The Content of Reindeer Male Vocalisations: Acoustic Cues to Age and Weight

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A Thesis

In the Department of

Biology

Presented in Partial Fulfillment of the Requirements

For the Degree of

Master of Science (Biology)

at Concordia University

Montreal, Quebec, Canada

March 2022

CONCORDIA UNIVERSITY

SCHOOL OF GRADUATE STUDIES

This is to certify that the thesis prepared

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Entitled: The Content of Reindeer Male Vocalisations: Acoustic Cues to Age and Weight

and submitted in partial fulfillment of the requirements for the degree of

Master of Science (Biology)

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Abstract

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Laura Puch

Acoustic signals are widely used by animals for communication and the regulation of interactions. Moreover, some acoustic parameters of animal vocalisations have been shown to reliably indicate male quality and play a role in mate and rival assessment. For example, in some deer species, formant frequencies act as cues to body size, and lower formant values and spacing are selected by females in a mate choice context. Reindeer possess a peculiar vocal tract anatomy involving a laryngeal air sac which probably acts as an additional filter, making it a candidate species for novel investigations in the field of bioacoustics. We investigated whether some acoustic parameters were good indicators of age and body weight (used as an index for body size). We did this by performing acoustic analyses using recordings collected from a semi-domesticated reindeer population in northern Finland. This study first highlights the complexity of acoustic parameter extraction from reindeer male vocalisations, likely due to the presence of the laryngeal air sac. We found age of subadult males (aged 2.5-4.5 years) to be negatively correlated with formant F3 and formant spacing, suggesting that their vocalisations convey information on the caller's age. Formant frequencies were not affected by male body weight, suggesting they may not provide information on male size in our population. In contrast, we found that heavier males had a lower formant spacing. It is thus probably a key acoustic parameter in mate and rival assessment as it gives an honest indication on male body weight. We discuss the importance of reliable acoustic cues to size and quality indices in sexual selection contexts. However, it is highly likely that reindeer males and females rely on other signals as well, such as visual and olfactory cues, to gain information on male attributes. We suggest that further investigation is needed to better understand how the laryngeal air sac affects vocal production.

Acknowledgements

I would like to thank my supervisor, Dr. Robert Weladji, for his support throughout this project, which has not been a long quiet river. I would also like to thank Jukka Siitari at the Natural Resources Institute of Finland (Luke) for the management of GPS collars data and for help in handling the data, and Mika Tervonen at the Reindeer Herder's Association in Finland for the management of reindeer in Kutuharju. I also thank Keenin Coombs who helped with data collection in the field and Lucie Barluet de Beauchesne at the ENES lab (Equipe de Neuro-Ethologie Sensorielle) who helped with acoustic analyses.

A big thank you to my lab mates, Alex and Shao in particular, who made me forget how lonely I felt during the first year of the Covid-19 pandemic. I am infinitely grateful for the support of my family and boyfriend. Without Raphael, who knows if this thesis would have seen the light of day?

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I would like to acknowledge here the contribution of the following collaborators on the project:

- Conception and design of the study: L, R, O
- Data collection: L, R, O
- Analysis and interpretation of data: L, R
- Drafting of manuscript: L
- Critical revision: L, R, O, and J

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Introduction

Acoustic signals are widely used by animals for communication and interaction regulation (Bradbury & Vehrencamp, 1998). The emitter can convey diverse information to the receiver(s) for group cohesion, danger avoidance, peer recognition, etc. (Bradbury & Vehrencamp, 1998). Identifying the type of information conveyed by animal acoustic signals is a central research focus in the field of bioacoustics (Bradbury & Vehrencamp, 1998). To this end, researchers show great interest for acoustic signals involved in mating.

In the context of reproduction, it has been shown that acoustic communication play an important role in sexual selection. The theory of sexual selection, introduced by Darwin (1871), is one of the main forces driving species evolution. He suggested that some traits might evolve if they are sexually selected, that is if they increase the individual's reproductive success, even at the expense of their survival. Acoustic signals are the traits most often shown to be sexually selected, as shown in insects, frogs, and birds (Andersson, 1994). Males can use acoustic signals for territory defense and in contests with other males to monopolise access to mates (Arak, 1983; Boake & Capranica, 1982; Krebs et al., 1978). Males can also use acoustic signals for attracting females (Gottlander, 1987; Hedrick, 1986; Robertson, 1986).

More recently, the study of mammal vocalisations has received growing attention (Taylor & Reby, 2010). Originally developed to study human speech, the source-filter theory (Fant, 1970) was generalised to all mammals given that the vocal production apparatus is fundamentally similar across mammalian species (Titze, 1994). Based on the mode of production of vocalisations, the source-filter framework has revealed how anatomical constraints of the mammalian vocal tract led to vocalisations conveying honest information on physical attributes of the caller, such as body size

(Taylor & Reby, 2010) (**Figure 1**). As this can have implications for the mating process, many studies have investigated the potential relationships between acoustic parameters of male mammal vocalisations and their attributes (see review by Taylor & Reby, 2010).

Following the source-filter framework, the role of two components in particular has been investigated, the fundamental frequency and formant frequencies (Taylor & Reby, 2010) (**Figure 1**). The fundamental frequency is determined by the source signal, generated by vibration of the vocal folds in the larynx. Conversely, formant frequencies result from the filtering of the signal by the vocal tract. Studies have investigated these acoustic features as indicators of age and body size (Charlton et al., 2009; Fitch, 1997; Reby & McComb, 2003b; Riede & Fitch, 1999; Sanvito et al., 2007; Vannoni & McElligott, 2008; Wyman et al., 2012). The fundamental frequency appears to be a poor predictor of body size (Charlton et al., 2011; Reby & McComb, 2003b; Taylor & Reby, 2010). This could be explained by the fact that the organs involved in the production of the source signal are soft and unconstrained by skeletal structures (Taylor & Reby, 2010). In contrast, the vocal tract is not as flexible, and thus the formant frequencies seem to be static cues to body size, directly reflecting the length of the vocal tract (Charlton et al., 2011; Reby & McComb, 2003b; Wyman et al., 2012).

Some deer are group-living animals, characterized by a social structure in which communication is crucial to regulate interactions (Vannoni et al., 2005). They are highly vocal animals and deer vocalizations are also notable for their diversity. In gregarious species, like red deer (*Cervus elaphus*) or reindeer (*Rangifer tarandus*), males compete intensely for females. They evolved armaments and sexual displays, such as vocal displays which are highly developed (Reby & McComb, 2003a). In the context of reproduction, calls are used for territorial defense, mate attraction and male-male competition (Reby & McComb, 2003a). These sexual calls have been widely studied in red deer but also in fallow deer (*Dama dama*), Père David's deer (*Elaphurus davidianus*), and sika deer (*Cervus nippon*) (Charlton & Reby, 2011; Feighny et al., 2006; Liu et al., 2016; Reby & McComb, 2003b; Volodin et al., 2015; Yen et al., 2013). Reindeer vocalisations have been poorly studied. Compared to other species, the acoustic repertoire of reindeer is quite small (Lent, 1975). Females and calves use vocalisations for mother-young communication and recognition (Espmark, 1971). Males are restricted to the rutting period, they use a characteristic vocalisation while establishing harems, herding females and chasing rivals (Espmark, 1964). This vocalisation, called 'grunt', consists of a series of short calls which sound hoarse, and are produced by a series of exhalations in rapid succession (Espmark, 1964). Grunts can be directed towards females in courtship contexts or towards males during agonistic interactions.

Unlike other cervids, reindeer have evolved an inflatable laryngeal air sac; and sexual selection seems to have favoured a considerable size of the air sac in males (Frey et al., 2007). Its acoustic function has yet to be clarified. Some other mammal species possess an air sac, but its role is also not yet well understood. The air sac seems to affect formant position and call loudness (Riede et al., 2008), or to allow re-breathing the air to avoid hyperventilation (Hewitt et al., 2002) in some primates. In the muskox (*Ovibos moschatus*), the air sac may increase the amplitude of roaring or darken the timbre of the roars (Frey et al., 2006). A recent study comparing three deer species showed that inter-specific acoustic variability is related to differences in vocal anatomy (Frey et al., 2021). Therefore, we hypothesize that the presence of this air sac in reindeer may affect the acoustic structure of their vocalisations by acting as an additional acoustic filter. This study thus aimed at investigating whether some acoustic parameters of reindeer male rutting vocalisations encode information on physical attributes of the sender. We first performed acoustic analyses using recordings collected from a semi-domesticated reindeer population in northern Finland. We further examined the relationships between acoustic parameters and both male age and body weight (used as an index for body size). We hypothesized that both physical attributes would predict formant frequencies and the related acoustic features. Then, we discuss the potential influence of the laryngeal air sac on these relationships and whether vocalisations have the potential to convey information on male quality that could be further used for mate and rival assessment.

Materials & Methods

i. *Study site and animals*

The data was collected at the Kutuharju field reindeer research station in Kaamanen, northern Finland (69°N, 27°E). The studied herd consists of about 100 animals every year (including males, females, and calves) from a semi-domesticated reindeer population monitored since 1969.

During the rut (mid-September – late October), animals are free-ranging in a large, fenced enclosure (Lauluvaara ~ 13.8 km^2). Before the mating season, males are weighed and fitted with VHF radio collars allowing telemetry tracking to find them in the enclosure. Females are fitted with coloured collars. All animals are of known age and individually recognisable thanks to the long-term book-keeping of the herd demography.

ii. *Acoustic recordings*

Male grunts were recorded during the rut 2019. The population included 15 reproductive males. Recordings were made with the Zoom SGH-6 directional microphone (Zoom Corporation, Tokyo, Japan), equipped with a windscreen, and connected to the Zoom H6 Handy Recorder (Zoom Corporation, Tokyo, Japan) (amplitude resolution: 24 bits, sampling rate: 96 kHz). The focal sampling method was used. We recorded vocalisations from 10 different males between September 30 and October 9.

iii. *Acoustic analyses*

We discarded vocalisations of poor quality (e.g., with excessive background noise) from the analyses. We selected and extracted 75 high-quality grunts from 9 different reproductive males (aged 2.5-5.5 years) based on both listening and visual inspection of spectrograms (**Figure 2**). We then conducted acoustic analyses with Praat software v. 6.1.38 (Boersma & Weenink, 2021).

Following the source-filter framework, we proceeded to extract the fundamental frequency and the first formant frequencies according to their potential to predict body size. By visualising the spectrogram and using the functions “Show pitch” and “Show formants” from Praat software, we noticed that the software was not able to extract the fundamental frequency at all and was not able to correctly extract the formants. That was expected as reindeer vocalisations tend to be noisy and unstructured. We thus applied “manual” methods to extract the acoustic parameters rather than using the automatic functions of the software.

To extract the fundamental frequency of a call, we focused on a stationary part of 0.05 s from the middle of the call in order to calculate it by the reciprocal of the period (i.e. mean distance between signal maxima in the time domain) as it was done by Frey et al. (2007). However, we could not distinguish the harmonic structures of calls (**Figure 2**), hence, we were unable to determine the fundamental frequency.

For formants frequencies, selected grunts were concatenated by selecting and extracting only the exhalation parts of grunts (**Figure 2**). For each vocalisation, we generated the spectrum using the ‘To Spectrum’ function from Praat software and applied the ‘Cepstral smoothing’ function with a 200-Hz bandwidth (Charlton et al., 2009, 2011). Based on both visual inspection of the spectrum and the list generated by the “To formant (peaks)” function, we extracted the seven first formants (F1, F2, F3, F4, F5, F6, and F7) (**Figure 3a**). If some of them could not be identified, we applied the ‘Cepstral smoothing’ function with a 50-Hz bandwidth.

To estimate the vocal tract length (VTL), we first calculated the formant spacing (ΔF) (spacing between any two consecutive formants) based on the linear regression line fitting method (when at least 4 formants were identified, N = 73 grunts) (Reby & McComb, 2003b) (**Figure 3b**). We then used the following equation: $VTL = \frac{c}{2\Delta F}$ where c is the speed of sound in air in a mammal's vocal tract (approximated as 350 m/s) (Reby & McComb, 2003b).

iv. Statistical analyses

We conducted statistical analyses in R v. 3.5.1 (R Core Team, 2018). To determine whether vocalisations have the potential to convey information on male quality, we tested whether extracted acoustic parameters (F1-F7, ΔF, VTL) were good predictors of male characteristics (age and body weight). Using the lme4 package (Bates et al., 2015), simple linear mixed-effects models were used to test the effect of age and body weight, respectively, on acoustic parameters. Each of the seven formant frequencies, formant spacing, and vocal tract length were successively used as response variables. Age, then body weight, was used as explanatory variable. Male identity was included as a random factor in all models to account for the repeated measurements on the same individuals, thereby controlling for pseudoreplication. We examined the normality of model residuals and then tested the significance of each model. We also used the MuMIn package (Barton, 2019) to calculate the conditional coefficient of determination (R^2) for each model, which represents the variance explained by the entire model, including both fixed and random effects.

We used a second method to investigate the effect of age on acoustic parameters as it has been shown that the pattern of body weight growth for reindeer males aged 0–5 years appears to plateau after reaching 4 years old (Melnycky et al., 2013). Accordingly, we adjusted our model assuming that the growth of the vocal tract might be affected in the same way. Indeed, as we hypothesised that acoustic parameters depend on vocal tract length, it is possible that age stops influencing acoustic parameters after 4 years old. To investigate this, we performed simple linear mixed-effects models to test the effect of age on acoustic parameters for subadult males (aged 2.5-4.5 years) only, removing adult males (aged ≥ 5 years). We also used t-tests to compare 4.5-year-old males (N = 1) and 5.5-year-old males (N = 3).

Results

i. *Descriptive statistics of acoustic parameters*

Out of the 75 analysed vocalisations, all seven formant frequencies were identified in 42 vocalisations. One formant could not be detected in 27 vocalisations, 2 formants could not be detected in 4 vocalisations, 3 in 1 vocalisation, and 4 in 1 vocalisation also. We discarded the latter two for further analyses; only vocalisations for which at least 5 formants were identified were kept (N = 73). F3 was the formant detected more often (97.26%) while F1 was the one less often (87.67%) detected (**Table 1**). Moreover, for F2 and F3, and F6 and F7, we observed an overlap in the frequency ranges (**Table 1**). The mean (± SD) calculated ΔF was 389.6 ± 9.4 Hz and the mean estimated VTL was 44.9 ± 1.1 cm (**Table 1**).

ii. *Formant frequencies and age*

The simple linear mixed-effects models on all males did not show any effect of age on acoustic parameters (all $P > 0.05$). When only subadult males were considered, age still did not predict F1, F2, F4, F5, F6, and F7. Conversely, we found a significant negative effect of age on F3 (estimate ± SE = -46.64 ± 19.45, $P = 0.016$, $R^2 = 0.440$; **Figure 4a**) and on ΔF (estimate ± SE = -6.683 ± 2.968, $P = 0.019$, $R^2 = 0.313$; **Figure 4b**), as well as a significant positive effect on VTL (estimate ± SE = 0.811 ± 0.342, $P = 0.018$, $R^2 = 0.323$; **Figure 4c**). For these variables, the t-tests then confirmed that there was no significant difference between males aged 4.5 years and males aged 5.5 years (all $P > 0.05$).

iii. Formant frequencies and body weight

We found no effect of body weight on the seven first formant frequencies F1 to F7 (all $P > 0.05$). On the other hand, we found that body weight significantly predicted ΔF and the resulting estimated VTL. Increasing body weight was associated with a decrease in ΔF (estimate ± SE = -0.233 ± 0.082, $P < 0.01$, $R^2 = 0.248$; **Figure 5a**) while heavier males had a higher VTL (estimate ± SE = 0.027 ± 0.0095, $P < 0.01$, $R^2 = 0.255$; **Figure 5b**).

Discussion

We extracted acoustic parameters from vocalisations of rutting male reindeer. This allowed us to estimate the vocal tract length of the animals and to investigate if reliable information on physical attributes of males is encoded in grunts. The aim was to test whether age and/or body weight predicted formant frequencies, formant spacing, and the estimated vocal tract length. We found formant F3 and formant spacing to be good indicators of male age in subadults, and formant spacing to be a good indicator of male body size.

i. Acoustic parameters extraction

This study showed that acoustic parameters extraction from reindeer vocalisations is challenging. Similarly to male North American bison bellows (Wyman et al., 2012), we were unable to detect and extract the fundamental frequency from the recorded vocalisations. Unlike bison bellows and red deer roars where formant frequencies are well defined (Reby & McComb, 2003a; Wyman et al., 2012), the extraction of formant frequencies is not straightforward from reindeer grunts. Indeed, some formants could not be detected. For instance, F1 had to be distinguished from the microphone noise using the 50-Hz bandwidth most of the time. Both bad definition of formant frequencies and inconsistencies between individuals resulted in range overlaps. To our knowledge, Frey et al. (2007) is the only other study investigating male reindeer vocalisation acoustic structure. They measured an average fundamental frequency of 55 Hz but did not report any measurements of formant frequencies as “only very few and ill-defined formant frequencies were detectable”. They also mentioned unpublished recordings by other researchers (referring to a personal communication by D. Reby in 2006) which revealed clearer vocal tract resonances and conclude that further studies are required to solve this inconsistency, which we agree with.

We estimated the vocal tract length from the formant spacing. The mean VTL estimated here was 44.94 ± 1.08 cm. Frey et al. (2007) measured the VTL with CT images and direct measurements, obtaining on average 37 cm for an adult male, slightly lower than our measure. By contrast, in both red deer and koala (*Phascolarctos cinereus*), the VTL estimated from acoustic measurements was two to three times as long as expected from their anatomy (Charlton et al., 2011; Tecumseh Fitch & Reby, 2001). This can be explained by the fact that these species possess a descended larynx which they extend when vocalising. This allows males to lower the formant frequencies of their vocalisations, hence, exaggerating their perceived body size. The small difference between our estimation and the measured vocal tract length thus suggests that reindeer may not be able to retract their larynx as far, corroborating the conclusion previously made by Frey et al. (2007) based on an in-depth study of the vocal tract anatomy of reindeer.

ii. Formant frequencies and physical attributes

We found that age predicted some acoustic parameters only for subadult males. The formant frequency F3 appears to provide a clear indication of male age, as well as formant spacing ΔF and VTL. We argue that this is probably due to the lengthening of the neck and vocal tract that occurs with body growth. Previous research suggested that maximum body weight of reindeer males is reached at 4 years old (Melnycky et al., 2013). It is likely that their vocal tract does not grow significantly after that age and, thus, formant frequencies within adults would not be expected to vary significantly with age.

Our results indicate that formant frequencies do not provide reliable information on male body weight in reindeer. This could be partly explained by the fact that formants in the lower half of the frequency spectrum (F1, F2) are more subject to variation in vocal tract shape (Fant, 1970).

This means that they cannot be good indicators of vocal tract length, hence body size (Reby & McComb, 2003b; Sanvito et al., 2007). Conversely, it is surprising that the upper formants (F3 to F7) did not reflect body weight even though they are known to be better indicators of body size (Reby & McComb, 2003b; Sanvito et al., 2007).

Formant spacing provides honest information on body weight in reindeer as we showed that heavier males had a lower formant spacing. As expected, a positive relationship was found between body weight and the estimated vocal tract length. These results are consistent with previous studies on other mammalian species (giant panda: Charlton et al., 2009; red deer: Reby & McComb, 2003b; elephant seal: Sanvito et al., 2007; bison: Wyman et al., 2012) and with our predictions, based on the assumptions that anatomical constraints influence the acoustic structure of animal vocalisations allowing some acoustic parameters to be reliable cues to the caller's body size. However, the variance in formant spacing explained by body weight in reindeer seems to be lower than for other mammals. The r-squared obtained in our study was 0.248, whereas it was 0.329 in bison (Wyman et al., 2012), 0.35 in elephant seal (body length used instead of body weight) (Sanvito et al., 2007), and 0.88 in giant panda (Charlton et al., 2009). This reduced capability of formant spacing to convey information about the caller's body size may be due to the complex vocal system of reindeer which includes a laryngeal air sac, potentially acting as an additional filter. A similar hypothesis was suggested for the study on elephant seals (Sanvito et al., 2007).

iii. Role of the laryngeal air sac

The role of air sacs present in some mammal species is not yet well understood. Just like for reindeer (Frey et al., 2007), several studies have observed a pronounced sexual dimorphism regarding the size of air sac. For instance, the occurrence of a paired lateral laryngeal ventricle is specific to males in the Mongolian gazelle (*Procapra gutturosa*) (Frey & Riede, 2003). A larger air sac in males has also been observed in some primate species (Hewitt et al., 2002). Sexual dimorphism is prevalent in polygynous species and often results from sexual selection pressures (Andersson, 1994). We can thus argue that the large laryngeal air sac of male reindeer may be conserved by sexual selection. To go further, we suggest investigating how the air sac influences the acoustic structure of vocalisations. In reindeer, Frey et al. (2007) observed that the increasing size of the air sac from juveniles to subadults and to adults correlates with the decrease of harmonic structures in the spectrograms of their respective calls. Here, we showed the complexity of acoustic analyses for this species. This is in agreement with Frey et al. (2007)'s observation, suggesting that the presence of the laryngeal air sac in reindeer is probably responsible of the difficulties we encountered when extracting acoustic parameters from vocalisations. Moreover, a possible effect of the air sac in adult reindeer suggested by Frey et al. (2007) is to mask formants of the vocal tract. This could explain the lack of relationship we reported between the upper formants and body weight. In the Mongolian gazelle, it is suggested that air sacs might function as resonators and increase amplitude during vocalisation, as in some primate species (Frey & Riede, 2003). Conversely, the expected relationship between body weight and formant spacing did not seem to be altered by the presence of the laryngeal air sac in reindeer. Its acoustic function and role thus need further investigation.

iv. Conclusions

Our results suggest that males may convey reliable information on physical attributes to receivers through vocalisations. Therefore, male vocalisations could be subject to sexual selection by playing a role in mate and rival assessment. Females could use formant frequencies as an indicator of male quality in the context of reproduction, just like it has been demonstrated in red deer (Charlton et al., 2007) and koala (Charlton et al., 2012). By choosing their mates based on quality criteria, females might gain indirect genetic benefits of more competitive offspring. On the other hand, males could use formant frequencies as an indicator of their rival's fighting ability to avoid fights with bigger males during the rut. For example, koala and red deer males are attentive to calls from other males and adjust their vocal responses accordingly, vocalising more when hearing calls from larger and potentially more dangerous males (Charlton et al., 2013; Reby et al., 2005).

However, the reduced capability of formants to convey information about the caller's body size in reindeer suggests that vocalisations are probably not a major determinant in mate and rival assessment in reindeer. Males and females may also rely on visual and/or olfactory cues for example (Andersson, 1994). For instance, an experiment showed that human males were able to accurately estimate the physical strength of other males from photos of their bodies and face (Sell et al., 2009). It has also been demonstrated that the agile antechinus (*Antechinus agilis*) females rely on olfactory cues to choose their mates as it provides them with information on genetic relatedness (Parrott et al., 2007). Moreover, we can hypothesize that the significant but low variance in formant spacing explained by male body weight does not result from direct sexual selection pressures. The latter probably affect mainly body size, which in turn shapes vocalisations. The primary role of grunts during the rut could simply be to get attention from females (Lent, 1975) and warn/threaten other males in forested areas when visual cues are not available.

Further investigation is needed to identify the potential different roles of male vocalisations in intersexual and intrasexual selection. Indeed, the social context of grunts should be considered to investigate whether there are differences in calls directed towards females and towards males. If so, we could assume the conveyed information is not the same. This has for example been reported for fallow deer (Charlton & Reby, 2011). More importantly, we need to investigate whether females and rival males perceive the variation of formant frequencies with the emitter's attributes and use it for mate and rival assessment. Playback experiments could be used to address that (Charlton et al., 2007, 2012; Jiang et al., 2022; Reby et al., 2005, 2010). We would expect females to show preference for grunts having formants with lower spacing, as the caller is likely to be larger and have higher fitness.

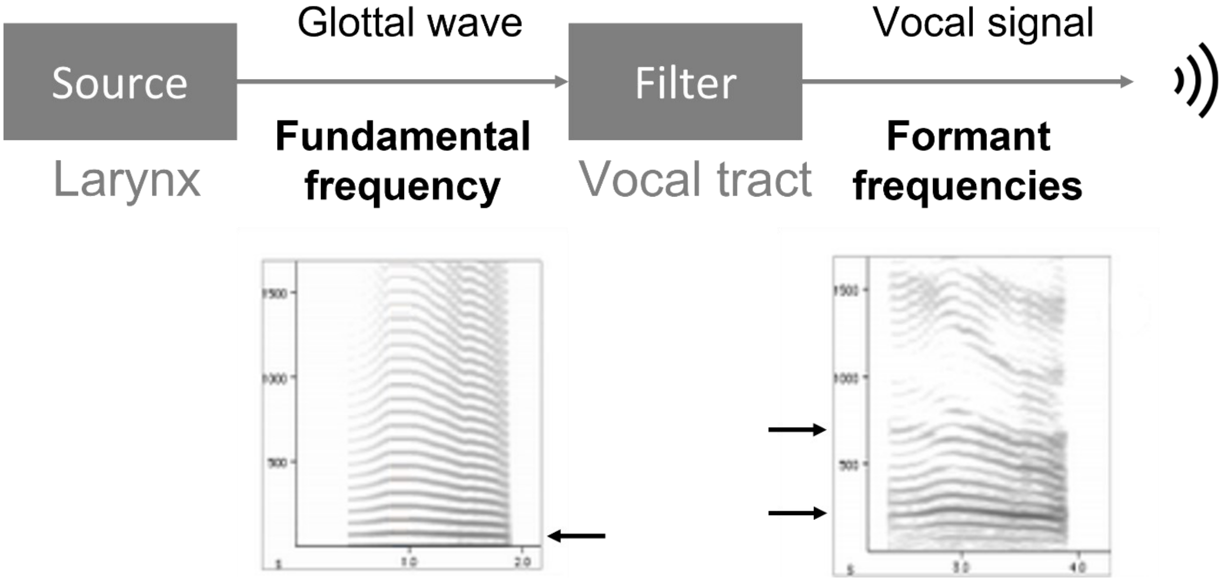

Figure 1. Illustration of the decomposition of mammal vocal signals according to the source-filter theory and identification of resulting characteristic acoustic features on spectrograms of a red deer roar (Reby & McComb, 2003a).

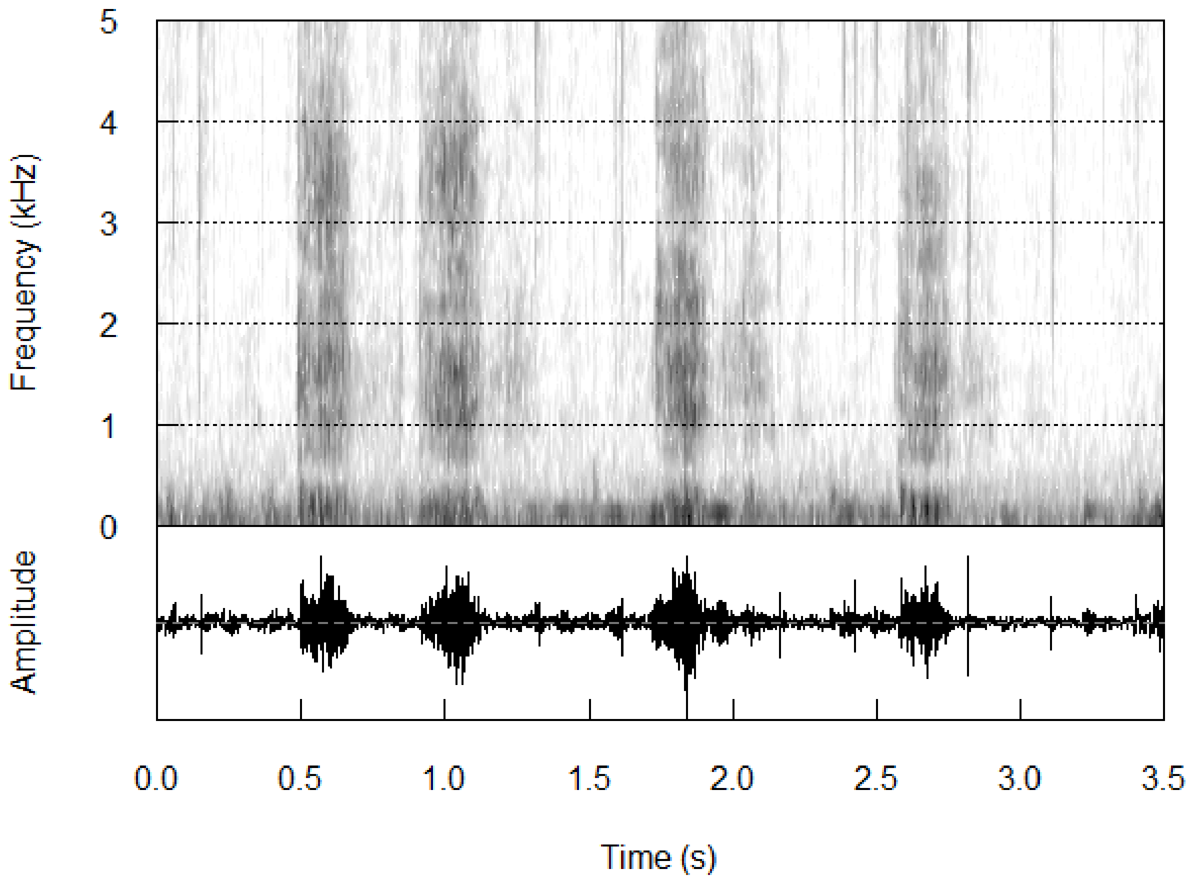

Figure 2. Waveform and spectrogram of a grunt from a reindeer male (aged 3.5 years). Darker vertical bands on the spectrogram are the exhalation parts of the grunt. Fundamental frequency, harmonics, and formant frequencies are poorly defined.

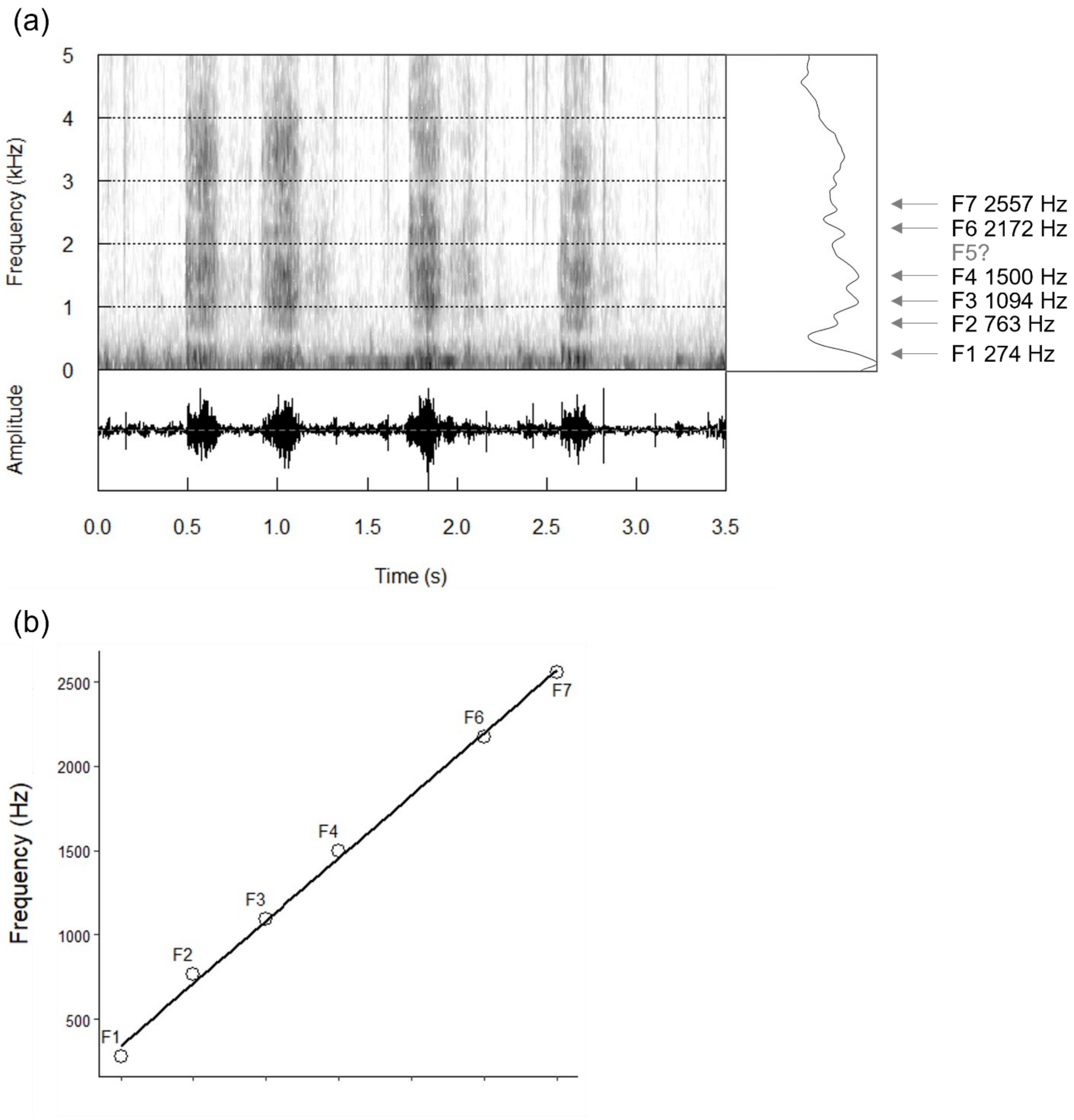

Figure 3. (a) Extraction of formants frequencies in a reindeer grunt. Only exhalation parts are selected and concatenated for cepstral smoothing. The first seven formant frequencies were identified when possible. (b) Estimation of formant spacing during a reindeer grunt using the linear regression line fitting method.

Table 1. Descriptive statistics of acoustic parameters (N = 73).

Acoustic parameter	Range	Mean ± SD	N	Detection %
F1 (Hz)	203 - 302	252.9 ± 24.8	64	87.67
F2 (Hz)	558 - 804	702.1 ± 65.3	68	93.15
F3 (Hz)	960 - 1223	1093.3 ± 64.8	71	97.26
F4 (Hz)	1310 - 1590	1451.9 ± 72.7	70	95.89
F5 (Hz)	1543 - 1866	1701.1 ± 75.7	65	89.04
F6 (Hz)	1915 - 2312	2134.3 ± 97	68	93.15
F7 (Hz)	2298 - 2644	2444 ± 94.5	70	95.89
ΔF (Hz)	373.9 - 411.4	389.6 ± 9.4	73	-
VTL (cm)	42.5 - 46.8	44.9 ± 1.1	73	-

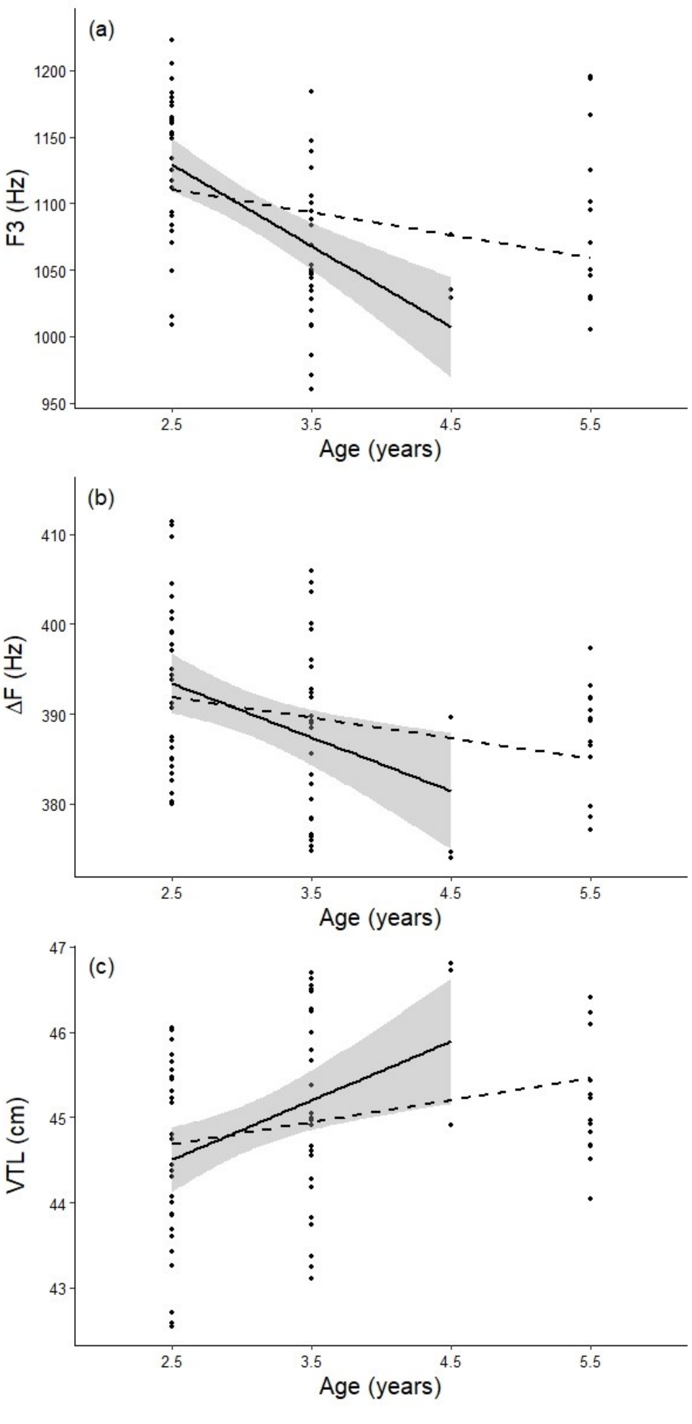

Figure 4. Scatter plots displaying the effect of age on (a) formant F3, (b) formant spacing, and (c) vocal tract length. Regression lines are shown for all males (dotted lines) (not significant) and for subadult males (aged 2.5-4.5 years) only (solid lines with grey bands representing 95% confidence intervals).

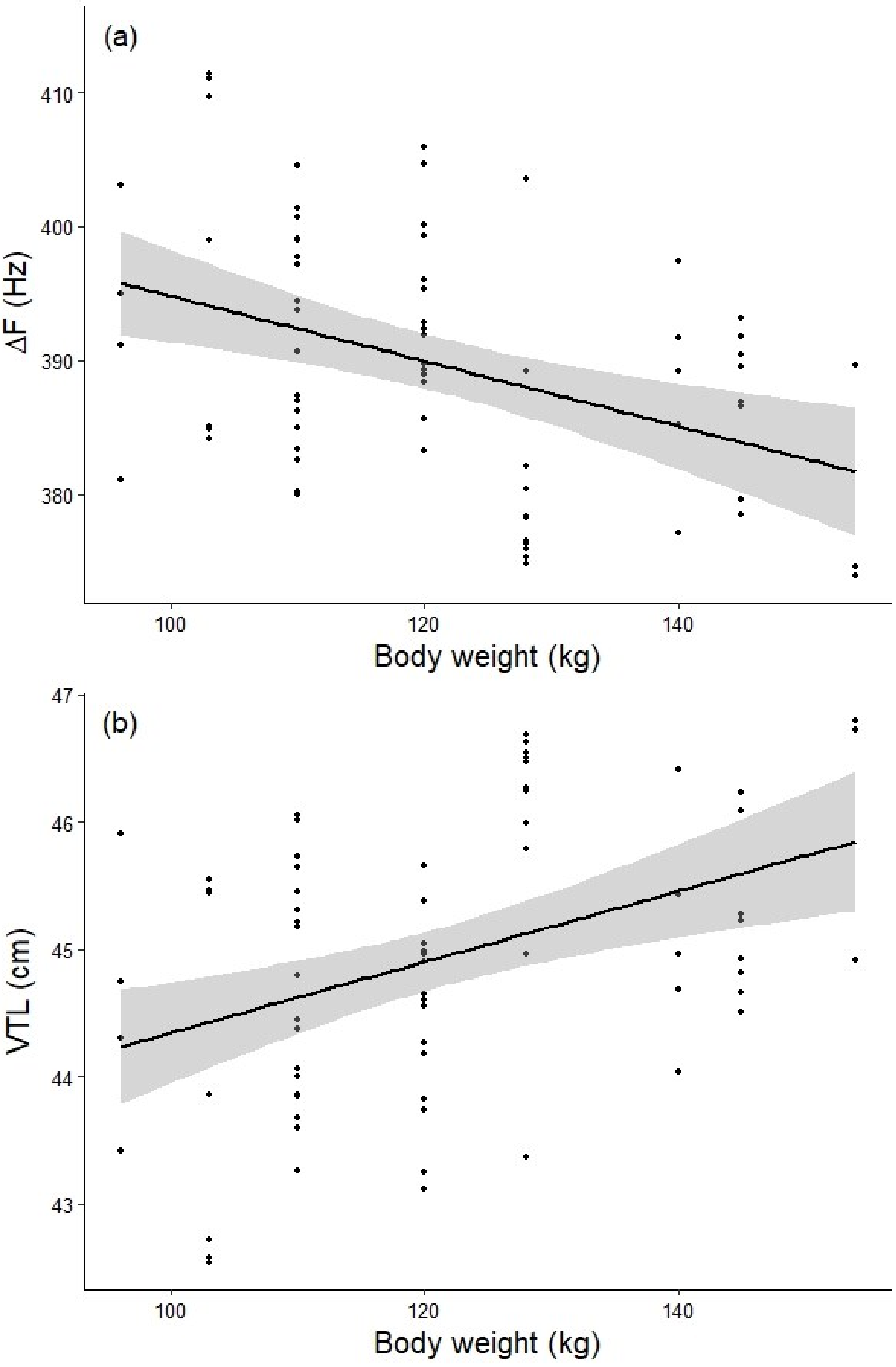

Figure 5. Scatter plots displaying the effect of body weight on (a) formant spacing, and (b) vocal tract length. Regression lines are shown with grey bands representing 95% confidence intervals.

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