July 14, 2022.



FIGURE 2 False killer whales (*Pseudorca crassidens*) that were recaptured between the two sightings in La Paz Bay, Gulf of California, Mexico. In the top row, one finds individuals photographed on July 14, 2022; in the bottom row, one finds the same individuals photographed on April 10, 2023. The yellow circles indicate the notches confirming the recapture; they are the same animals.

Pro 1.6.1 at 1,024 Fast Fourier Transform (FFT), Hanning window, and 50% overlap. Spectral and temporal parameters were extracted: start (StaF), end (EndF), minimum (MinF) and maximum (MaxF) frequency, bandwidth (BanF), duration (Dur), interwhistle intervals (IWI), and number of inflection points (InfP).

Whistles with stable contours that have occurred repeatedly were classified as stereotyped whistles (STW). The remaining whistles, with varying contours, were classified as nonstereotyped whistles (NSW). The whistle emission sequence, and its IWI, were analyzed through the SIGnature IDentification (SIGID) method (Janik et al., 2013); possible signature whistles (PSW) were identified based on STW categories, with at least four whistles. Therefore, if one or more of the whistles occurred during the sequential bout analysis (75%), within 1–10 s of another whistle, it would be considered as the PSW type (Janik et al., 2013). Whistle classification was made by one experienced observer. All STW that did not pass the SIGID and NTW criteria were defined as non-SWs for the analysis.

Ten naïve independent observers (veterinary medical students), who had no previous experience with bioacoustics experiments, assessed a randomly chosen data subset to confirm the reliable identification of different PSW types. Each observer received five randomly chosen examples of all identified PSW types (n = 8; Cones et al., 2023; Janik, 1999; Sayigh et al., 2007). They were instructed to split them into groups of five, based on contour similarity; no further guidance was given.

In addition, the first author of this publication (R.R.), who created the STW catalog and classified the PSWs, selected three STW samples that were extracted from audio of underwater footage (GoPro Hero 7; 48-kHz sample rate and 16-bit resolution; in total, 64.5% (125/188) of whistles recorded within 4.3 min were classified as STW) that was recorded on July 14, 2022, for a short-term whistle stability assessment. These STW samples belonged to the group comprising some of the same individuals (confirmed dorsal fins Photo ID; Figure 2). They were also compared by naïve observers to identify the PSW types of false killer whales.

Descriptive statistical approach was applied to all frequency and temporal parameters. Emission rate was calculated by dividing the number of whistles by the number of minutes within the recorded whistle-time, which was defined as the time interval between the first and last registered acoustic signal (clicks or whistles).

In total, 410 whistles were extracted from the recorded whistle-time (10.1 min), and it led to emission rate of 40.6 whistles/min. Out of the 410 whistles, 69.0% (283) were classified as STW, 44.9% of them (127) met the SIGID bout criteria for bottlenose dolphin SW, and thus were classified as PSW. These repeated call types represented



FIGURE 3 Examples of whistle spectrograms of eight identified possible signature whistle (PSW) types (PSW1, PSW2, PSW3, PSW4, PSW5, PSW6, PSW7, and PSW8) emitted by a "Gulf of California population" of false killer whales (*Pseudorca crassidens*) living close to La Paz Bay, Mexico, according to the SIGID method. Frequency (kHz) is on the *y*-axis; it ranged from 0 to 15 kHz. Time (s) is on the *x*-axis. Scaling was the same for all items. Spectrogram settings: Fast Fourier Transform size = 1,024, Hanning window, overlap = 50%. The PSW number at the top of each panel represents the identified number for each PSW type.

31.0% (127/410) of all analyzed whistles, which were, subsequently, categorized into one of the eight PSW types. Figure 3 shows the spectrograms of all identified PSW types.

Temporal and spectral parameters recorded for each PSW type are displayed in Table 1. The number of samples per PSW type ranged from eight (PSW2 and PSW5) to 38 (PSW1). The recorded frequency parameters have shown mean MinF and MaxF of 2.54 ± 0.47 kHz (mean \pm standard deviation) (PSW6), and 10.56 ± 0.20 kHz (PSW7), respectively. Whistle duration (Dur) has shown mean values ranging from 0.47 ± 0.05 s (PSW2) to 1.04 ± 0.11 s (PSW4), with pooled mean value of 0.77 ± 0.20 kHz, for all PSW types.

The visual task showed a perfect match (100%) to the adopted classification created by the first author of the present publication (R.R.). Moreover, the three STW samples extracted from July 14, 2022, showed 100% overlap with two PSW types (two whistle samples with PSW5 and one with PSW7), and this is reinforced by the presence of shared individuals in both sightings. This finding is also consistent with the author's classifications.

In this study, we have shown the existence of stereotyped whistles in false killer whales in the Gulf of California. While we believe these may be SWs, we use the term "possible signature whistles (PSWs)" because the small acoustic data set does not allow us to rule out the possibility that the stereotyped acoustic signals identified were shared, repeated call types. Moreover, the SIGID method was developed for bottlenose dolphins; therefore, direct comparisons between these dolphins and false killer whales must consider the specificities of each species. Accordingly, false killer whale whistles are overall less complex in comparison to bottlenose dolphins (Thode et al., 2016; Weir et al., 2013), and this could affect the information conveyed through SW and their visual identification by human beings. False killer whale's behavior of congregating in many small subgroups, spread out over kilometers could also change the SW rate. However, there was always a seaplane flying in the area adjacent to the sightings during both encounters (15-km radius); in both cases, there was no record of other groups of false killer whales or other toothed whales present. Occasionally, false killer whales were observed accelerating and jumping out of the water, during the

		Frequency (k	(Hz)				Shape		
₽	2	Start (StaF)	End (EndF)	Minimum (MinF)	Maximum (MaxF)	Bandwidth (BanF)	Duration (s) (Dur)	Interwhistle (s) (IWI)	Inflection (n) (InfP)
PSW1	38	4.22 ± 0.53	5.86 ± 0.69	3.60 ± 0.23	5.90 ± 0.71	2.30 ± 0.64	0.78 ± 0.22	2.56 ± 2.20	2.68 ± 1.28
PSW2	œ	4.76 ± 0.43	9.31 ± 1.22	4.35 ± 0.04	9.56 ± 0.64	5.21 ± 0.65	0.47 ± 0.05	3.81 ± 1.72	0.25 ± 0.46
PSW3	13	4.25 ± 0.42	5.62 ± 0.49	4.25 ± 0.42	6.47 ± 0.12	2.22 ± 0.37	0.63 ± 0.13	2.30 ± 1.59	1.15 ± 0.38
PSW4	16	3.26 ± 0.21	4.98 ± 0.09	3.26 ± 0.21	5.02 ± 0.05	1.76 ± 0.22	1.04 ± 0.11	2.29 ± 0.78	0.13 ± 0.50
PSW5	8	7.02 ± 1.03	8.37 ± 1.29	5.18 ± 0.44	8.65 ± 1.06	3.47 ± 0.98	0.73 ± 0.24	1.01 ± 0.97	1.00 ± 0.00
PSW6	21	2.54 ± 0.47	9.03 ± 0.09	2.54 ± 0.47	9.06 ± 0.15	6.52 ± 0.46	0.76 ± 0.10	3.46 ± 2.82	0.05 ± 0.22
PSW7	13	4.17 ± 0.11	5.14 ± 1.74	4.17 ± 0.11	10.56 ± 0.20	6.40 ± 0.21	0.73 ± 0.06	1.81 ± 1.96	1.00 ± 0.00
PSW8	10	6.38 ± 0.33	4.45 ± 0.09	4.45 ± 0.09	6.38 ± 0.33	1.93 ± 0.33	0.80 ± 0.08	6.23 ± 2.61	0.00 ± 0.00
Total	127	4.20 ± 1.30	6.44 ± 1.85	3.72 ± 0.78	7.29 ± 1.92	3.57 ± 2.00	0.77 ± 0.20	2.87 ± 2.35	1.28 ± 1.36

TABLE 1 Mean \pm standard deviation recorded for acoustic parameters offset of eight possible signature whistle (PSW) types (n = 127) produced by false killer whales (Pseudorca crassidens) living in Mexican Pacific coastal waters, close to La Paz Bay. present study; however, it is unclear if they were feeding. This could be important to know, because such factors (e.g., environmental, social, behavioral, genetic, or cultural aspects) can increase acoustic emission rates and, consequently, SW production and rates (May-Collado & Wartzok, 2008; Quick & Janik, 2008). When dolphins are isolated from their conspecifics, for example, the SW rate in their acoustic repertoire can reach up to 100% (Caldwell et al., 1990; Janik & Slater, 1998; Sayigh et al., 2007).

Based on our results, we suggest that at least one third (31.0%) of coastal false killer whale whistles produced by free-ranging animals from La Paz Bay, Gulf of California, could be PSW. This could even be higher, if one takes into consideration that SIGID is a conservative criterion with success rate of 50%, and no possibility of showing false positives (Janik et al., 2013). This percentage may range from 38% to 70% among free-swimming bottlenose dolphins (Buckstaff, 2004; Cook et al., 2004; Janik & Sayigh, 2013; Watwood et al., 2005), which belong to the most assessed dolphin species.

The visual similarity in value judgment by naïve external observers has confirmed the herein adopted PSW classification type, with perfect overlap; this finding suggested short-term whistle stability for false killer whale PSW.

Overall, mean values recorded for whistle temporal and frequency parameters of false killer whales (non-SW, STW, and PSW) from the Mexican Pacific are similar to bioacoustics data observed in the few studies available in the literature about this species' vocalizations (Oswald et al., 2003, 2007; Rio, 2023b; Weir et al., 2013). False killer whales often produce relatively short-duration, little-modulation, low-frequency whistles (4–10 kHz), at markedly narrow frequency range (BandF; Murray et al., 1998; Oswald et al., 2003; Rio, 2023b; Thode et al., 2016; Weir et al., 2013).

It is important to continuously monitor the acoustics of this false killer whale population, in Mexico and other locations, to confirm our results and deepen knowledge about this species' individual recognition, cognitive skills, long-term stability, and other interesting SW aspects. Moreover, research about the individual recognition skills of these animals could provide valuable information for the yet limited understanding of this species' social structure (Kratofil et al., 2020; Martien et al., 2019), in La Paz Bay, especially because recent genetic evidence shows unique haplotypes for this population (Blanco-Jarvio et al., 2023).

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AUTHOR CONTRIBUTIONS

Raul Rio: Conceptualization; data curation; formal analysis; funding acquisition; investigation; methodology; project administration; resources; software; supervision; validation; visualization; writing – original draft; writing – review and editing. **Hiram Rosales-Nanduca:** Conceptualization; funding acquisition; investigation; methodology; project administration; resources; supervision; writing – review and editing.

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