

CARDIAC EXAMINATIONS OF ANESTHETIZED STELLER SEA LIONS (*EUMETOPIAS JUBATUS*), NORTHERN FUR SEALS (*CALLORHINUS URSINUS*), AND A WALRUS (*ODOBENUS ROSMARUS*)

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Abstract: Pinniped hearts have been well described via dissection, but in vivo measurements of cardiac structure, function, and electrophysiology are lacking. Electrocardiograms (ECGs) were recorded under anesthesia from eight Steller sea lions (*Eumetopias jubatus*), five northern fur seals (*Callorhinus ursinus*), and one walrus (*Odobenus rosmarus*) to investigate cardiac electrophysiology in pinnipeds. In addition, echocardiograms were performed on all eight anesthetized Steller sea lions to evaluate in vivo cardiac structure and function. Measured and calculated ECG parameters included P-wave, PQ, QRS, and QT interval durations, P-, R-, and T-wave amplitudes, P- and T-wave polarities, and the mean electrical axis (MEA). Measured and calculated echocardiographic parameters included left ventricular internal diameter, interventricular septum thickness, and left ventricular posterior wall thickness in systole and diastole (using M-mode), left atrium and aortic root dimensions (using 2D), and maximum aortic and pulmonary flow velocities (using pulsed-wave spectral Doppler). ECG measurements were similar to those reported for other pinniped species, but there was considerable variation in the MEAs of Steller sea lions and northern fur seals. Echocardiographic measurements were similar to those reported for southern sea lions (*Otaria flavescens*), including five out of eight Steller sea lions having a left atrial to aortic root ratio <1, which may indicate that they have an enlarged aortic root compared to awake terrestrial mammals. Isoflurane anesthesia likely affected some of the measurements as evidenced by the reduced fractional shortening found in Steller sea lions compared to awake terrestrial mammals. The values reported are useful reference points for assessing cardiac health in pinnipeds under human care.

INTRODUCTION

Comparative studies have found that mammalian cardiac morphology,⁴⁵ heart mass,⁴³ electrical signal conduction,^{15,32,36} and left ventricular function,^{23,24,28} are similar among many terrestrial and aquatic species of varying body size. However, pinnipeds (seals, sea lions, and walrus), which have long been suspected of having specialized cardiac adaptations to support extended breath holding during diving, have been underrepresented in these analyses. As a result, additional in vivo studies of pinniped hearts are required to evaluate cardiac health in this taxonomic family.

Despite the noted overall commonality among mammals, some structural differences have been

observed between the hearts of pinnipeds and terrestrial mammals. For example, dissections of pinniped hearts have revealed dorsoventral compression,^{34,50} a tendency towards a bifid apex,^{9,34,45,53} and enlarged ascending aortas (aortic bulbs).^{9,26,34,49} Although dissections effectively demonstrate broad anatomical differences, they cannot reveal changes in electrical or mechanical function. To address these questions, electrocardiography and echocardiography can be performed.

Cardiac examinations of pinnipeds are limited, with only 5 of 33 species being represented in the scientific literature. Electrocardiography has been performed on at least 54 awake harbor seals (*Phoca vitulina*),^{12,19} 3 awake California sea lions (*Zalophus californianus*),¹⁹ 13 awake^{19,56} and 1 anesthetized northern elephant seal (*Mirounga angustirostris*),¹⁹ 24 anesthetized southern elephant seals (*Mirounga leonina*),¹¹ and 13 anesthetized southern sea lions (*Otaria flavescens*).⁷ There are even fewer reports of echocardiographic examinations of pinnipeds, represented by only four awake southern sea lions.⁴ Although these data provide a starting point for monitoring cardiac health in pinnipeds, the dearth of pinniped species represented makes application problematic. More

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data from additional pinniped species are needed to improve our understanding of cardiac health in these species.

The objective of the current study was to examine the structure and electrical and mechanical function of pinniped hearts using electrocardiography and echocardiography. These examinations provide valuable reference points for cardiac examinations of pinnipeds cared for in zoos, aquariums, and rehabilitation facilities.

MATERIALS AND METHODS

Overview

Electrocardiography and echocardiography were used to assess cardiac structure and electrophysiology in anesthetized, female Steller sea lions (*Eumetopias jubatus*). Additional electrocardiograms (ECG) were collected from anesthetized female northern fur seals (*Callorhinus ursinus*) and an anesthetized male walrus (*Odobenus rosmarus*) to better understand pinniped cardiac electrophysiology. All research was conducted under the approval of the Animal Care Committees at the Vancouver Aquarium and the University of British Columbia (Permit A17-0092).

Animals

Eight adult female Steller sea lions (14–21 y; 166–227 kg), five adult female northern fur seals (10 y; 23.5–38.0 kg), and one juvenile male walrus (23 mo; 366 kg) were evaluated. All were maintained in human care at either the Vancouver Aquarium (Vancouver, BC, Canada) or the UBC Open Water Research Station (Port Moody, BC, Canada). Health was assessed by the Vancouver Aquarium's head veterinarian (MH) via routine physical examinations and bloodwork. Each individual was deemed clinically healthy and exhibited normal behavior at the time of the study. None of the individuals studied were pregnant.

Anesthetic protocols

All animals were induced via inhalational gas anesthesia using a face mask according to previously established protocols under the supervision of a veterinarian (MH).²¹ The Steller sea lions and northern fur seals were induced with 5% isoflurane (Fresenius Kabi Canada Ltd, Toronto, Ontario, M9W 0C8, Canada) delivered through a precision vaporizer in 100% oxygen and maintained with 1.5–2.5% isoflurane in 100% oxygen, whereas the walrus was induced with 8% sevoflurane (Sevorane, AbbVie Corporation, Stain-

Laurent, Quebec, H4S 1Z1, Canada) in 100% oxygen and maintained with 3–5% sevoflurane in 100% oxygen. Two of the Steller sea lions, F03RO and F03IZ, were administered diazepam (Apo-diazepam, Apotex Inc, Toronto, Ontario, M9L 2Z7, Canada; ~0.14 mg/kg) orally about 30 min prior to anesthesia.

All animals were out of water for the duration of the procedure. Procedures at the Vancouver Aquarium were performed indoors, whereas those at the Open Water Research Station were performed in a semienclosed shelter. No attempt was made to keep air temperature constant at either location.

Electrocardiography of pinnipeds

Six-lead ECGs were recorded using a CardioPet ECG Device (IDEXX Laboratories, Inc, Westbrook, ME 04092, USA) and CardioPet software version 6.2. All ECG recordings were made in the frontal plane with the animals in the sternal position. Clip positioning was species-dependent. In Steller sea lions and northern fur seals, clips were attached to the axillary region or caudal webbing of each fore flipper adjacent to the axilla, and either to the tibiotarsal joint or a digit of each hind flipper. To obtain a recording from the walrus, clips were attached to subcutaneous needles positioned approximately 2–3 finger widths dorsomedial to the insertion of each fore flipper, and dorsal to the walrus's tibiotarsal joint. Clips and the surrounding skin were wetted with 70% isopropyl alcohol. Two 1-m ECG recordings were collected from each individual using a paper speed of 50.0 mm/s and amplitude of 10 mm/mV.

ECG measurements were taken by a veterinary cardiologist (MM) following standard procedures. All ECG measurements were made digitally using the IDEXX VetMedStat online web application. Measurements were taken from three consecutive good-quality ECG complexes from lead II. The following ECG parameters were measured in all individuals: P-wave, PQ, QRS, and QT interval durations, P-, R-, and T-wave amplitudes, P- and T-wave polarities, and the net direction of myocardial depolarization (mean electrical axis of the QRS on the frontal plane) was calculated. The mean electrical axis (MEA) for each individual was calculated using

$$MEA = \arctan(DI_{amp}, aVF_{amp}) \times \frac{180}{\pi}$$

where DI_{amp} is the amplitude of the R-wave in lead I, and aVF_{amp} is the amplitude of the R-wave in lead aVF.³⁰ Rhythm assessment was determined

by visual inspection of the ECG. Sinus arrhythmia was defined as >10% variation in the R–R interval.¹⁷

Echocardiography of Steller sea lions

Echocardiography was performed by the veterinary cardiologist (MM) using a LOGIQ e portable ultrasound (GE Healthcare, Milwaukee, WI 53201, USA) with an ECG module and a 4C-RS Convex Array (2.0–5.5 MHz) ultrasound probe (GE Medical Systems [China] Co, Ltd, Wuxi, Jiangsu, 214028, China). The haircoat was wetted with 70% isopropyl alcohol to flatten the hair and eliminate trapped air and then ultrasound gel was applied. Measurements were made by the veterinary cardiologist (MM) on the LOGIQ e.

The Steller sea lions were positioned for echocardiography only after ECG recordings (procedure above) were completed. Two configurations were used to perform echocardiography on the Steller sea lions. In the first two cases, Steller sea lions F00BO and F97SI were imaged from above in left lateral recumbency with the probe positioned on the right side of the chest near the flipper insertion. They were then positioned in dorsal recumbency with the probe placed on the left side of the chest to record flow velocities. This positioning was determined to be suboptimal for image quality and was subsequently modified for the remainder of the echocardiograms.

Echocardiograms of F97HA, F03RO, F03AS, F03WI, F03IZ, and F00YA were performed on a purpose-made echocardiography table with a similar design to tables used for small animal echocardiography. Two large wooden platforms were constructed and arranged with a V-shaped gap between them where the chest of the Steller sea lion could be positioned. Steller sea lions were positioned in right lateral recumbency and images were obtained from below. To record flow measurements, the platforms were repositioned so that the cutout was on the opposite side—the sea lion was positioned in left lateral recumbency with its chest over the gap and images were obtained from below. Using a purpose-made table resulted in clearer images that could be reliably obtained in less time and required less pressure from the transducer on the Steller sea lions' chests.

Right parasternal long axis, right parasternal short axis, and left parasternal apical views were obtained.⁵⁴ Reported values are an average of measurements from 1–3 cardiac cycles for each individual. M-mode measurements were made following the guidelines of the American Society

of Echocardiography,⁴⁶ and included: left ventricular internal diameters, interventricular septum thickness, and left ventricular posterior wall thickness in systole and diastole. Left ventricular measurements were made using the leading edge to leading edge technique. Diastole was defined as the onset of the QRS and systole was defined as the smallest diameter of the left ventricle and coincided either with the maximum downward excursion of the septum or maximum upward excursion of the free wall. Fractional shortening, the percentage decrease of the left ventricular internal dimension (LVID) from diastole to systole, was calculated using:

$$\frac{LVIDd - LVIDs}{LVIDd} \times 100\%$$

where LVIDd is left ventricular end-diastolic diameter, and LVIDs is left ventricular end-systolic diameter.

Two-dimensional (2D) measurements included the left atrium and aortic dimensions. Measurements of the left atrium and aortic root were obtained from the right parasternal short axis view following the Hansson method.²⁰ Measurements were obtained on the first frame showing a closed aortic valve.

Pulsed-wave spectral Doppler was used to measure maximum aortic and pulmonary flow velocities. The pulmonary valve was interrogated from the right parasternal short axis view. The aortic valve was interrogated from the left parasternal apical view. To measure blood flow velocities, the pulsed-wave gate was positioned at the level of the tips of the open valve (aortic and pulmonary). The cursor could not always be positioned in perfect alignment with blood flow, but the angle was always within an estimate of 10–20°.

Other than LA: Ao, echocardiographic measurements were not standardized to aortic diameter because Steller sea lions are known to have an enlarged ascending aorta that could make the results of such calculations smaller than they actually are, rendering these types of standardizations ineffective for comparing Steller sea lions to terrestrial mammals. In addition, echocardiographic measurements were not standardized to body weight because it has not yet been demonstrated that heart structures scale with body mass in this species.

Analysis

Normality was assessed by visual inspection of histograms and Q-Q plots. Normally distributed data are reported as mean ± SD and nonnormally

Table 1. Mean \pm SD electrocardiogram parameters for eight Steller sea lions, five northern fur seals, and one walrus under anesthesia. Mean electrical axes are reported as ranges of individual averages.

	Steller sea lion ($n = 8$)	Northern fur seal ($n = 5$)	Walrus ($n = 1$)
Mass (kg)	198.8 \pm 20.1	29.4 \pm 5.0	366.5
Heart rate (bpm)	56.5 \pm 3.7	113.0 \pm 15.5	76.0
P-wave duration (ms)	136 \pm 12	74 \pm 6	146.67
P-wave amplitude (mV)	0.17 \pm 0.05	0.18 \pm 0.03	0.30
PQ interval duration (ms)	255 \pm 24	195 \pm 95	244.67
R-wave amplitude (mV)	0.41 \pm 0.27	0.73 \pm 0.28	0.43
QRS complex duration (ms)	130 \pm 9	66 \pm 11	182.33
QT interval duration (ms)	386 \pm 35	234 \pm 18	368.67
T-wave amplitude (mV)	0.14 \pm 0.23	0.16 \pm 0.20	0.39
MEA ($^{\circ}$)	-124 to 8	-79 to 80	35

distributed data are described as median \pm IQR. The heart rate range recorded for each species in this study was compared to the general mammalian model using the equation heart rate = $(241 \pm 9) \times \text{body weight}^{-0.25 \pm 0.02}$.⁵² Mean body weights for the Steller sea lions and northern fur seals were used in these calculations.

RESULTS

Steller sea lion, northern fur seal, and walrus ECG results

ECG characteristics are listed in Table 1. All species displayed P, Q, R, S, and T deflections typical of all vertebrates and had sinus rhythm. Sinus arrhythmia was detected in five out of eight Steller sea lions (F00BO, F97HA, F03IZ, F03RO, F00YA), but not in any of the northern fur seals or the walrus. MEAs of Steller sea lions ranged from an individual average of -124° to 8° (Fig. 1), whereas those of northern fur seals ranged from an individual average of -79° to 80° (Fig. 2), and the average MEA of the walrus was -35° .

Mean heart rate while anesthetized was 56.5 \pm 3.7 bpm in Steller sea lions, 113.0 \pm 15.5 bpm in northern fur seals, and 76.0 bpm in the walrus (Table 1). Based on the allometric equation relating heart rate to body weight,⁵² Steller sea lions weighing 198.8 kg are predicted to have a heart rates ranging from 55.6 to 74.0 bpm, northern fur seals weighing 29.4 kg are predicted to have heart rates ranging from 93.1 to 114.9 bpm, and a walrus weighing 366.5 kg is predicted to have a heart rate ranging from 47.1 to 64.3 bpm.

Steller sea lion P- and T-wave polarities were fairly consistent between individuals for each lead (Table S1). In each lead, five or more Steller sea lions had positive P-waves, except for in lead aVR (for which all eight had negative P-waves). Simi-

larly, in each lead, six or more Steller sea lions had positive T-waves except for in leads aVR (one out of eight positive) and aVL (two out of eight positive). P- and T-wave polarity in northern fur seals was consistent with the pattern observed for Steller sea lions with the exception of all five northern fur seals having negative P-wave polarity in lead aVL (Table S2). The P- and T-wave polarities of the walrus were almost the same as the majority of the Steller sea lions, with the exception of having a biphasic T-wave in lead I (Table S3).

QRS depolarization patterns for each lead were fairly consistent among the Steller sea lions (Fig. 1 and Table S4) but appeared to be unique for each northern fur seal with few observable trends (Table S5). No comparisons could be made for the walrus with just one set of measurements (Table S6).

Steller sea lion echocardiography results

In all eight Steller sea lions, the right parasternal long axis view (Fig. 3) and right parasternal short axis views at the level of the aortic valve (Fig. 4), mitral valve, and papillary muscles (Fig. 5) of the heart were obtained.⁵⁴ The M-mode image of the left ventricle was obtained in the parasternal short axis view at the level of the papillary muscles (Fig. 6) and allowed for measurements in all individuals. No evidence of structural cardiac disease was noted on echocardiography for any individual. The only notable observation was the presence of a false tendon in the left ventricle of one of the Steller sea lions (F00YA).

All echocardiographic parameters were successfully measured in all study animals unless specifically noted. Echocardiographic parameters for Steller sea lions under anesthesia were summarized as medians \pm IQR (Table 2). Median heart rate determined in M-mode was 50.5 \pm 7.1

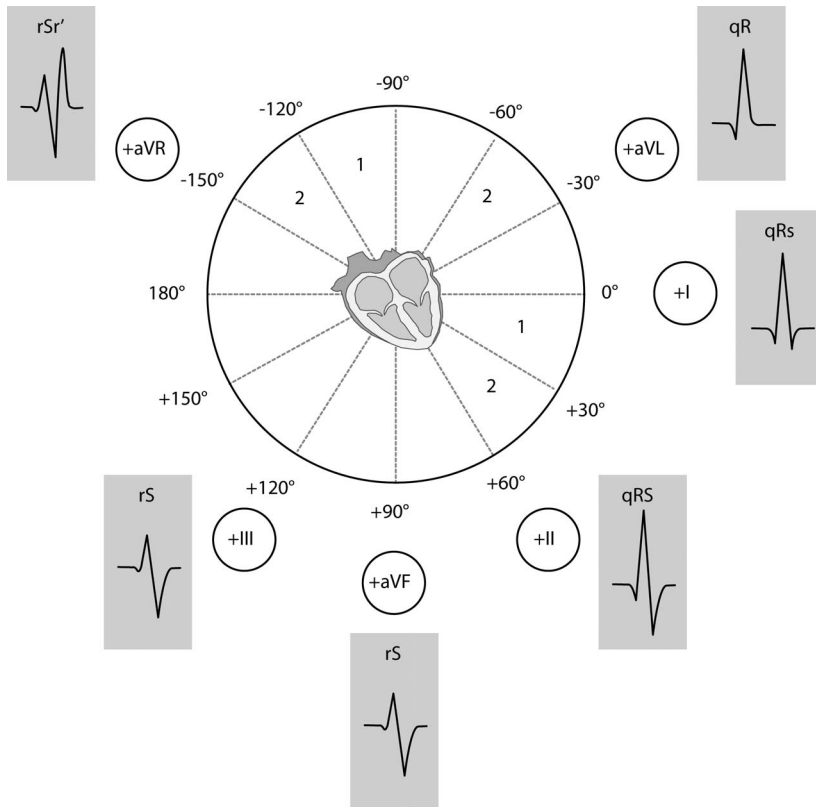


Figure 1. Mean electrical axes and typical QRS complexes of anesthetized Steller sea lions ($n=8$). The number of individuals with a mean electrical axis falling within each 30° interval is shown. Typical QRS morphology based on the shape displayed by the highest proportion of sea lions for each lead is depicted with descriptive nomenclature. Lowercase letters indicate small deflections, capital letters indicate large deflections, and apostrophes indicate a secondary wave of the given type.

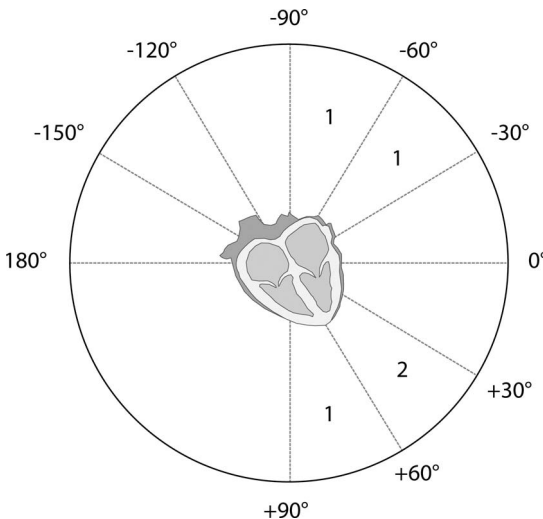


Figure 2. Mean electrical axes of anesthetized northern fur seals ($n=5$). The number of individuals with a mean electrical axis falling within each 30° interval is shown.

bpm. Left atrial to aortic root ratio (LA:Ao) was 0.92 ± 0.17 , with the ratio <1 in 5 of the 8 Steller sea lions. Fractional shortening was $31.9 \pm 11.5\%$. Peak aortic velocity (Fig. 7) measured in 7 of the 8 Steller sea lions had a median of 0.64 ± 0.26 m/s, and peak pulmonary velocity (Fig. 8) measured in 6 of 8 Steller sea lions had a median of 0.83 ± 0.21 m/s.

DISCUSSION

This study reports the first cardiac examinations for healthy anesthetized Steller sea lions, northern fur seals, and one walrus, offers a method for performing echocardiograms on large pinnipeds, and provides values for future pinniped health assessments. Cardiac data are reported for 14 pinnipeds representing 3 species, adding to existing literature to provide a total of 126 individual pinnipeds representing 8 species. All three species displayed characteristic vertebrate ECG waveforms, but Steller sea lions and northern fur seals

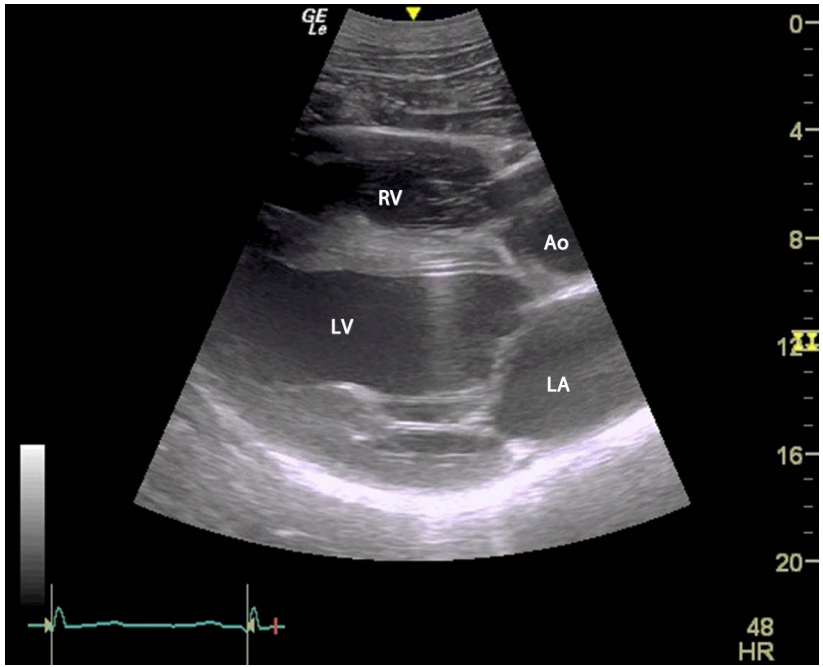


Figure 3. Right parasternal long axis view of a Steller sea lion heart showing the left ventricle (LV), right ventricle (RV), left atrium (LA), and aortic root (Ao).

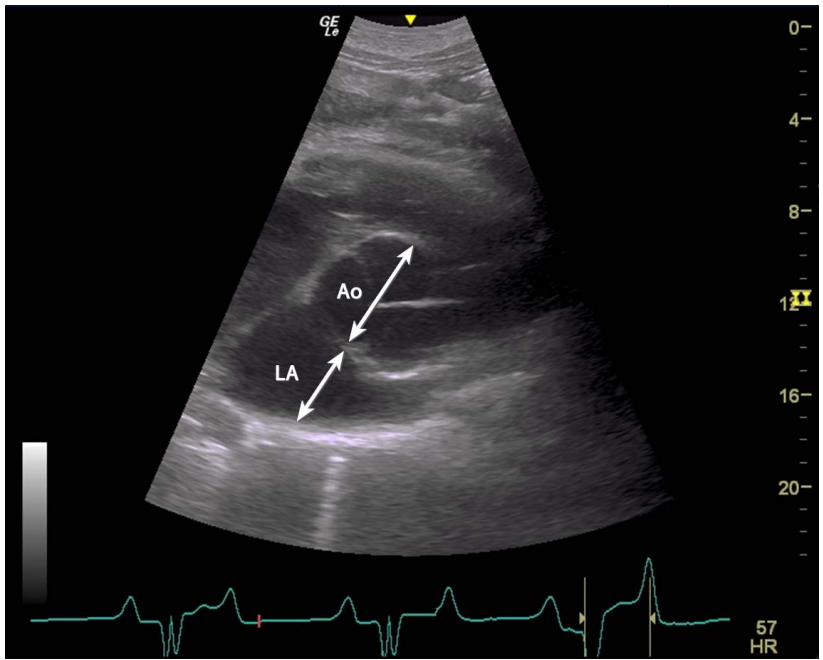


Figure 4. Right parasternal short axis view of a Steller sea lion heart at the level of the aortic valve showing the left atrium (LA) and aortic root (Ao).

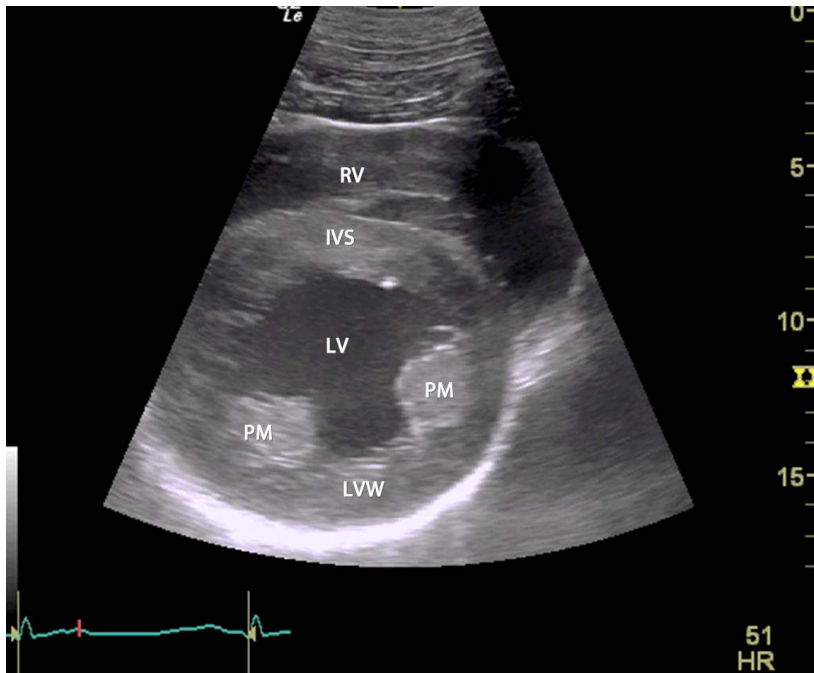


Figure 5. Right parasternal short axis view of a Steller sea lion heart at the level of the papillary muscles showing the left ventricle (LV), right ventricle (RV), interventricular septum (IVS), left ventricular free wall (LVW), and the papillary muscles (PM).

displayed considerable variation in the MEA. Steller sea lion cardiac morphology appeared normal, but there were indications that anesthesia affected fractional shortening. Interspecies and interindividual variation in ECG and echocardiographic parameters highlight the need to perform cardiac examinations on a number of pinnipeds to generate meaningful reference values.

Although the ECG and echocardiographic measurements presented in this study all come from anesthetized animals, they are valuable points of reference because animals are often anesthetized for clinical exams. Performing echocardiography on Steller sea lions required considerable transducer pressure on their chests, which may not be tolerated by individuals that are awake, even with training. Therefore, transthoracic echocardiography may be practically restricted to anesthetized Steller sea lions, even though measurements on conscious individuals (e.g., southern sea lions, a smaller species) have been successfully obtained.⁴

Steller sea lion, northern fur seal, and walrus electrocardiography

Heart rates for each species were close to the expected range for their average body mass based

on the general mammalian model.⁵² The average heart rate recorded during electrocardiography for Steller sea lions and northern fur seals fell within the predicted ranges, whereas the average heart rate for the walrus was slightly higher than predicted.

Sinus arrhythmia was detected in five of the eight Steller sea lions, but not in the walrus or northern fur seals. Sinus arrhythmia is considered a variant of normal in many species and is particularly common at lower heart rates often experienced under anesthesia. Sinus arrhythmia was more common in Steller sea lions with slightly lower heart rates (49–59 bpm compared to 60–61 bpm) except for F03AS with a heart rate of 54 bpm and normal sinus rhythm. Considering that all three species were anesthetized, it is unclear why sinus arrhythmia was only observed in the Steller sea lions, but not in the walrus or northern fur seals. Perhaps the five Steller sea lions displaying sinus arrhythmia had greater vagal tone or were under a deeper plane of anesthesia at the time of the ECG recording.

There was no overarching trend in the MEAs of Steller sea lions, northern fur seals, and the walrus. Right cranial (three Steller sea lions), left cranial (two Steller sea lions, two northern fur

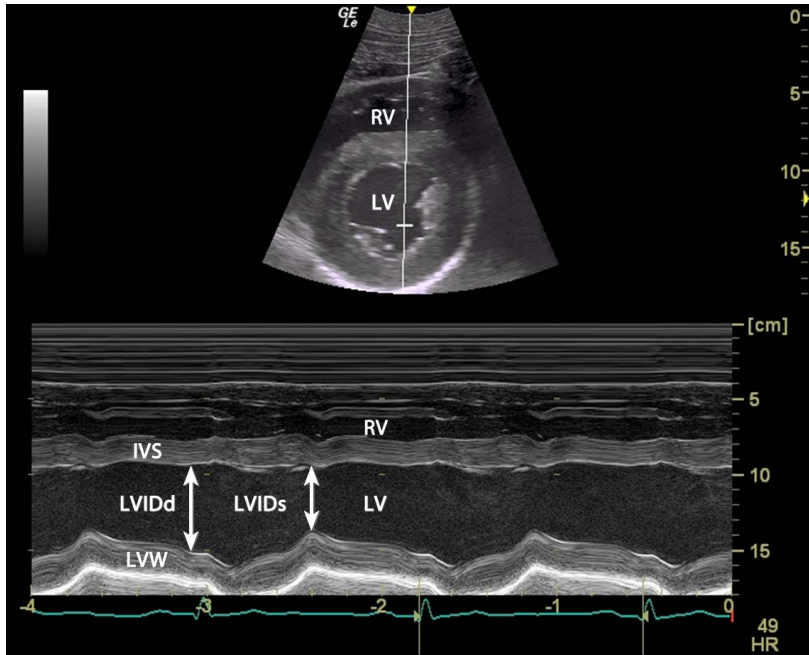


Figure 6. Right parasternal short axis view of a Steller sea lion heart at the level of the papillary muscles (2D image, top), and motion of the cardiac chambers over time (M-mode image, bottom) showing the left ventricle (LV), right ventricle (RV), interventricular septum (IVS), and posterior wall of the left ventricle (LVW). Left ventricular end-diastolic diameter (LVIDd) and left ventricular end-systolic diameter (LVIDs) were measured as shown by the white arrows using the leading edge to leading edge technique. Diastole was defined as the onset of the QRS and systole was defined as the smallest diameter and coincided either with the maximum downward excursion of the septum or maximum upward excursion of the free wall.

Table 2. Echocardiographic measurements for eight anesthetized Steller sea lions as well as a group summary (median \pm IQR). All values are M-mode measurements except for LAD, Ao, LA:Ao (2D), and AV Vmax and PV Vmax (Doppler). Abbreviations: LAD = left atrial diameter, Ao = aortic root diameter, LA:Ao = left atrial to aortic root ratio, LVIDd = left ventricular internal diameter in diastole, LVIDs = left ventricular internal diameter in systole, IVSd = interventricular septum thickness in diastole, IVSs = interventricular septum thickness in systole, LVWd = left ventricular free wall thickness in diastole, LVWs = left ventricular free wall thickness in systole, FS = fractional shortening, AV Vmax = peak systolic aortic blood velocity, PV Vmax = peak systolic pulmonary blood velocity, HR = heart rate. NA indicates measurement not available.

	Individual measurements								Median \pm IQR
	F00BO	F97HA	F03RO	F03AS	F03WI	F03IZ	F00YA	F97SI	
Age (y)	17	21	14	14	14	15	18	20	16.0 \pm 4.5
Mass (kg)	166.5	173.0	192.0	199.2	204.4	208.6	219.5	227.0	201.8 \pm 24.1
LAD (cm)	4.53	4.15	4.20	3.01	4.28	5.23	4.60	4.71	4.41 \pm 0.44
Ao (cm)	4.25	4.47	5.13	4.90	5.25	4.90	4.25	5.17	4.90 \pm 0.73
LA:Ao	1.07	0.93	0.82	0.61	0.77	1.07	1.08	0.91	0.92 \pm 0.26
LVIDd (cm)	8.26	5.57	5.23	6.45	5.93	6.25	6.00	9.16	6.13 \pm 1.06
LVIDs (cm)	5.50	3.48	3.64	5.34	4.65	4.00	4.43	5.69	4.54 \pm 1.47
IVSd (cm)	1.13	1.68	1.45	1.46	1.48	1.79	1.47	1.51	1.47 \pm 0.10
IVSs (cm)	1.70	2.15	2.10	1.97	2.20	2.85	1.70	2.28	2.12 \pm 0.32
LVWd (cm)	1.18	1.69	1.91	1.63	2.15	1.68	1.60	1.96	1.69 \pm 0.30
LVWs (cm)	1.80	2.23	2.30	1.84	2.40	2.12	2.32	2.44	2.26 \pm 0.29
FS (%)	33.4	37.5	30.5	17.3	21.2	36.0	26.2	37.9	31.9 \pm 11.5
AV Vmax (m/s)	1.06	0.49	0.58	0.64	0.94	0.58	0.75	NA	0.64 \pm 0.26
PV Vmax (m/s)	0.80	0.60	0.86	0.56	0.87	1.01	NA	NA	0.83 \pm 0.21
HR (bpm)	56.1	41.5	62.5	50.5	48.0	NA	46.8	56.2	50.5 \pm 8.7

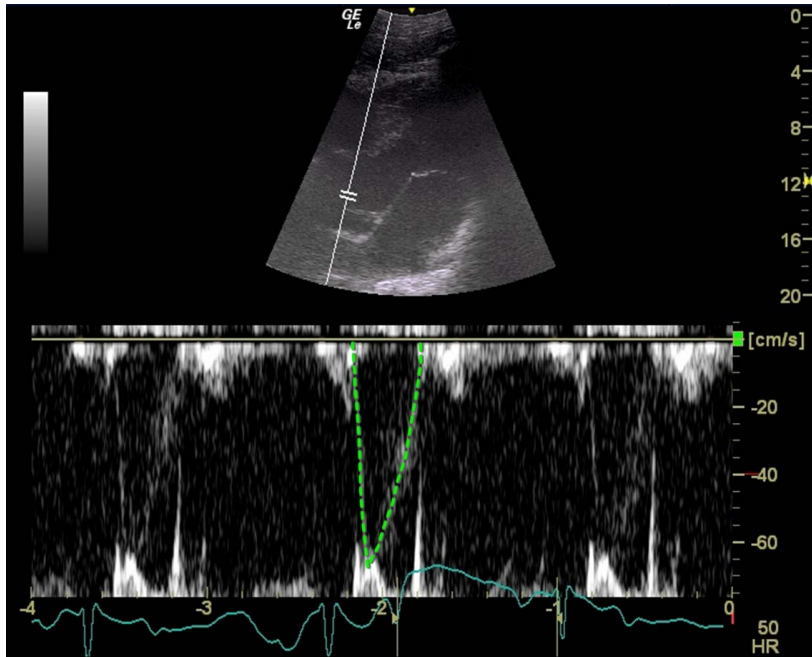


Figure 7. Pulsed-wave spectral Doppler recording of aortic blood flow in a Steller sea lion. The aortic valve was interrogated from the left parasternal apical view and the pulsed-wave gate was positioned at the level of the tips of the open valve. The cursor could not always be positioned in perfect alignment with blood flow, but the angle was always within an estimate of 10–20°. Flow profile is traced in green.

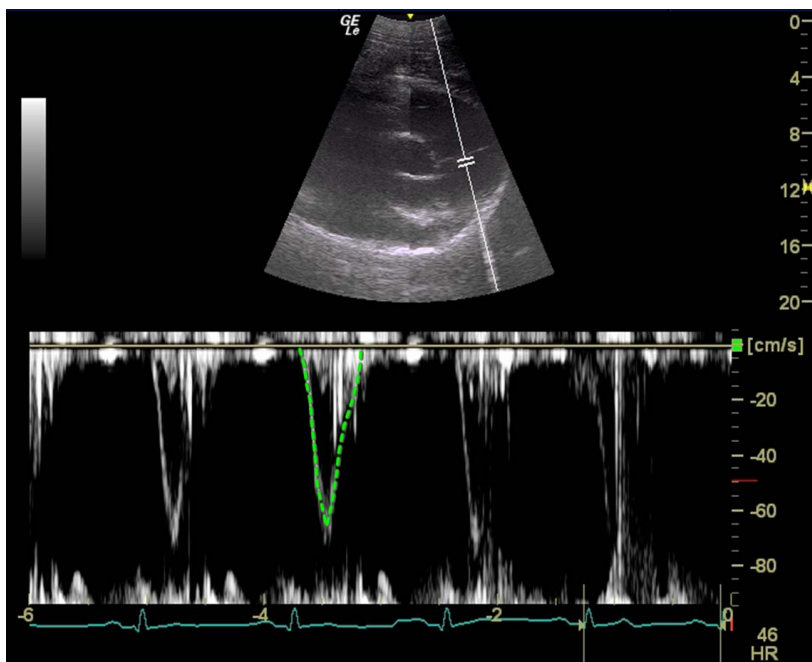


Figure 8. Doppler recording of pulmonary blood flow in a Steller sea lion. The pulmonary valve was interrogated from the right parasternal short axis view and the pulsed-wave gate was positioned at the level of the tips of the open valve. The cursor could not always be positioned in perfect alignment with blood flow, but the angle was always within an estimate of 10–20°. Flow profile is traced in green.

seals, one walrus), and left caudal orientations (three Steller sea lions, three northern fur seals) were recorded. In comparison, other species of pinnipeds (northern elephant seals, southern elephant seals, southern sea lions, California sea lions, and one harbor seal) tend to have left cranial MEA orientations.^{7,11,19} In the present study, slight differences in ECG methodology between individuals may have caused or minimally contributed to the observed MEA variability. Regardless of the possible effects of procedural differences, the variation in MEA warrants further investigation with more individuals to determine the normal range for pinnipeds or if true species-specific differences exist.

Steller sea lion cardiac structure and left ventricular function

To the authors' knowledge, this is the first reported echocardiographic study of a Steller sea lion, and only the second echocardiographic study of a pinniped to date.⁴ All hearts appeared normal; the only observation of note was the presence of a false tendon in one of the sea lions (F00YA). False tendons are bands of fibrous tissue that stretch across the left ventricle, and can be incidental in healthy mammals, especially cats.²⁵

Steller sea lions were found to have a larger aortic root diameter than left atrial diameter, similar to findings reported from an echocardiographic study of southern sea lions.⁴ A ratio of LA:Ao < 1 is commonly reported to be within the normal range of many species of terrestrial mammals.² However, these findings could be consistent with the enlarged ascending aorta reported in necropsy studies of more than 14 species of pinnipeds to date,^{9,10,26,44,50,53} including Steller sea lions.³⁴ It is unknown if the range of aortic root diameters (4.25–5.23 cm) reported for Steller sea lions in the current study represents an enlargement compared to size-matched terrestrial mammals because there is currently no standard scaling relationship to compare to. More echocardiographic assessments of large free-ranging species are needed to make this determination.

Average fractional shortening in Steller sea lions in this study (32%) was less than the median value calculated from 14 species of terrestrial mammals imaged awake (40%).^{1,8,13,14,16,18,29,33,37,40,47,55} This mildly reduced fractional shortening likely resulted from the negative inotropic effects of isoflurane anesthesia, which commonly reduces fractional shortening in healthy mammals.^{39,48,51} Indeed, when considering only studies of anesthetized terrestrial

mammals, median fractional shortening (32%) was identical to values from the present study. Considerable variability has been reported in the fractional shortening of anesthetized terrestrial mammals (11–47%) which may be due to species-specific differences or the anesthetic protocols used. Further support that anesthesia had an effect on fractional shortening in Steller sea lions comes from the finding that reduced fractional shortening was not observed in bottlenose dolphins and southern sea lions that were examined without anesthesia.^{4,5} Measurements of fractional shortening in conscious Steller sea lions are needed for confirmation.

Limitations

One limitation of this study was the use of inhalational anesthesia because of its known effects on cardiac characteristics.^{31,35,41} Despite this limitation, the reported values are still relevant for clinical purposes because examinations in these species are usually made under anesthesia.

During the procedures, animals were maintained on the lightest plane of anesthesia possible to accomplish the procedures safely, and monitored for essential physiologic parameters, such as heart rate and respiration rate, that would indicate a need for intervention. Variation in physiologic parameters that were not monitored during data collection may have affected the results of this study. Body temperature, blood gas parameters, and anesthetic depth have known effects on the cardiovascular system and potentially varied among individuals at the time that measurements were recorded. For example, pinnipeds may breath hold during anesthesia, which can result in hypercapnia and associated arrhythmias.^{3,22} Without measurements of end-tidal CO₂ or the partial pressure of CO₂ in the blood, it is unknown if hypercapnia contributed to the reported variation in cardiac rhythm between individuals and species. Variation in cardiac rhythm could also be explained by variations in anesthetic depth and body temperature as arrhythmias have been associated with deeper planes of anesthesia,⁴² and hypothermia.⁶ This could be confirmed in future studies by monitoring the expired fraction of inhalational anesthetic and body temperature. Bradycardia is also influenced by physiological status and can result from excessive anesthetic depth, hypothermia, and hypoxemia.^{27,38} These effects have been reported in anesthetized otariids,²¹ and walruses,³ and may have resulted in lower heart rate measurements than expected in some individuals in

this study. In future studies, uncertainty surrounding the effects of physiological status on cardiovascular measurements can be reduced with greater physiologic monitoring during anesthesia.

Another limitation of this study was the use of a convex array probe, which is not the preferred probe type for echocardiography. This probe was selected because low-frequency probes are needed to perform echocardiography on animals this large, and it was the lowest frequency probe available. This limitation could be overcome in future studies by using a phased-array probe with a low frequency.

Transthoracic echocardiography was challenging to accomplish in Steller sea lions primarily due to their large size and limited acoustic windows. As a result, ideal alignment was not always possible. Future transthoracic studies of Steller sea lions could use Anatomical M-mode, a software application that allows the angle of the M-mode cursor to be adjusted, to compensate for suboptimal alignment. Aortic and pulmonary flow velocities were also likely affected by a suboptimal angle of insonation, as shown by the aortic flow velocity being lower than the pulmonary flow velocity. Transesophageal echocardiography may prove to be the better alternative for evaluating blood flow velocity in Steller sea lions.

CONCLUSION

The cardiac measurements obtained for anesthetized Steller sea lions, northern fur seals, and a walrus demonstrate the feasibility of recording ECGs and echocardiograms from these species. Using a purpose-made echocardiography table with the Steller sea lions positioned in right lateral recumbency rendered better echocardiographic images. ECG waveforms and echocardiographic findings were similar to results in terrestrial mammals.

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