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Aerial Vocalizations by Wild and Rehabilitating Mediterranean Monk Seals (*Monachus monachus*) in Greece

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Abstract

It is important that all aspects of the life history and behavior of Mediterranean monk seals (*Monachus monachus*) are documented to ensure effective management and conservation of this critically endangered species. Little is known about either airborne or underwater vocalizations for this species. Field research and rehabilitation of Mediterranean monk seal pups by MOm (MOm/Hellenic Society for the Study and Protection of the Monk Seal) provided opportunistic video recordings of the airborne sounds of this species. This study is the first to report acoustic features of airborne vocalizations from eight wild adult females and six of their pups, short-term recordings of a sick pup, and longer-term recordings of three rehabilitating pups. Although sample sizes are small, every effort was made to garner the most acoustic information from these recordings. The aims of this study were to document the acoustic properties and types of aerial vocalizations from wild and rehabilitating Mediterranean monk seals and to compare the acoustic features of their sound repertoire to the airborne vocalization characteristics of other monachids.

Audio-tracks were spectrographically analyzed using six frequency and five time variables with *Spectrogram* software. Through examination of real-time spectrograms and audibly distinguishable characteristics, this study classified four airborne vocalization types (bark, snort, scream, and chirp) for adult females. Three aerial vocalization types (bark, gaggle, and squawk) were documented from pups. The bark was the most common vocalization type from wild and rehabilitating pups. Wild adult female Mediterranean monk seal vocalizations ranged in frequency from 438 to 3,050 Hz and consisted of 1 to 6 components within a series with a total duration from 42 to 8,171 ms. Airborne vocalization types of rehabilitating pups ranged in frequency from 269 to 1,584 Hz and consisted of 1 to 11 components within a series with a total duration from 88 to 12,006 ms.

Using Pearson's correlation analyses, many of the frequency and time variables were highly correlated. Principle Component Analysis (PCA), using nine variables, produced a 3-factor model that explained 98.8% of the variability in the acoustic features of the whole repertoire. A more parsimonious, 3-factor PCA model, using only six variables, still explained a high percentage of variability (81.5%). These results indicated that collecting only two frequency measurements (beginning frequency and first harmonic interval) and four time measurements (number of components within a series, total duration, component duration, and the component interval) is sufficient to distinguish among species-specific vocalizations.

Results of this acoustical analysis are from a small number of Mediterranean monk seals, and further acoustic research is warranted to record adult males, and to increase the sample sizes of vocalizations from wild adult females and pups. In addition, more recordings of ill, stressed, and captive monk seals are needed. Lastly, vocalizations of wild Mediterranean monk seals should be studied at the other two main population sites (i.e., Archipelago of Madeira and Cabo Blanco region), at other times of the year, and outside of the breeding season to provide a better understanding of the overall vocal behavior of this species. The preliminary analyses presented herein hold promise that with sufficient data on acoustic features of airborne vocalizations researchers could acoustically monitor wild Mediterranean monk seals and determine their sex, pup age, and perhaps their health.

Key Words: Mediterranean monk seal, ontogeny, *Monachus monachus*, behavior, acoustic, monachid, vocalization

Audio Files: See www.aquaticmammalsjournal.org/index.php?option=com_content&view=article&id=533&Itemid=21 for .wav files of each Mediterranean monk seal vocalization type.

Introduction

Mediterranean monk seals (*Monachus monachus*) have been heavily exploited by humans for thousands of years (Johnson & Lavigne, 1999; Stringer et al., 2008), and the once continuous distribution of this species which ranged from the Black Sea throughout the Mediterranean Sea to the northwestern coasts and islands of North Africa has been fragmented into three isolated populations in the northeastern Mediterranean Sea, the Archipelago of Madeira, and the Cabo Blanco region in Morocco/Mauritania. Currently, fewer than 600 individuals are estimated to remain in the wild (Johnson et al., 2006). Mediterranean monk seals are listed as Critically Endangered by the International Union for Conservation of Nature (IUCN) (2010) and are considered now to be the most endangered pinniped in the world. They are threatened by habitat destruction and fragmentation, negative interactions with fisheries, pollution, and stochastic events, while lack of information regarding basic aspects of their biology is considered to hinder the development of effective conservation strategies (Johnson et al., 2006).

Vocalizations by pinnipeds are a natural biological signal that can convey the species, age, sex, and behavioral context of the caller. Recordings and analyses of phocid vocalizations have been used to document discrete breeding populations, diel activity patterns, the onset of breeding, territorial defense, and mother-pup interactions, as well as the age and sex of the caller (Bartholomew & Collias, 1961; Thomas & DeMaster, 1982; Thomas & Kuechle, 1982; Abgrall et al., 2003; Collins et al., 2005; Charrier & Harcourt, 2006). Analysis of vocalizations is particularly useful because these data can be collected remotely without disturbing the seals.

Mediterranean monk seals belong to the phocid subfamily Monachinae (Berta & Sumich, 1999). Social systems in monachids vary from solitary to seasonal monogamy to colonial polygyny (Stirling & Thomas, 2003). Underwater acoustic behavior of adult Antarctic monachid seals (Weddell seal [*Leptonychotes weddellii*], crabeater seal [*Lobodon carcinophagus*], Ross seal [*Ommatophoca rossii*], and leopard seal [*Hydrurga leptonyx*]) has been studied by several investigators (Thomas & Kuechle, 1982; Thomas & Golladay, 1995; Stirling & Thomas, 2003; Terhune et al., 2008; McCreery & Thomas, 2009). Typically, in-air vocalizations by Antarctic monachids are produced by mothers and pups during the nursing period to locate each other and establish a mother-pup bond (Thomas & Kuechle, 1982; Noe, 2001), while underwater vocalizations are produced by males to establish and defend aquatic territories and to attract mates. Similarly, Hawaiian monk seal (*Monachus schauinslandi*) females reside on beaches while nursing pups, but mating occurs in the water. In contrast, the monachid elephant seal (*Mirounga leonina* and *M. angustirostris*) has a harem mating system in which the male defends a group of females on the beach using a series of loud thumps; mating occurs on land, and mothers and pups exchange airborne vocalizations while hauled-out (Bartholomew & Collias, 1961; Hayes et al., 2004).

There is a paucity of publications on the airborne vocal communication in temperate phocids (Bartholomew & Collias, 1961; Van Parijs & Kovacs, 2002; Hayes et al., 2004; Sanvito et al., 2008). There are virtually no publications on aerial vocalizations by Arctic phocids, but there are a few publications on Antarctic phocids (Van Opzeeland et al., 2010). Among monk seals, the only existing study of airborne vocalizations was conducted on the Hawaiian monk seal by Job et al. (1995), and nothing is known about the acoustic behavior of either the Mediterranean or the Caribbean monk seal (*Monachus tropicalis*). Unfortunately, because of their extinction, nothing will ever be known about the vocal behavior of the Caribbean monk seal.

A hearing study on a captive Hawaiian monk seal indicated the best hearing range under water was from 12 to 28 kHz; and below 8 kHz, hearing was less sensitive than in other phocids tested. The best hearing range should be an indicator of the vocalization range of Hawaiian monk seals and perhaps other monk seals (Thomas et al., 1990; Thomas, 1991).

An understanding of the vocal behavior of Mediterranean monk seals may provide acoustic characteristics of the species, which could be monitored and assist in population studies. The aims of this study were to document the acoustic features and usage of aerial vocalizations by wild Mediterranean monk seal mothers and pups, as well as by rehabilitating pups, to determine the (1) frequency and time characteristics of the vocal repertoire typical for this species, (2) number of vocalization types for mothers and for pups, (3) acoustic properties of vocalization types, (4) acoustic differences between mother and pup vocalizations, (5) relative usage of vocalization types by mothers and pups, (6) individual or sex variability in vocalizations, and (7) vocal ontogeny from known-age pups. Based on studies of vocal behavior in other monachids, the anticipated results are that the species, age, and sex of Mediterranean monk seals can be distinguished from their vocalization features.

Materials and Methods

Field Study Site and Data Collection

Airborne vocalizations of wild Mediterranean monk seals were collected opportunistically by researchers of MOm/Hellenic Society for the Study and Protection of the Monk Seal during field work carried out from December 2003 through April 2009 throughout Greece. Data were collected on one channel of a handheld Sony video recorder (Model DCR TRV900E) with a linear frequency response up to 22 kHz. Recordings were made of eight different adult females and six of their pups while the monk seals were on the beach (either within or near the pupping cave) or as mothers were in the water vocalizing to their pups. (See map of five locations of recordings in Figure 1.) Adult females were identified from their external appearance (Samaranch & Gonzalez, 2000) and their proximity to a pup, while pups were designated based on their external appearance and small size (Dendrinos et al., 1999).

Figure 1. Map of Greece indicating the five locations of audio and video recordings of wild Mediterranean monk seals

MOm Rehabilitation Facility

Four orphaned pups, all 1 to 2 wks of age, were transported to the Monk Seal Rehabilitation Center of MOm on the island of Alonissos, Northern Sporades. One pup, "sick pup," died 2 d into treatment. During their rehabilitation, the other three pups were housed indoors in a small pool; the exterior wall of the rehabilitation facility was made of PVC. Keepers fed and cared for the pups on a daily basis. All three pups grew and were released into the wild at approximately 50 kg in weight. The approximate rehabilitation period for "Artemis" was 5 mo; for "Victoria," 3.5 mo; and for "Dimitris," 5 mo (Dendrinos et al., 2007a).

Airborne vocalizations and video recordings were collected during the daytime, using the same camera equipment as for wild monk seals. Recordings were made by leaving the video-camera on a tripod near the pups, and recordings were made when no researchers or keepers were present. Data on three pups (two females, Artemis and Victoria, and one male, Dimitris) were collected at least ½ h/wk throughout the entire rehabilitation process.

Two audio/video recordings were obtained from the sick pup. It is difficult to say with certainty to what extent the researchers affected the acoustic behavior of the rehabilitating monk seals while collecting these data. Because the three monk seal pups in rehabilitation increased in weight and eventually were released, their vocalizations likely were typical of wild pups; whereas the sounds from the sick pup could have been atypical or affected by illness or stress.

Acoustic Analyses

During analyses, the audio-track of the video data was digitized and analyzed using *Spectrogram* Version 16 software to investigate six frequency and five time variables for all vocalizations. Vocalization types of adult females and pups were assigned by audible discrimination and visual differences in spectrograms. To standardize analysis of a single sound with a series of sounds, frequency and time measurements were taken on the first component of each vocalization. For each vocalization, the measured time variables included whether a vocalization was single or produced in a series, number of components in a series (i.e., for a series of three barks the number of components was 3), the component duration (i.e., duration of the first element in a series), component interval (i.e., time between the first and second components), total vocalization duration, and vocalization rate (i.e., accelerating, retarding, or irregular). For each vocalization, the measured frequency variables included the beginning frequency, ending frequency, maximum frequency, minimum frequency, presence or absence of harmonics, and the first harmonic interval (Figure 2).

Statistical Analyses

Statistical analyses was conducted using *MYSTAT 12* software (SPSS, Chicago, IL USA). Descriptive statistics were calculated on all frequency and time variables for each vocalization type for eight adult females, six wild pups, one sick pup, and three rehabilitating pups. Normality and homogeneity of variance of each variable required for parametric tests was ensured by the Anderson-Darling Test, and an

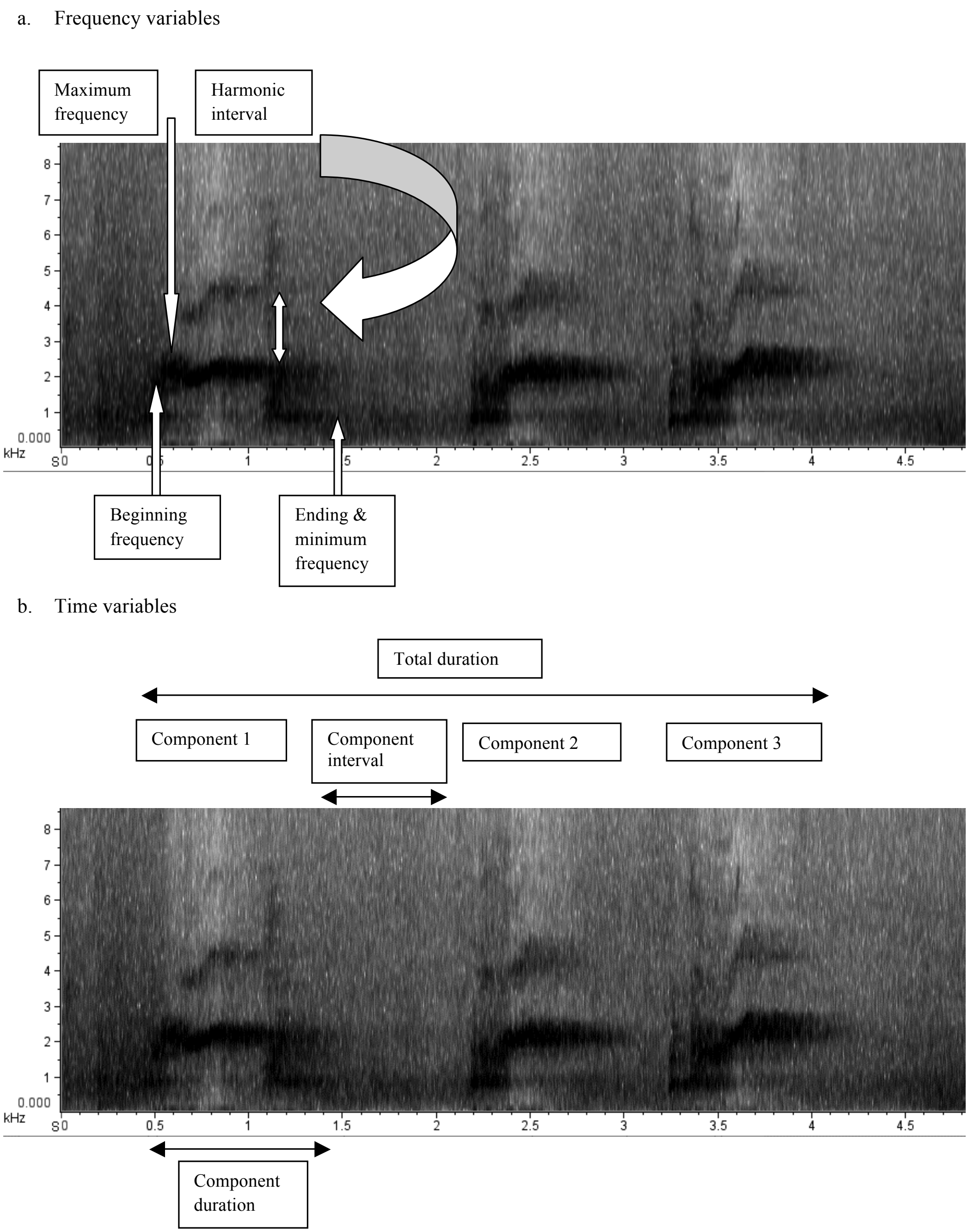


Figure 2. Spectrograms illustrating a. frequency variables and b. time variables measured on Mediterranean monk seal vocalizations

alpha level of 0.05 was used for all statistical tests. Pearson's correlation analyses were conducted on all acoustic variables to examine whether any variables were highly correlated and, therefore, could be eliminated from the analysis. Principle component analysis (PCA) was conducted for all frequency and time variables to examine the most influential variables in classifying vocalization types (Jolliffe, 2002). One-way ANOVAs were conducted on all frequency and time variables to examine possible age, sex, or individual differences in vocalizations. Nonparametric contingency table analyses were conducted on the usage rate of each vocalization type to examine possible age, sex, or individual differences among monk seals.

Results

Figure 1 illustrates the five different locations that eight wild adult female and six pups were audio- and videotaped (four in the Aegean Sea on the eastern side and one in the Ionian Sea on the western side of Greece). A total of 534 min of recordings were made of wild and rehabilitating monk seals. The duration of these recordings varied from 14 s to 160 min. Each recording of rehabilitating pups was 30 min in duration. Days recorded were 19 for Artemis, 12 for Dimitris, and 5 for Victoria. Two audio/videotapes of the sick pup were analyzed, and all of the vocalizations on 36 audio/video recordings (from wild, sick pup, and during rehabilitation of three pups) were analyzed. All vocalizations from the three rehabilitating pups were analyzed as they grew, so they should represent vocalization features of normal pups; however, vocalizations for the sick pup could reflect illness or stress.

Vocal Repertoire of the Species

Wild Mediterranean monk seals had four airborne call types. Three vocalization types were unique to adult females (scream, chirp, and snort) and one vocalization type (the bark) was shared by adult females and pups. Descriptive statistics for frequency and time variables of vocalizations of wild Mediterranean monk seals are shown by vocalization type in Tables 1 and 2.

Airborne vocalization of wild Mediterranean monk seals ranged in frequency from 438 to 3,050 Hz, varied from 1 to 6 components within a series, and had a total duration from 42 to 8,171 ms (see spectrograms for each vocalization type in Figure 3).

Rehabilitating pups had two unique airborne vocalization types (squawk and gaggle). They also produced barks similar to wild pups (see spectrogram of pup vocalization types in Figure 4). Descriptive statistics for frequency and time variables of airborne vocalizations by one male and two female rehabilitating Mediterranean monk seal pups

Table 1. Descriptive statistics for frequency variables (in Hz) measured on the first component of aerial vocalizations of wild Mediterranean monk seal females and their pups and rehabilitating pups. Note the small sample size of adult female barks; however, these were recorded at different locations and dates, so they are likely from different individuals.

Site	Seal age	Vocalization type	Statistic	Beginning	Ending	Maximum	Minimum	First harmonic interval
Wild	Adult	Bark	*n*	3	3	3	3	3
Wild	Adult	Bark	mean	1,449.1	1,530.2	1,601.6	1,441.5	1,717.2
Wild	Adult	Bark	SD	663.6	507.2	420.7	658.0	374.3
Wild	Adult	Scream	*n*	10	10	10	10	10
Wild	Adult	Scream	mean	1,975.1	1,826.6	2,095.3	1,699.2	617.6
Wild	Adult	Scream	SD	284.0	279.8	278.0	402.9	181.0
Wild	Adult	Chirp	*n*	19	19	19	19	5
Wild	Adult	Chirp	mean	1,963.5	1,952.2	2,118.6	1,799.4	2,187.9
Wild	Adult	Chirp	SD	317.6	372.3	351.2	318.4	113.3
Wild	Adult	Snort	*n*	1	1	1	1	NA
Wild	Adult	Snort	mean	1,236.1	1,236.1	1,236.1	1,236.1	NA
Wild	Adult	Snort	SD	0	0	0	0	NA
Wild	Pup	Bark	*n*	33	33	33	33	16
Wild	Pup	Bark	mean	1,212.6	1,208.3	1,241.2	1,192.5	839.1
Wild	Pup	Bark	SD	658.7	675.8	677.0	666.3	998.0
Rehab	Pup	Bark	*n*	95	95	95	95	93
Rehab	Pup	Bark	mean	1,177.1	1,194.2	1,216.6	1,169.8	242.9
Rehab	Pup	Bark	SD	279.0	276.4	275.5	274.3	260.2
Rehab	Pup	Squawk	*n*	6	6	6	6	6
Rehab	Pup	Squawk	mean	1,173.1	1,214.5	1,266.6	1,156.4	135.1
Rehab	Pup	Squawk	SD	335.9	385.8	409.3	325.2	27.7
Rehab	Pup	Gaggle	*n*	4	4	4	4	4
Rehab	Pup	Gaggle	mean	1,013.6	1,013.8	1,013.8	1,013.9	298.2
Rehab	Pup	Gaggle	SD	118.6	118.3	118.4	118.1	334.0

Table 2. Descriptive statistics for time variables measured (in ms) on the first component of aerial vocalizations of wild Mediterranean monk seal females and their pups and rehabilitating pups. Note the small sample size of adult female barks; however, these were recorded at different locations and dates, so they are likely from different individuals.

Site	Seal age	Vocalization type	Statistic	Number components	First component duration	First component interval	Total duration
Wild	Adult	Bark	*n*	2	3	2	3
Wild	Adult	Bark	mean	5.5	426.6	671.3	4,644.6
Wild	Adult	Bark	SD	0.7	204.2	169.0	4,048.8
Wild	Adult	Scream	*n*	5	10	5	10
Wild	Adult	Scream	mean	2.6	993.2	743.1	1,981.1
Wild	Adult	Scream	SD	0.5	1,138.2	319.0	1,361.9
Wild	Adult	Chirp	*n*	15	19	15	19
Wild	Adult	Chirp	mean	3.3	335.5	574.6	2,181.1
Wild	Adult	Chirp	SD	1.3	207.2	457.4	1,968.5
Wild	Adult	Snort	*n*	1	1	1	1
Wild	Adult	Snort	mean	2	355.5	206.7	1,072.2
Wild	Adult	Snort	SD	0	0	0	0.0
Wild	Pup	Bark	*n*	21	33	21	33
Wild	Pup	Bark	mean	2.8	148.6	713.8	1,351.1
Wild	Pup	Bark	SD	1.2	57.1	335.3	1,238.5
Rehab	Pup	Bark	*n*	95	95	56	95
Rehab	Pup	Bark	mean	2.5	367.2	782.1	2,086.8
Rehab	Pup	Bark	SD	2.9	164.4	418.0	2,362.7
Rehab	Pup	Squawk	*n*	6	6	1	6
Rehab	Pup	Squawk	mean	1.2	1,571.6	686.5	1,895.9
Rehab	Pup	Squawk	SD	0.4	630.5	0	1,242.3
Rehab	Pup	Gaggle	*n*	4	4	1	4
Rehab	Pup	Gaggle	mean	1.5	991.9	499.0	1,248.1
Rehab	Pup	Gaggle	SD	1.0	1,393.8	0	1,297.2

are presented in Tables 1 through 3. The frequency range of rehabilitating pup vocalizations was from 269 to 1,584 Hz, the number of sounds within a series varied from 1 to 11 components, and the total duration varied from 88 to 12,006 ms per series.

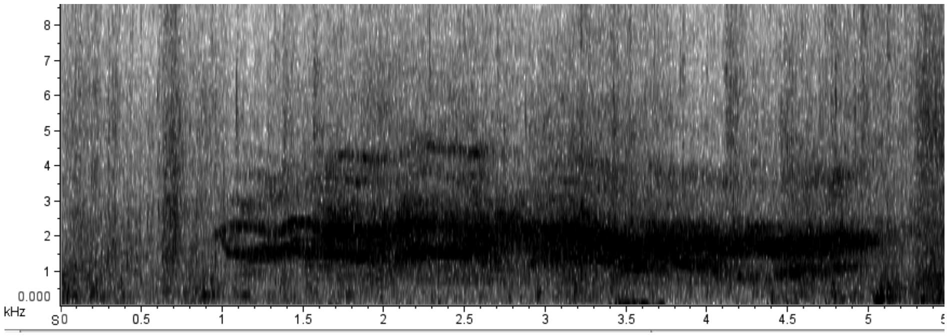
Using a one-way ANOVA showed that all frequency parameters varied significantly among the four vocalization types of wild monk seals: beginning frequency ($F_{(4,166)} = 23.31$, $p < 0.01$), maximum frequency ($F_{(4,166)} = 28.92$, $p < 0.01$), ending frequency ($F_{(4,166)} = 18.47$, $p < 0.01$), minimum frequency ($F_{(4,166)} = 12.86$, $p < 0.01$), and first harmonic interval ($F_{(4,166)} = 19.37$, $p < 0.01$). The only time variable which varied significantly among the four vocalization types of wild monk seals was component duration ($F_{(4,166)} = 29.00$, $p < 0.01$), with the bark having the shortest component duration of 142 ms from a single individual and the scream having the longest component duration of 2,018 ms from another individual.

Pearson's correlation analyses were used to examine the relationships among the six frequency variables and among the five time variables collected to describe the airborne vocal repertoire of the observed Mediterranean monk seal mothers and pups. Four frequency variables (beginning frequency, ending frequency, maximum frequency, and minimum frequency) were highly, positively correlated ($r = 0.97$, df = 65, $p < 0.01$) in both wild and rehabilitating monk seals. The first harmonic interval was not correlated with any other frequency variable.

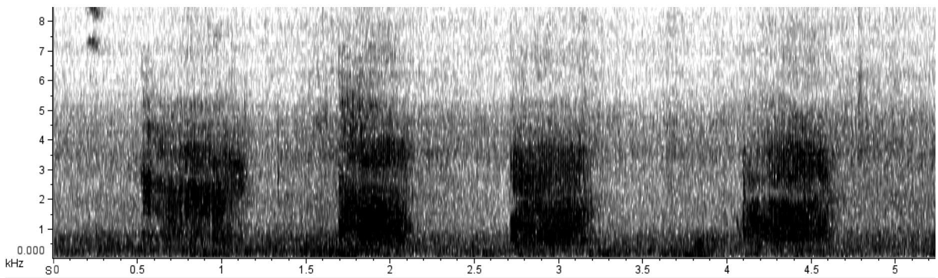
For time variables, Pearson's correlation analyses ($r = 0.89$, df = 65, $p < 0.01$) indicated that the number of components was positively correlated with duration, but component duration and component interval were negatively correlated in wild Mediterranean monk seals (females and their pups combined). In the three rehabilitating pups, the number of components and vocalization duration were positively correlated, but the number of components and component intervals were negatively correlated ($r = -0.62$, df = 61, $p < 0.01$).

PCAs were conducted to generate a model of frequency and time variables that best described the species' repertoire. Because many frequency and time variables were correlated, all combinations of including and excluding the correlated frequency and time variables of monk seal vocalizations were examined. The 3-factor PCA model

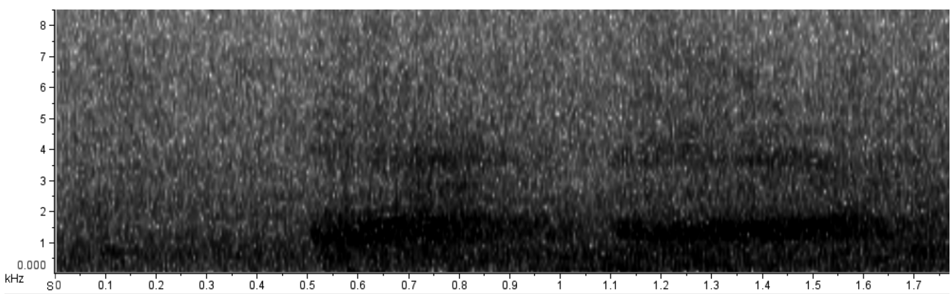
a. Adult female scream



b. Adult female bark



c. Adult female snort



d. Adult female chirp

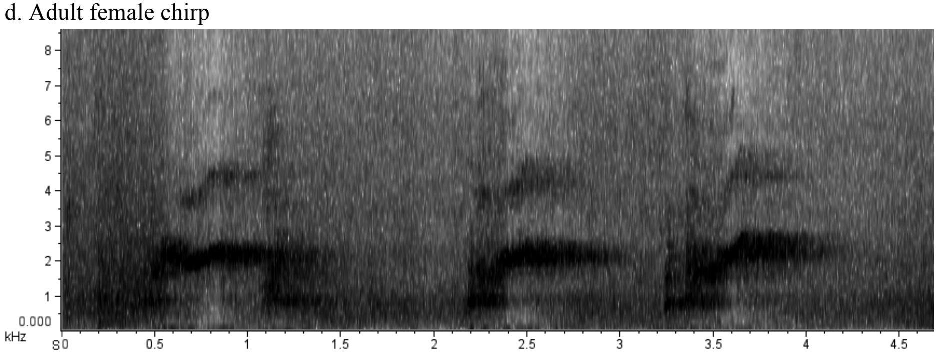


Figure 3. Spectrograms of airborne vocalizations from wild adult Mediterranean monk seals: a. scream, b. bark, c. snort, and d. chirp

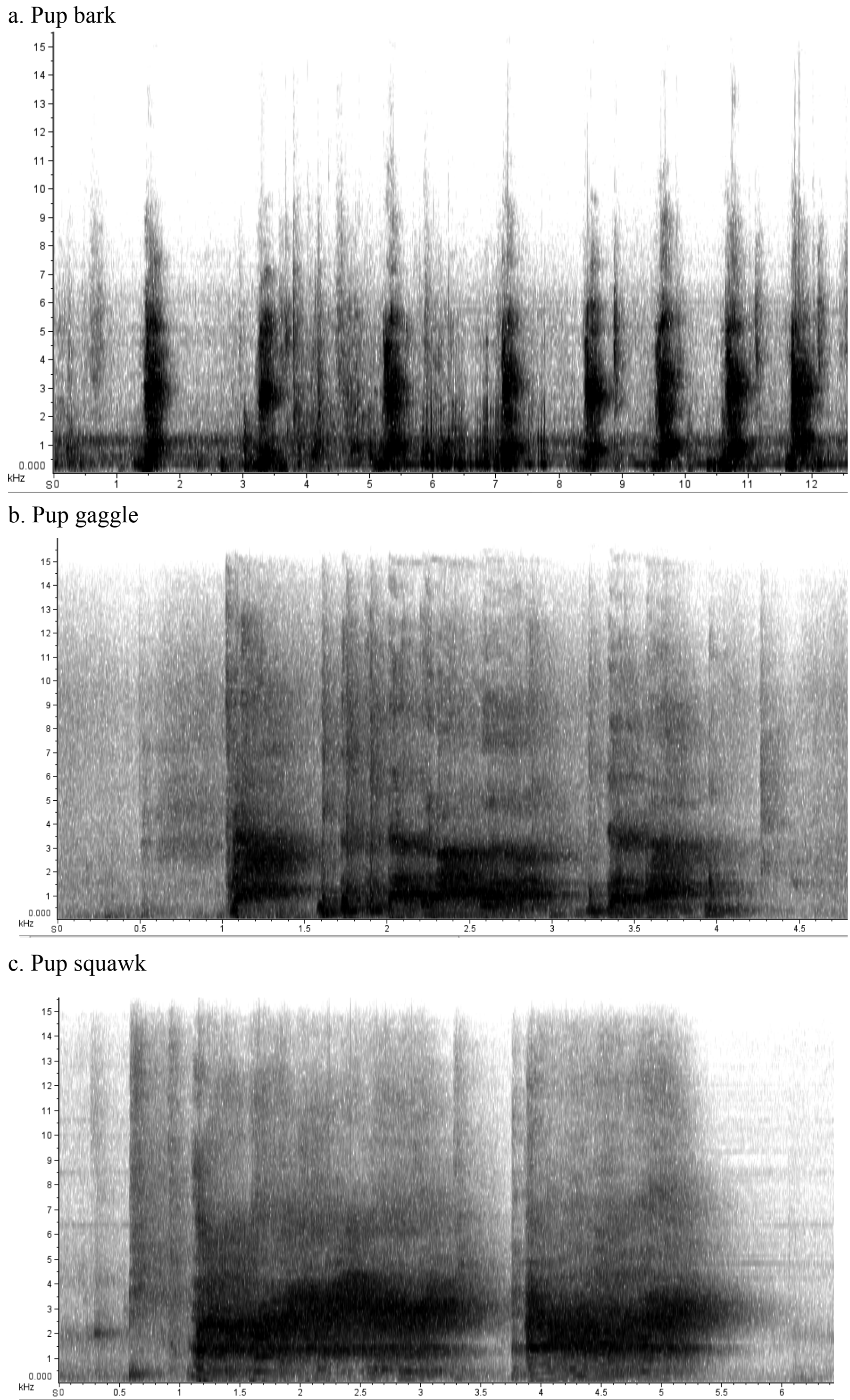


Figure 4. Spectrograms of airborne vocalizations from rehabilitating Mediterranean monk seal pups: a. bark, b. gaggle, and c. squawk

Table 3. Frequency and time features measured on the first component of vocalizations for each rehabilitating Mediterranean monk seal pup

		Frequency variables (Hz)					Time variables (ms)			
Pup	Statistic	Begin	Maximum	End	Minimum	First harmonic interval	Component number	Duration	Interval	Total duration
Artemis	*n*	49	49	49	49	49	49	49	26	49
	Minimum	269	269	313	313	14	1	119	209	196
	Maximum	1,506	1,584	1,584	1,472	1,041	11	2,373	2,000	11,052
	Mean	1,235.1	1,264.6	1,247.5	1,226.2	267.8	2.3	507.2	773.7	2,021.6
	SD	284.2	286.6	276.4	273.1	307.6	2.0	436.5	426.5	2,387.8
Dimitris	*n*	36	36	36	36	36	36	36	19	36
	Minimum	269	269	269	269	101	1	110	209	133
	Maximum	1,506	1,584	1,584	1,472	1,022	6	1,508	1,519	7,460
	Mean	1,207.5	1,248.0	1,231.8	1,200.9	223.5	2.1	425.1	806.8	1,759.9
	SD	266.1	264.8	269.1	264.1	237.1	1.4	266.8	426.2	1,738.1
Victoria	*n*	14	14	14	14	14	14	14	10	14
	Minimum	618	674	663	618	146	1	94	223	261
	Maximum	1,360	1,360	1,360	1,360	315	8	595	1,556	7,070
	Mean	980.7	1,059.3	1,011.2	976.7	193.7	3.2	305.1	723.0	2,358.1
	SD	217.2	240.2	232.7	217.7	44.5	2.1	129.4	427.4	1,933.7
Sick Pup	*n*	6	6	6	6	6	6	6	3	6
	Minimum	640	640	573	573	112	1	88	539	88
	Maximum	1,360	1,506	1,438	1,360	269	8	386	1,130	12,006
	Mean	1,082.5	1,125.5	1,084.3	1,062.0	161.0	2.7	194.6	769.6	2,735.1
	SD	245.6	282.1	283.4	266.0	59.0	2.7	102.5	316.1	4,617.7

which explained the highest amount of variation in the vocal repertoire (98.9%) of wild Mediterranean monk seals (females and pups combined) is presented in Table 4a. The 3-factor PCA model which explained the highest amount of vocal variation (83.21%) in the three rehabilitating pups is presented in Table 4b. Both of these 3-factor PCA models indicated that maximum frequency, ending frequency, beginning frequency, and minimum frequency variables had primary effects; the number of components, vocalization duration, and component duration were of secondary importance; and first harmonic interval and component interval variables were of tertiary importance in explaining the variability in the acoustic repertoire of Mediterranean monk seals. The PCA model for rehabilitating pups only differed in that component duration was added in factor 3.

To simplify analysis, a more parsimonious 3-factor PCA model was generated for all monk seal vocalizations. This simpler model included only six variables (two frequency variables and four time variables), rather than all nine variables, and this model still explained a high percentage of variability (81.5%). This simpler 3-factor model included the number of components, vocalization duration, and component interval in factor 1; first harmonic interval in factor 2; and first harmonic interval, beginning frequency, and component duration in factor 3 (Table 4c).

Table 4. Component loadings for the PCA of the acoustic variables from airborne vocalization of a. wild adult females and Mediterranean monk seal pups, b. rehabilitating pups, and c. the most parsimonious PCA model for all vocalizations. All Eigenvalues were ≥ 1; bold values indicate a significant variable in the PCA model.

a. Wild females and pups

Acoustic variables	Factor 1	Factor 2	Factor 3
Maximum frequency	**0.979**	0.093	0.126
Ending frequency	**0.972**	0.092	0.167
Beginning frequency	**0.949**	0.108	0.265
Minimum frequency	**0.941**	0.124	0.293
First harmonic interval	**0.848**	0.044	-0.317
Number of components	0.066	**-0.932**	0.148
Vocalization duration	-0.016	**-0.931**	0.323
Component duration	**0.492**	**-0.580**	-0.343
Component interval	**-0.494**	0.210	**0.767**

b. Rehabilitating pups

Acoustic variables	Factor 1	Factor 2	Factor 3
Maximum frequency	**0.98**	0.13	0.01
Beginning frequency	**0.98**	0.14	0.01
Minimum frequency	**0.98**	0.15	0.03
Ending frequency	**0.98**	0.16	0.04
Number of components	0.21	**-0.93**	-0.07
Vocalization duration	0.24	**-0.89**	-0.25
Component interval	-0.24	**0.79**	-0.10
First harmonic interval	0.09	-0.12	**0.72**
Component duration	0.16	0.22	**-0.64**

c. Wild females, wild pups, and rehabilitating pups

Acoustic variables	Factor 1	Factor 2	Factor 3
Number of components	**0.951**	0.134	-0.038
Vocalization duration	**0.907**	0.233	0.207
Component interval	**-0.819**	0.118	0.177
First harmonic interval	0.12	**-0.841**	**-0.067**
Beginning frequency	0.176	**-0.666**	**0.515**
Component duration	-0.099	0.249	**0.894**

Number of Vocalization Types

Wild Mediterranean monk seals produced four airborne vocalization types. Three vocalization types were unique to adult females (scream, chirp, and snort), while the bark was produced by both adult females and pups. Rehabilitating pups had two unique vocalization types (squawk and gaggle) that were not produced by either wild adult females or wild pups. The rehabilitating pups also produced barks that were similar to wild pup barks (Figure 4).

Acoustic Properties of Vocalization Types

Wild Seals—Descriptive statistics of frequency and time variables on the first component in vocalizations by wild Mediterranean monk seals are presented in Tables 1 and 2 by vocalization type. Vocalization of wild Mediterranean monk seals ranged in frequency from 438 to 3,050 Hz, varied from 1 to 6 components within a series, and had a total duration between 42 and 8,171 ms (see spectrograms for each vocalization type in Figure 3).

One-way ANOVAs indicated that all frequency parameters of the first component (beginning, ending, maximum, minimum, and first harmonic interval) varied significantly among the five vocalization types for wild seals (df = 166, $p < 0.01$). The only time variable that varied significantly among the first component of each vocalization type from wild monk seals was component duration, with the bark having the shortest first component duration of 142.69 ms from one monk seal and the scream having the longest first component duration of 2,018 ms from another monk seal.

Rehabilitating Pups—Descriptive statistics for frequency and time variables on the first component of vocalizations for rehabilitating Mediterranean monk seal pups are presented in Tables 1 through 3 by vocalization type. The frequency range of the first component in vocalizations by rehabilitating pups was from 269 to 1,584 Hz, the number of components within a series varied from 1 to 11, and the total duration varied from 88 to 12,006 ms. Four frequency parameters of the first component (beginning, ending, maximum, and minimum) varied significantly among the three vocalization types from rehabilitating monk seal pups (df = 103, $p < 0.01$); however, there was no significant difference in the first harmonic interval of the three vocalization types from rehabilitating pups. Component duration, number of components, total duration, and component interval varied significantly among the first components of the three vocalization types of rehabilitating monk seal pups (df = 13, $p < 0.01$). The number of components did not vary significantly among vocalization types in rehabilitating pups.

Acoustic Differences Between Mother and Pup Vocalizations

Analysis of age-related differences in vocalizations of wild adult females and pups was hampered by the small sample of only three barks by adult females and unbalanced sample sizes of vocalization types by the two age classes. However, using one-way ANOVAs indicated that age class (adult vs pup) of wild monk seals had a significant effect on all frequency variables. Adult females had a significantly higher beginning frequency ($F_{(2, 32)} = 26.88$, $p < 0.01$), higher maximum frequency ($F_{(2, 32)} = 34.66$, $p < 0.01$), and first harmonic intervals ($F_{(2, 32)} = 6.23$, $p = 0.03$) than wild pups. Wild pups had a significantly lower ending frequency ($F_{(2, 32)} = 22.93$, $p < 0.01$) and lower minimum frequency ($F_{(2, 32)} = 15.47$, $p < 0.01$) than adult females. Adult females had significantly longer component durations ($F_{(2, 32)} = 10.64$, $p < 0.01$), longer total durations ($F_{(2, 32)} = 5.19$, $p = 0.03$), and more components in a series ($F_{(2, 32)} = 9.12$, $p = 0.008$) than wild pups. Wild pups had significantly longer component intervals than adult females ($F_{(2, 32)} = 11.88$, $p < 0.01$).

Usage of Vocalization Types

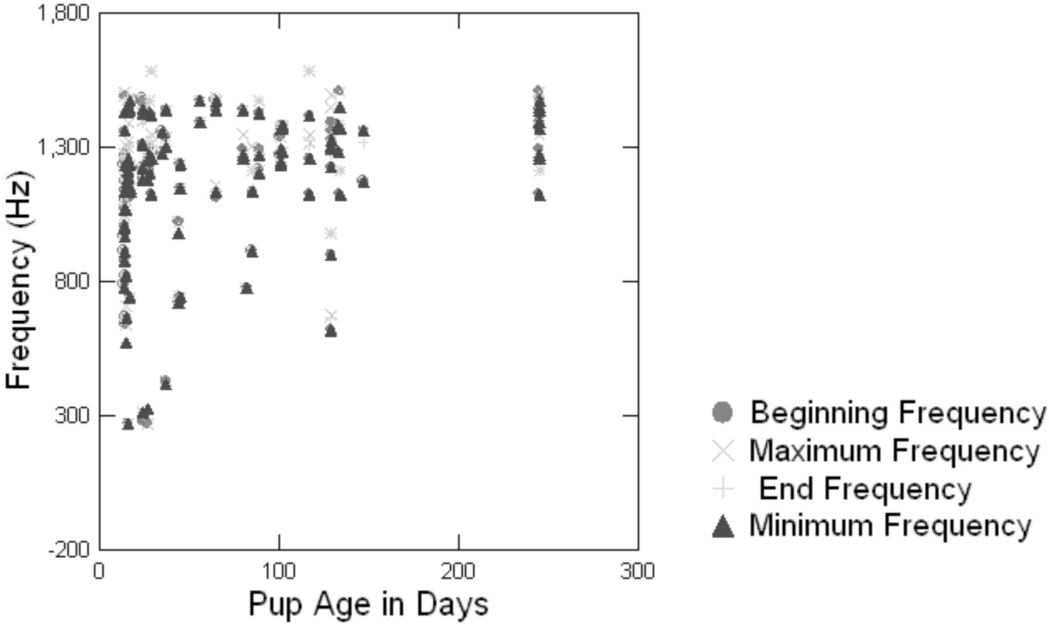
Contingency table analysis indicated that wild adult females used chirps and screams more often than wild pups (chirp: $\chi^2_{(1, 4)} = 23.33$, $p < 0.01$; scream: $\chi^2_{(1, 4)} = 17.13$, $p < 0.01$). There was no significant difference in usage of any vocalization type among wild adult females. The bark was the most common vocalization type used by both wild and rehabilitating pups. The bark was the only vocalization type used by wild pups, whereas the squawk, gaggle, and bark were used by rehabilitating pups. Because sample sizes were small, these results might not be indicative of typical vocalization types used by wild adult females or pups.

Acoustic Differences in Vocalizations of Individuals

Because wild adult females and pups were not marked, analysis of individual variation in these monk seals was not possible. Only individual differences in vocalizations were analyzed among the three rehabilitating pups. A one-way ANOVA showed that Victoria produced vocalizations with significantly different frequency structure than the other two pups (Figure 5): a larger first harmonic interval ($F_{(3,101)} = 4.83$, $p < 0.01$), a lower beginning frequency ($F_{(3,101)} = 3.65$, $p = 0.02$), a lower ending frequency ($F_{(3,101)} = 3.34$, $p = 0.02$), and a lower minimum frequency ($F_{(3,101)} = 3.75$, $p < 0.01$). Otherwise, there were no significant individual differences in a specific vocalization type for the other frequency or time variables.

A one-way ANOVA comparing the vocalization features of the male sick pup to Dimitris,

a. All rehabilitating pup vocalizations



b. Bark

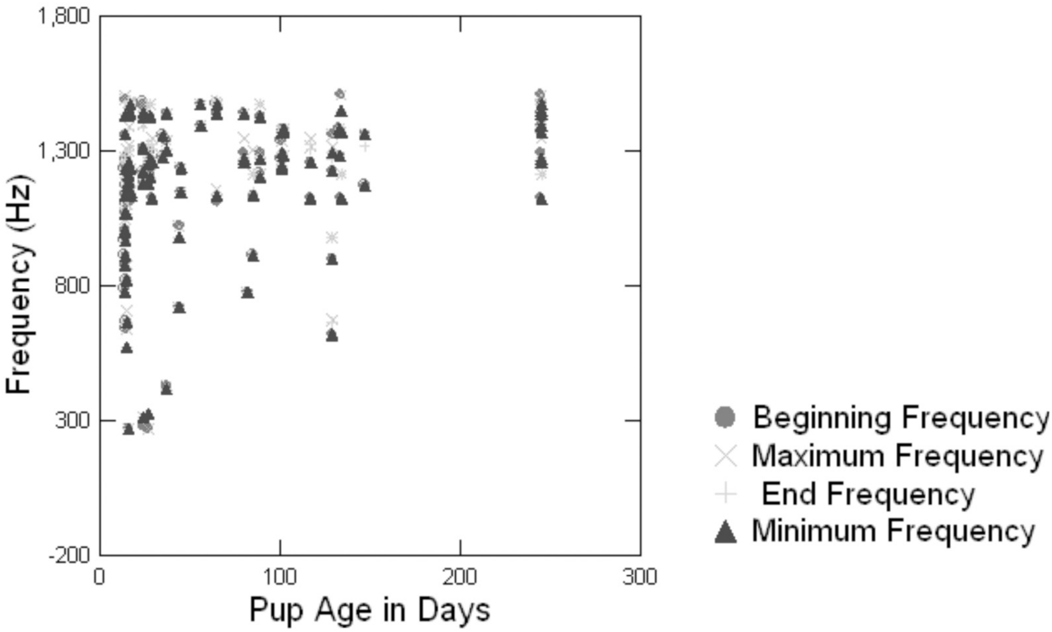


Figure 5. Frequency changes by age for three rehabilitating Mediterranean monk seal pups: a. all vocalizations and b. the bark.

the successfully rehabilitated male pup, showed that all frequency and time variables were significantly different between the two male pups (p = 0.000; see Table 3). The frequency structure of vocalizations by the sick pup were significantly different compared to sounds by Dimitris in having a larger first harmonic interval, lower beginning frequency, lower ending frequency, lower maximum frequency, and lower minimum frequency. The time structure of vocalizations by the sick pup was significantly different compared to sounds by Dimitris in having a greater number of components within a series, longer component durations, and longer total durations. In summary, the vocalizations by the male sick pup were lower in frequency, longer, and repeated more often in a series compared to the other male pup. These acoustic differences in vocalizations were possibly related to his poor physical state.

Sex Differences in Vocalizations

Because no recordings were made of adult males, it was not possible to examine sex differences between adult monk seals. Sex differences in vocalizations were analyzed only for two female and one male rehabilitating pup. One-way ANOVA tests demonstrated that the sex of the rehabilitating pup did not have a significant effect on any frequency or time variables for any vocalization type. However, a contingency table analysis showed that the sex of the rehabilitating pup had a significant effect on the usage of barks (χ^2 = 27.15, df =16, $p = 0.04$), with female pups producing more barks than the male pup. However, this sample size is too small to conclude that female pups always bark more often than male pups.

Ontogeny of Vocalizations of Known-Age Pups

Changes in frequency and time variables of rehabilitating pups were examined by pup age in days to determine whether vocalization features changed as pups grew older. For the two female rehabilitating pups, four acoustic variables were positively correlated with pup age: call duration ($r = 0.34$, df = 103, $p < 0.01$), maximum frequency ($r = 0.26$, df = 103, $p < 0.01$), ending frequency ($r = 0.26$, df = 103, $p < 0.01$), and first harmonic interval ($r = 0.31$, df = 103, $p < 0.01$). For the rehabilitating male monk seal pup, five acoustic variables were positively correlated with pup age: beginning frequency ($r = 0.41$, df = 103, $p < 0.01$), maximum frequency ($r = 0.35$, df = 103, $p < 0.01$), ending frequency ($r = 0.36$, df = 103, $p < 0.01$), minimum frequency ($r = 0.41$, df = 103, $p < 0.01$), and first harmonic interval ($r = 0.31$, df = 103, $p < 0.01$).

For both the female pups and the male pup, as they grew older, frequency variables became higher in frequency (Figure 6a); however, this was predominantly seen in the bark rather than in the squawk call type (Figure 7). In contrast, no time variables were correlated with pup age (Figure 6b).

Correlations were run between vocalization types and pup age in days across all three rehabilitating pups. The rate of the gaggle call type ($r = 0.58$, df = 103, $p < 0.01$) was positively correlated with pup age, and the rate of the bark call type ($r = -0.57$, df = 103, $p < 0.01$) was negatively correlated with pup age (i.e., older pups produced fewer barks).

Discussion

One reason for the paucity of literature on the vocal behavior in *Monachus* spp. is their low numbers and inaccessibility for long-term study and recordings. These monk seals typically inhabit remote, isolated locations, which would require long-term study at known sites. In contrast, the Antarctic monachids are numerous and accessible during the austral summer breeding season, and their distributions are known. The audio/video recordings of the airborne sounds of the wild and rehabilitating Mediterranean monk seals described in this study provided a unique opportunity to document the airborne acoustic repertoire of a few adult females and pups. Airborne recordings require being close to the monk seal with the appropriate recording equipment. Unless it is a specific goal of field researchers to study vocal behavior, appropriate recording equipment often is not available for opportunistic encounters with the elusive monk seals.

Parsimonious PCA Model

Most frequency and time variables were highly correlated in both wild adult females and pups and among rehabilitating Mediterranean monk seal pups. This is typical of many phocid vocalizations and indicates that, depending on the research topic, it may not be necessary to examine all the many frequency and time variables that can be measured on spectrograms to accurately describe vocalization types (Thomas & Golladay, 1995; Stacey, 2006; Clark, 2007). The goal of the PCA analyses was to recommend which of the many acoustic variables should be examined to efficiently distinguish vocalization type, age class, and/or sex of the caller. These results indicated that collecting only two frequency (i.e., beginning frequency and first harmonic interval) measurements and four time variables (i.e., number of components in a series, the duration of a vocalization series, vocalization duration, and the component interval) is sufficient to distinguish vocalization types for monk seals. In the comprehensive 3-factor PCA model using nine variables, which explained 98.9% of the variation, frequency variables were predominant in factor 1. It is interesting that the simplest 3-factor PCA model used only six variables and that time variables had a primary effect, whereas frequency variables had secondary and tertiary effects.

Possible Effects of Captivity on Acoustic Behavior

The effect of transport, captivity, and interactions with humans on behavioral research is a wide and popular criticism (Castellote & Fossa, 2006). At the L'Oceanográfic in Spain, Castellote & Fossa (2006) observed a persistent decrease in acoustic activity of beluga whales (*Delphinapterus leucas*) after air transportation to the facility, and again after the introduction of four harbor seals to the beluga whales' pool. Scarpaci et al. (2002) found that bottlenose dolphins (*Tursiops truncatus*) exhibit increased whistling in the presence of "swim-with-dolphin" tour operations.

In this Mediterranean monk seal study, more than half of the analyzed vocalizations were recorded from captive individuals. The authors cannot rule out that the captive environment affected the acoustic behavior of rehabilitating pups. However, the rehabilitating pups were not given ototoxic drugs nor were they sedated, so their hearing should have been normal. Mediterranean monk seals were the only species at the MOm rehabilitation facility, so there was no chance of recording sounds from other pinniped species or vocal behavior being affected by another species' sounds. The rehabilitating pups were housed indoors in a pool, and the exterior walls of the facility were of PVC. Reverberation of sounds was minimal, and no signs of echoes were detected in spectral analysis of the vocalizations.

All three rehabilitating pups steadily grew during their time at the MOm facility and eventually were released into the wild. All rehabilitating pups frequently barked, which could have been to elicit the keeper's attention. A similar repetition of barks by wild pups may occur to elicit a response from their mother. The fact that PCA produced similar 3-factor models for wild and captive seals supports the notion that captivity had little effect on vocal behavior, but sample sizes were too small to be sure.

Number of Vocalization Types

All monachids have a few airborne vocalizations used between mothers and pups. This study documented three airborne vocalization types by Mediterranean monk seal pups and four vocalization types by adult females. The gaggle was produced only once by a rehabilitating pup being fitted with a satellite tag, which was likely a stressful situation. The bark was the only vocalization type shared by both Mediterranean monk seal adult females and pups and by wild and rehabilitating pups. The bark was used between a mother and her pup, presumably to maintain the mother-pup bond. Mothers vocalized to their pup when they were in the water swimming near the pupping caves, while hauled-out on the beach, and while inside the cave with the pup. The captive monk seal pups barked at their keepers, presumably to get their attention and be fed.

There are few studies on airborne vocalizations in monachids. Noe (2001) and Collins et al. (2005, 2006) analyzed airborne vocalizations produced by Weddell seal pups and described a "primary" vocalization and a mother-pup "contact" vocalization. Job et al. (1995) analyzed the in-air vocalizations of Hawaiian monk seal mothers and pups and found pup vocalizations to be simple in structure with no varying types. Clearly, recording and analyzing the number of sound types, acoustic features, and behavioral contexts of airborne vocalizations in monachids is an important area for future research.

Frequency of Vocalizations

Mediterranean monk seals seem to have higher frequency airborne vocalizations compared to Weddell seals and Hawaiian monk seals. Vocalization of wild Mediterranean monk seals ranged in frequency from 438 to 3,050 Hz. The frequency range of rehabilitating pup vocalizations was from 269 to 1,584 Hz. Noe (2001) reported that Weddell seal pup airborne vocalizations had a mean maximum frequency of 719 Hz and a mean minimum frequency of 490 Hz. Collins et al. (2005, 2006) reported slightly lower frequency Weddell seal pup vocalizations: the primary pup vocalization had a mean maximum frequency of 386 Hz and a mean minimum frequency of 217 Hz. The Weddell seal pup contact vocalizations had a mean maximum frequency of 337 Hz and a mean minimum frequency of 189 Hz. These differences could stem from age differences in the pups examined in the two studies. In-air vocalizations of Hawaiian monk seal pups have a fundamental frequency of 99 Hz (Job et al., 1995). Alternatively, the differences between Hawaiian monk seal pup vocalizations described by Job et al. and Mediterranean monk seal pup sounds in this study could be due to age differences or environmental differences in wild vs captive animals.

Number of Components Within a Series

Noe (2001) found the number of vocalization components in Weddell seal pups ranged from 1 to 5, but the majority (71%) of vocalizations for both mothers and pups consisted of one component. Job et al. (1995) reported that Hawaiian monk seal pup vocalizations were simple in structure with no varying vocalization types. The number of components within a series by wild Mediterranean monk seals varied from 1 to 6, while the number of components in rehabilitating monk seal pups varied from 1 to 11. The number of components likely reflects motivation—that is, a pup that has lost contact with its mother will vocalize more often (Morton, 1977). The highest number of components was seen in the rehabilitating monk seal pups and may represent the absence of a mother, stress of captivity, or trying to get the attention of a caretaker.

Many phocids produce a long series of repeated elements with an increasing repetition rate, especially during aggressive encounters (Thomas & Kuechle, 1982; Noe, 2001; Sanvito et al., 2008). In the recordings of rehabilitating Mediterranean monk pups, the number of components and component intervals often were negatively correlated, which indicates that repeated vocalizations generally had an accelerating repetition rate. In addition, the number of components and the vocalization duration were positively correlated, whereas component interval and vocalization duration were negatively correlated. In other words, as the duration of a rehabilitating pup vocalizations increased, the number of elements in the vocalizations was greater, and the vocalizations typically had an accelerating repetition rate. This tendency to produce long, accelerating vocalizations could be an artifact of captivity where a pup is vocalizing to get the keeper's attention as it might vocalize in the wild to get its mother's attention.

Vocalization Duration

The large number of components in vocalizations by rehabilitating monk seal pups resulted in long duration vocalizations. This may have been related to their captivity—that is, vocalizing to the keeper—or it could be an attempt to contact a mother as the pup would do in the wild. In contrast, the duration of wild monk seal pup airborne vocalizations had fewer components within a series and shorter total durations. The total duration and number of components of wild adult females and pups was comparable to that of other monachids. According to Collins et al. (2005, 2006), the mean duration for a single Weddell seal pup primary vocalization was 612 ms and for a single pup contact vocalization was 1,275 ms. The duration of a single Hawaiian monk seal pup vocalization was 665 ms (Job et al., 1995).

Individual Differences in Pup Vocalizations

The authors are not aware of other studies on airborne vocalizations of monachid seals which examined individual variability in pup sounds, especially over a long timeframe. Overall, individual rehabilitating Mediterranean monk seal pups had similar frequency and time values for all acoustic variables at a given age. Victoria had slightly lower frequency vocalizations compared to the other pups; however, with such a small sample size, it is unknown whether this result is due to an individual difference among animals or a difference between females and a male pup. At the end of his 144 d in captivity and while being fitted with a satellite tag (Dendrinos et al., 2007a), Dimitris simultaneously produced a loud slapping noise with his flippers and vocalized a new vocalization type, the gaggle. This vocalization type had many short bursts in series. It is difficult to know whether this new vocalization type was used only by a male, by an agitated monk seal, or was unique to Dimitris.

The male sick pup died after 2 d of treatment and produced vocalizations with a larger first harmonic interval, a lower beginning frequency, a lower ending frequency, a lower minimum frequency, and slightly longer component durations than the three other rehabilitating pups. These acoustic differences in vocalizations by "sick pup" were possibly related to his poor physical state. Other studies have shown that ill or stressed dolphins produce different frequency, contour, and time characteristics in their whistles. Riess et al. (2009) reported that several dolphin species produced a unique whistle when ill or in rehabilitation. Therien et al. (in press) documented a unique whistle from a captive, male bottlenose dolphin (*Tursiops truncatus*) produced 2 d before his death. As a result, acoustic features of Mediterranean monk seal vocalizations could be used as an indicator of an individual's health status, but more data are needed on this topic.

Sex Differences in Pup Vocalizations

There are notable differences in the airborne vocal behavior of adult male and female elephant seals (Bartholomew & Collias, 1961; Hayes et al., 2004); however, the authors are not aware of any other studies that documented differences in airborne vocalizations in phocids by sex. Differences in airborne vocalizations by sex in adult Mediterranean monk seals could not be examined because recordings of adult males were not available. Sample sizes for analysis of sex differences among vocalizations in the rehabilitated pups were small: two females and one male. Results presented herein suggest that the sex of the rehabilitating pups did not have a significant effect on any frequency or time variables in any vocalization types; however, more data from a greater number of both sexes of pups is required to fully address this question. Still, both rehabilitating female pups barked more often than the male pup.

Vocal Ontogeny in Pups

Another unique feature about data presented herein is that recordings were made of known-age rehabilitating Mediterranean monk seal pups. This information provided the opportunity to examine whether vocalization features could be used as an age determination method for wild pups. The single male and two female pups all produced vocalizations with maximum frequency, end frequency, and a first harmonic interval that were positively correlated with age in days. Therefore, these frequency variables might be a good indicator of age in wild Mediterranean monk seal pups, but sample sizes were very small.

In mammals, as the vocal tract and body size grow, juvenile vocalizations typically decrease in frequency and eventually achieve the adult vocalization range (Sebeok, 1977). Most monachid species (elephant seals [Sanvito et al., 2008]; Weddell seals [Noe, 2001]; and, to a lesser extent, Hawaiian monk seals [Job et al., 1995]) conform to this trend. However, in Mediterranean monk seal pups, the barks increased in frequency with age to approximate the adult vocalization. This seems counterintuitive relative to growth and vocal tract trends, although to an experienced listener, Mediterranean monk seal mother and pup vocalizations do seem higher in frequency (pitch to the human ear) than in Weddell seals, another monachid, studied by Noe (2001) and J. A. Thomas (pers. obs., 2010).

Frequency and time properties of vocalizations for each rehabilitating pup were similar when compared at the same age in days. The frequency of Artemis's vocalizations changed at the very end of her time in captivity (107 d), but there was no statistical significance to this change compared to the two other rehabilitating pups. Perhaps pups were released back into the wild before any major changes in physical appearance or maturity was reflected in their vocalization structure. The older captive Mediterranean monk seal pups barked less often, which could either indicate a feature typical of maturation or that the pups were adapting to captivity.

Conclusions: Management and Conservation Implications

Vocal behavior is a natural signal that can provide information about the species, sex, age, and behavior of monk seals, perhaps without even seeing the animal. Since the critically endangered Mediterranean monk seal gives birth almost exclusively in coastal caves, it is imperative that pupping sites be identified and protected to ensure the survival of the species (Dendrinos et al., 2007c). Remote, automated sensing of vocal behavior in these elusive pinnipeds is imperative for a better understanding and protection of the species and should be handled as non-invasively as possible.

Despite substantial logistic difficulties, Dendrinos et al. (2007b), Gücü (2009), and Karamanlidis et al. (2010) monitored critical monk seal habitat in the eastern Mediterranean Sea using video cameras in pupping caves. Results from acoustic analyses of several monk seal vocalizations presented herein support the notion that remote monitoring and censusing of Mediterranean monk seals in critical cave habitats via a continuous or timed-interval audio-recording system is feasible. With recent advances in downsizing digital recording devices with large memory capacity, and long-lasting batteries (e.g., iPods, MP3 recorders, or cell phones with recording applications), it is feasible and economical to use small digital units to continuously or periodically monitor the acoustic behavior of Mediterranean monk seals in pupping caves. Such digital units, which are adapted to recording human speech, would also cover the frequency range of Mediterranean monk seal vocalizations, decrease the number of times data are retrieved and batteries replaced, and minimize monk seal disturbance.

Results of this acoustical analysis of sounds from a small number of Mediterranean monk seals indicated that further acoustic research on this species is warranted and should include airborne recordings of adult males and increased sample sizes of vocalizations from wild adult females and pups. In addition, more recordings of ill, stressed, and captive monk seals are needed. The preliminary analyses presented herein hold promise that with sufficient data acoustic features of airborne vocalizations could indicate monk seal age, sex of a pup, approximate age of a pup, and perhaps even the health of a monk seal.

A few Mediterranean monk seals have been housed at European marine parks, and Hawaiian monk seals are regularly kept at Sea Life Park in Hawaii under the Captive Care and Release Research Project by the National Marine Fisheries Service (2008). However, little acoustic research on those individuals has been conducted or is represented in the literature. The authors recommend that when these endangered monk seals are held in captivity, every effort should be made to conduct non-invasive acoustical research to better understand their vocal behavior, among other topics.

Future field research on the acoustic behavior of Mediterranean monk seals should include analysis of the vocal ontogeny of juveniles through the age of sexual maturity, the airborne vocalizations of adult males, and the underwater vocal repertoires of all age and sex classes. Additional data from rehabilitating pups will help identify more subtle variations in vocal ontogeny of this critically endangered pinniped. Lastly, the vocalizations of wild Mediterranean monk seals should be studied at the other two main population sites (i.e., Archipelago of Madeira and Cabo Blanco region), at other times of the year, outside of the breeding season, and at different locations than pup-rearing caves to provide a better understanding of the overall vocal behavior of this species.

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