ORIGINAL ARTICLE

Individual identity of alarm calls in wild-living Brandt's voles (*Lasiopodomys brandtii*)

Ilya A. Volodin[1] · Vadim E. Kirilyuk[2] · Nina A. Vasilieva[3] · Elena V. Volodina[4]

Abstract

We investigated acoustic variation responsible for the individuality of alarm calls produced by 50 Brandt's voles *Lasiopodomys brandtii* derived from 50 different wild-living colonies. For the first time, we described the calling pattern of Brandt's voles, producing a long series of short alarm calls with short inter-call intervals. The alarm calls displayed four different contours of fundamental frequency but were nevertheless strongly individually distinct within a series of 50 successive alarm calls per caller (2500 analyzed alarm calls). The average value of correct assignment of alarm calls to individuals with discriminant function analysis was 15 times higher than the value expected by chance and was robust, not decreasing with cross-validation. We discuss that the highly individualistic alarm calls provide a basis for individual recognition of callers by colony members. At the same time, heterogeneity of call contours makes the long call series less monotonous, potentially preventing habituation and promoting alertness of conspecific call recipients.

Introduction

In many taxa of small colonial mammals, alarm calls represent an important part of responses to predation risk (Blumstein 2007; Pollard 2011; Volodin et al. 2024a). Alarm calls may encode variation in a predatory context (Da Silva et al. 1994; Wilson-Henjum et al. 2019; Eddington et al. 2024) and may also reflect caller attributes, including individual identity (Conner 1985; Schibler and Manser 2007;

Matrosova et al. 2011; Volodin et al. 2018, 2021a). In the alarm calls of rodents, sex and age features are commonly less expressed than traits imparting acoustic individuality (Matrosova et al. 2007, 2011; Swan and Hare 2008; Volodina et al. 2010; Goncharov et al. 2021).

Among rodents, individually distinct alarm calls are known in marmots (Blumstein and Munos 2005; Matrosova et al. 2011), ground squirrels (Hare 1998; McCowan and Hooper 2002; Matrosova et al. 2009, 2010a, 2010b; Schneiderová and Policht 2010; Schneiderová et al. 2017; Goncharov et al. 2021) and prairie dogs (Loughry et al. 2019). Individualistic alarm calls allow conspecifics to distinguish reliable from unreliable callers that call to non-dangerous objects, e.g., cows (Hare and Atkins 2001; Blumstein et al. 2004). Individualistic alarms may also provide information on how many individuals are simultaneously calling and/or indicate the degree of urgency of antipredator responses for colony members (Weary and Kramer 1995; Wilson and Hare 2003; Blumstein et al. 2004; Sloan and Hare 2006, 2008; Thompson and Hare 2010).

Among four Arvicolinae species producing alarm calls, acoustic individuality has been investigated only for Brandt's voles *Lasiopodomys brandtii* (Rutovskaya 2012). This analysis was based on the similarity of alarm calls within audio files recorded from anonymous callers and thus constitutes

✉ Ilya A. Volodin
volodinsvoc@gmail.com

1   Department of Vertebrate Zoology, Faculty of Biology, Lomonosov Moscow State University, Vorobievy Gory, 1/12, Moscow 119234, Russia

2   Institute of Natural Resources, Ecology and Cryology, Siberian Branch of Russian Academy of Sciences, Chita 672002, Russia

3   Department of Population Ecology, Severtsov Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow 119071, Russia

4   Department of Behaviour and Behavioural Ecology of Mammals, Severtsov Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow 119071, Russia

only a preliminary study of vocal individuality (Rutovskaya 2012). While these data revealed a high degree of acoustic individuality, they have yet to be confirmed with data from individually identified callers.

The alarm calls of Brandt's vole are high-frequency and short, with a maximum fundamental frequency of 8.9–10.7 kHz and a duration of 28–33 ms (Nikolskii and Sukhanova 1992; Rutovskaya 2012). In wild Brandt's voles, alarm calls are interspersed by silence ranging from 180 to 220 ms at moderate danger when a researcher is 15–20 m from the caller to 100–180 ms where researchers are immediately adjacent to callers, followed by escape to a burrow while uttering the final call having a distinctive structure (Nikolskii and Sukhanova 1992). With increasing threat, calls also become higher-frequency, probably reflecting the increasing emotional arousal of the callers (Nikolskii and Sukhanova 1992). In addition to arousal-rate variation, geographical variation of alarm calls was reported for three wild populations of Brandt's voles, two from Mongolia and one from Russia (Rutovskaya 2012).

Brandt's voles are small rodents with body-and-head length of 110–135 mm in the wild (Allen 1940) and 94 ± 7.6 mm in captivity (Dymskaya et al. 2022). This species inhabits steppes and pastures of Mongolia, northern China, and the Transbaikalia region of Russia (Zahler et al. 2004; Enkhbold et al. 2014; Li et al. 2017). Brandt's voles live in extended family colonies consisting of a few adult males and females and their offspring (Zhong et al. 2007; Batsuren et al. 2022; Gromov 2023). Natal dispersal commences in the middle of July; however, dispersal of most animals from family colonies occurs after wintering (Gromov 2023). Brandt's voles display population outbreaks in cycles of about 5 years (Zahler et al. 2004). Over the last few decades, this species has shifted its distribution to the north and west and reduced its range in the south perhaps owing to the influence of global warming (Enkhbold et al. 2014; Bai et al. 2022). The main predators of Brandt's voles are corsacs *Vulpes corsac*, steppe polecats *Mustela eversmanni*, Pallas' cats *Otocolobus manul*, saker falcons *Falco cherrug*, upland buzzards *Buteo hemilasius* and steppe eagles *Aquila nipalensis* (Samjaa et al. 2000). Brandt's vole is an important plague vector (Xu et al. 2015; Guo et al. 2023) and has a significant impact on vegetation (Samjaa et al. 2000; Cui et al. 2020).

The aim of this study was to evaluate individuality in the alarm calls of wild-living Brandt's voles and to estimate the relationship between the individualization of alarm calls and their acoustic structure. We investigated acoustic parameters of alarm calls emitted within a prolonged series of 50 successive calls from each of 50 individual Brandt's voles, one 50-call sequence per individual, produced in the presence of a surrogate predator (a standing researcher). This experimental design models a single predatory event, during which conspecific receivers would acquire information about the individual identity of a caller from a single series of alarm calls. Previously, this approach proved useful for evaluating the individuality of alarm calls in wild-living ground squirrels (McCowan and Hooper 2002; Matrosova et al. 2011) and pikas (Volodin et al. 2018, 2021a).

Methods

Study site and subjects

Alarm calls of Brandt's voles were recorded in Dauria (Transbaikalia, Russia) around the Teli field station of the Daurian Nature Reserve, from 29 June to 14 July 2019 (50.06° N, 115.44° E). This area is a grassy steppe undulating at elevations of 600–1100 m (Kirilyuk et al. 2013; Obyazov et al. 2021). The colonies of Brandt's voles were located on bottoms of dried lakes Zun-Torey and Barun-Torey and along the shore of the lake Zun-Torey and the wide isthmus between the lakes. 2019 was the end of a dry period before the return of a wet period within the 25–30-year water cycle of these lakes (Obyazov et al. 2021). The Brandt's voles preferred the hard-packed soil and stone grounds and avoided plots with high grass. The population peaked in 2019; the minimal distance between neighboring family-based colonies of Brandt's voles was about 70–100 m.

Colonies of Brandt's voles were clearly identifiable by degraded vegetation and visible open burrow entrances. The colonies contained both adults and pups of the current year; pups were from half to two-thirds of adult size. The Brandt's voles mainly produced their alarm calls from burrow entrances, with only part of their head visible outside (Supplementary information Movie S1). As callers were visible to researchers in less than half of the cases and only partially, their age and sex could not be identified, but for vole species, precise age estimation is commonly complicated even when the animals are in researcher's hands and may require sophisticated instrumental methods (Nikonova et al. 2024).

Acoustic recording

For acoustic recording (sampling rate 48 kHz, 16-bit resolution), we used two solid-state digital recorders (Marantz PMD-660; D&M Professional, Kanagawa, Japan) with hand-held microphones (Sennheiser K6-ME66; Sennheiser electronic, Wedemark, Germany), having a flat frequency response (± 2.5 dB) from 0.04 to 20 kHz. Acoustic recordings were made primarily in the first half of the day (from 10:00 to 12:00 approximately), avoiding recording during strong winds, which compromised the quality of call recordings.

Although individuals were unmarked, recordings of alarm calls were done on an individual basis, by collecting a single prolonged series of alarm calls from each focal individual calling from one point uninterruptedly. Researchers (IAV or EVV) slowly walked (about 2 km/h) from colony to colony and stopped when they heard an individual Brandt's vole producing alarm calls. Each researcher recorded calls alone, but in a similar way, remaining motionless at a distance of 12–20 m from the focal caller. Recording from a fixed position dictated by the onset of calling in response to an approaching researcher provides a standardized approach to data collection that can be applied in subsequent studies. Alarm calls of Brandt's voles were rather faint and could be recorded at good quality from distances not exceeding 15–20 m. A researcher tried to record successive alarm calls for as long as possible (at least 50 alarm calls) from each focal individual (one per family colony). The recording lasted until the caller escaped to the burrow, or if the recording was interrupted by strong wind. After the end of the recording, each researcher walked to another family colony (located at least 70–100 m from the preceding colony) to record a new focal individual.

To avoid repeated recordings of the same individuals, researchers never visited the same colony again. Thus, each recording session included only one focal individual, associated with a particular colony. Each recording was stored as a separate wav-file. In total, two researchers collected 93 acoustic recordings from 93 different focal individuals.

Call samples

The selection of alarm calls for acoustic analyses was conducted using Avisoft SASLab Pro software v. 5.2.12 (Avisoft Bioacoustics, Berlin, Germany). As a rule, each series of alarm calls from one focal individual included several hundred alarm calls. For acoustic analyses, we selected 50 audio files, one file per individual Brandt's vole, each containing 50 successive alarm calls of good quality, appropriate for analysis of acoustic parameters, 2500 alarm calls in total. Criteria for selection were series unbroken by wind and not superimposed by calls of other Brandt's voles. Each series of alarm calls was recorded by a researcher, standing immobile, so, intervals between calls within series were approximately equal. Among individuals, the total length of the sequence of the successive 50 alarm calls varied from 6.38 to 12.75 s, on average, 9.96 ± 1.46 s.

Call analyses

Before analyses, calls were high-pass filtered at 1.0 kHz to reduce the low-frequency background noise using Avisoft, because the lowest fundamental frequency (f0) of alarm calls in the Brandt's vole always exceeds 2 kHz (Nikolskii and Sukhanova 1992; Rutovskaya 2012). In each of the 2500 alarm calls, we manually measured 7 acoustic parameters: two temporal, three variables of f0 and two power parameters (Fig. 1). We measured call duration with the standard marker cursor in the spectrogram window of Avisoft (sampling frequency 48 kHz, Hamming window, FFT 512 points, frame 50% and overlap 93.75%; frequency resolution 94 Hz and temporal resolution of 0.67 ms). We measured the beginning (f0beg) and end (f0end) fundamental frequency at the onset and end point of a call and the maximum fundamental frequency (f0max) at call contour maximum with the free reticule cursor (Fig. 1). Over the entire call power spectrum (between the onset
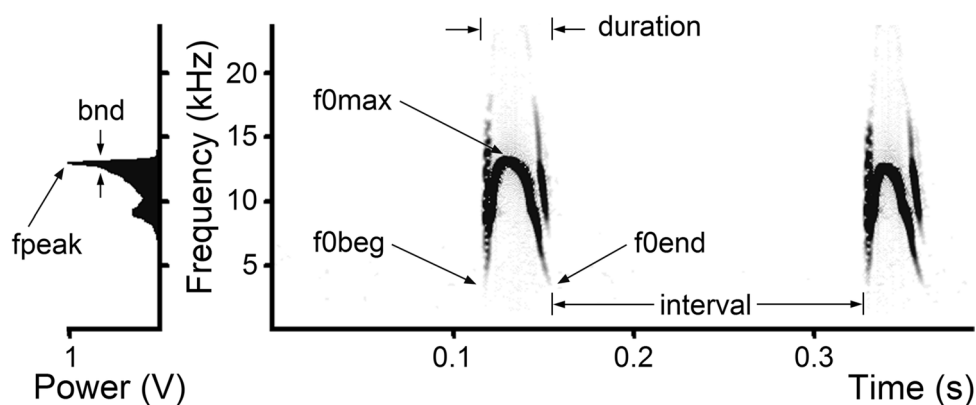

Fig. 1 Measured acoustic parameters for the alarm calls of wild Brandt's voles. Spectrogram (right) and the mean power spectrum of the first call (left). Designations: duration, call duration; interval, the inter-call interval from the end of a preceding call to the start of the next call; f0max, the maximum fundamental frequency; f0beg, the fundamental frequency at the onset of a call; f0end, the fundamen-tal frequency at the end of a call; fpeak, the frequency of maximum amplitude within a call; bnd, the bandwidth of fpeak at the distance of −10 dB from the maximum. The spectrogram was created at 48 kHz sampling frequency, FFT length 512, Hamming window, frame 50%, overlap 93.75%

and end point of a call), we measured fpeak, representing the value of the maximum amplitude frequency, and bandwidth (bnd) of fpeak at minus 10 dB from the maximum (Fig. 1). We measured the inter-call interval, from the end of a preceding call to the start of the next call (Fig. 1). All measurements were exported automatically to Microsoft Excel (Microsoft Corp., Redmond, Washington).

Each alarm call was classified based on f0max and fpeak values and contour shapes: "basic", "middle-peak upstretch", "high-peak upstretch" and "low-frequency" (Fig. 2, see "Results" for details). We also checked each alarm call for the presence of subharmonics and biphonations (Fig. 1) following similar analyses of nonlinear vocal phenomena (Wilden et al. 1998) in squeaks of vole species (Volodin et al. 2021b; Dymskaya et al. 2022). We scored the presence of nonlinear vocal phenomena only if the call portion containing subharmonics or biphonations exceeded 10% of the total call duration (Volodin et al. 2021b; Dymskaya et al. 2022, 2024; Rutovskaya et al. 2024).

Statistical analyses

All statistical analyses were carried out with STATISTICA v. 13 (StatSoft, Tulsa, OK, USA). We also used R 4.1.0 (R Development Core Team 2022) for conducting the randomization test for the probability of incorrect classification based on Solow (1990) using a custom-made script created with R. Descriptive statistics were indicated as mean $\pm SD$, all tests were two-tailed and differences were considered significant whenever $P < 0.05$.

Since we aimed to compare different sources of variability in the sample of alarm calls, we employed several methods of analysis of variance. We used a one-way ANOVA, to compare within and between individual variation of acoustic parameter values for the 7 measured acoustic parameters. We used a two-way ANOVA with Unequal N HSD for the unequal call samples with inclusion in analysis animal ID (individual identity) as a random factor to compare the variability of acoustic parameters between alarm calls with different contours
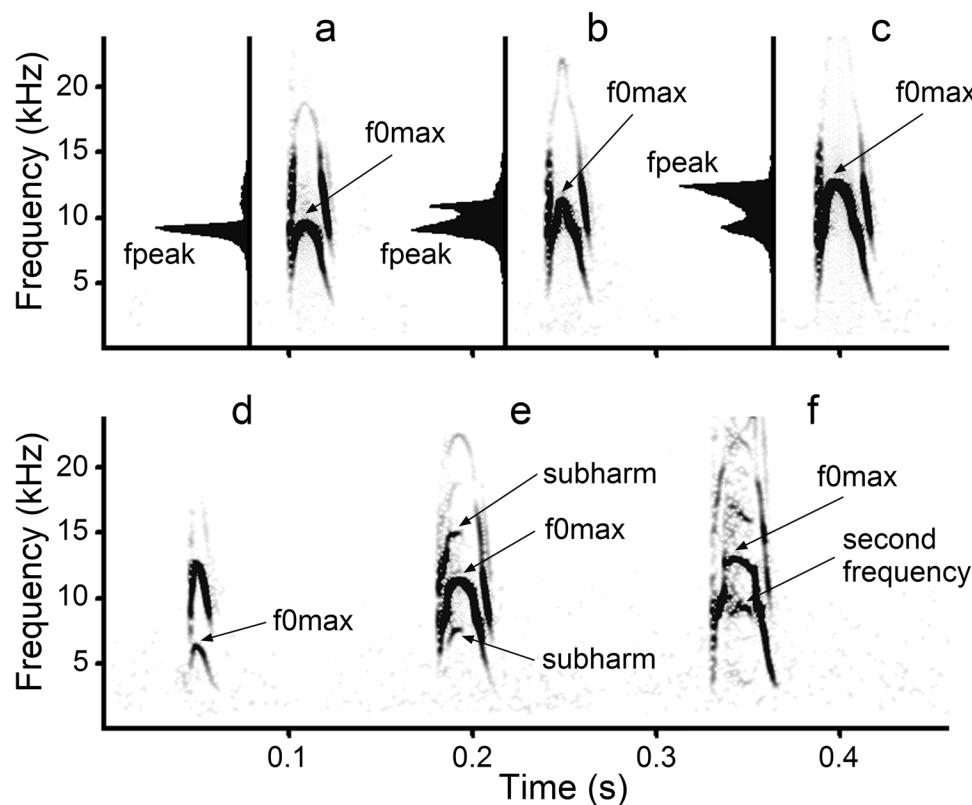

Fig. 2 Spectrograms illustrating four different contours of fundamental frequency (f0), subharmonics and biphonation in the alarm calls of Brandt's voles. (**a**) Basic contour; the mean power spectrum of this call is left of the spectrogram; (**b**) the middle-peak upstretch contour; the mean power spectrum of this call is left of the spectrogram; (**c**) the high-peak upstretch contour, the mean power spectrum of this call is left of the spectrogram; (**d**) the low-frequency contour; (**e**) an alarm call with subharmonics; (**f**) an alarm call with biphonation. Designations: f0max, the maximum fundamental frequency; fpeak, the frequency of maximum amplitude within a call; subharm, subharmonics of 1/3 of f0; second frequency, the second independent fundamental frequency of the biphonic call. The spectrogram was created at 48 kHz sampling frequency, FFT length 512, Hamming window, frame 50%, overlap 93.75%

of fundamental frequency. We used discriminant function analysis (DFA), to calculate the probability of assignment of alarm calls to the correct individual. The DFA standard (complete) procedure in STATISTICA includes a built-in option which tests variables for correlations and does not allow the inclusion of highly correlated variables in the analysis. All these variables met DFA assumptions.

For validation of DFA results, we used half-and-half cross-validation on the basis of odd (training set) and even (test set) order numbers of the alarm calls of each individual. The training set contained 1250 alarm calls, 25 per individual, from 50 subject animals. The test set contained the remaining 1250 alarm calls, 25 per individual from 50 subjects. DFA for the test set was conducted using discriminant functions, generated by the acoustic variables of the calls from the training set.

We used Wilks' Lambda values to estimate how strongly the acoustic variables contribute to the discrimination of individuals. We used repeated measures ANOVA to compare the values of correct assignment of calls to individuals between DFAs. For validating the DFA results, we calculated the probability of correctly classifying calls to individuals, by applying the randomization test for the probability of incorrect classifying in DFA with a custom-made script created in R based on Solow (1990). Random values were calculated from DFA on 1000 randomized permutations of datasets (Solow 1990; Mundry and Sommer 2007). For each distribution obtained with permutations, we noted whether the observed value exceeds 95% (950 values), 99% (990 values) or 99.9% (999 values) within the distribution (Solow 1990; Mundry and Sommer 2007). If the observed value exceeded 95%, 99% or 99.9% of values within this distribution, we established that the observed value differed significantly from the random value with a probability $P < 0.05$, $P < 0.01$ or $P < 0.001$, respectively (Solow 1990; Briefer et al. 2010; Chelysheva et al. 2023; Sedova et al. 2023).

For each individual, on the basis of percentages of alarm calls with particular contours, we calculated the index of qualitative variation *IQV* (Wilcox 1973):

$$IQV = K \times (100^2 - \Sigma\, p^2)/100^2 \times (K - 1).$$

where K is the number of categories (4 in this study), p is the percentage of alarm calls with a particular contour and $\Sigma\, p^2$ is the sum of all squared percentages. *IQV* can vary from 0 to 1, where 0 indicates no variability and 1 indicates maximum variability (Wilcox 1973; Chrdileli and Borchia 2024).

To evaluate the variation of acoustic variables for each individual, we calculated the coefficient of variation (*CV*) for each alarm call variable. We used Pearson's correlation (with Bonferroni correction) between *IQV*s, coefficients of variation (*CV*s) and mean values of acoustic parameters of alarm calls from one side and percent of calls correctly assigned with DFAs to individuals from another side. We used a one-way ANOVA to compare *IQV*s, *CV*s and the means of acoustic variables between callers with high and low individuality of alarm calls.

Ethical statement

The authors adhered to the "Guidelines for the treatment of animals in behavioural research and teaching" (Anim. Behav., 2020, 159, I-XI) and the legal requirements of Russia pertaining to the protection of animal welfare (StateStandard 3 53434–2009). The recording procedure was purely observational and was approved by the Committee of Bio-ethics of Lomonosov Moscow State University, research protocol # 2011–36.

Results

Alarm call acoustics

Alarm calls displayed a rapid increase of f0 from about 5.04 kHz at call onset to about 11.84 kHz at the point of the f0max, followed by a rapid decrease of f0 to about 4.93 kHz at call end (Table 1). Call duration was about 30.42 ms (ranging from 17 to 42 ms); the inter-call interval for successive alarm calls was about 169.7 ms. The peak frequency of the alarm calls was about 10.87 kHz, and the bandwidth of the peak frequency was about 2.07 kHz (Table 1).

Table 1 Mean $\pm SD$ and min–max values for acoustic variables of alarm calls of 50 individual Brandt's voles (calculated as the average of within-individual averages) and ANOVA results for individual differences. Designations: *duration*, call duration; *f0beg*, the fundamental frequency at the onset of a call; *f0max*, the maximum fundamental frequency; *f0end*, the fundamental frequency at the end of a call; *fpeak*, the frequency of maximum amplitude; *bnd*, the bandwidth of the frequency of maximum amplitude; *interval*, inter-call interval, from the end of a preceding call to the start of the next call; $N = 50$ individuals

Acoustic variable	Mean $\pm SD$	Min–Max	ANOVA
duration (ms)	30.42 ± 4.69	17.07–42.18	$F_{49,2450} = 95.81$, $P < 0.001$
f0beg (kHz)	5.04 ± 1.07	3.14–7.98	$F_{49,2450} = 223.9$, $P < 0.001$
f0max (kHz)	11.84 ± 1.38	7.65–13.87	$F_{49,2450} = 155.3$, $P < 0.001$
f0end (kHz)	4.93 ± 1.44	3.06–10.99	$F_{49,2450} = 398.4$, $P < 0.001$
fpeak (kHz)	10.87 ± 1.24	8.10–13.47	$F_{49,2450} = 46.92$, $P < 0.001$
bnd (kHz)	2.07 ± 0.70	0.90–4.26	$F_{49,2450} = 30.53$, $P < 0.001$
interval (ms)	168.7 ± 29.7	97.4–226.4	$F_{49,2450} = 21.90$, $P < 0.001$

On the basis of the f0max and fpeak values and correspondence between those, we classified each alarm call into one of four contours: basic, middle-peak upstretch, high-peak upstretch and low-frequency (Fig. 2, Table 2). Calls with basic contours had moderate f0max, coinciding with fpeak. Calls with middle-peak upstretch contours had on the top of the call contour an additional short elevation of about 2 kHz, having either the form of a dropper or a short plateau (Fig. 2). The fpeak of alarm calls with middle-peak upstretch contours was located at approximately the same area as fpeak of calls with basic contours (Table 2). Calls with the high-peak upstretch contours had a more pronounced rise of f0; their fpeak values were shifted up and coincided with values of f0max (Fig. 2, Table 2). Calls with low-frequency contours appeared to be incomplete calls with basic contours with f0 rising quickly and then declining, resulting in a shorter duration triangle-shaped f0 run (Fig. 2, Table 2). The ANOVA revealed pronounced differences in all measured acoustic variables between alarm calls with basic, middle-peak upstretch and high-peak upstretch contours of f0 (Table 2). Calls with low-frequency contours were excluded from the analysis because of insufficient sample size, with only 16 calls manifesting this contour in total (Table 2).

Nonlinear phenomena occurred in only 7 of 50 individuals (2 had biphonations, 2 had subharmonics and 3 had biphonations and subharmonics in different calls) (Fig. 2). Biphonations were noted only in 84 (3.36%) alarm calls of 5 individuals (from 1 to 38 calls per individual). Subharmonics were noted only in 42 (1.68%) alarm calls of 5 individuals (from 1 to 21 alarm calls per individual).

Individuality of alarm calls

One-way ANOVA revealed pronounced individual differences in all measured acoustic variables of the alarm calls (Table 1). We also conducted three DFAs for assigning alarm calls to correct individuals (Table 3). DFA1, based on the 7 measured acoustic variables, classified the total sample

of 2500 calls to 50 individuals with an accuracy of 65.52%, which was significantly higher than the random level of $4.32 \pm 0.35\%$, min $= 3.24\%$, max $= 5.68\%$ (permutation test, 1000 permutations, $P < 0.001$). The cross-validating DFA2, conducted on the training set of 1250 alarm calls (odd calls in the 50 individual series of alarm calls), classified the calls to individuals with an accuracy of 64.72%. The cross-validating DFA3, conducted on the test set of remaining 1250 alarm calls (even calls in the 50 individual series of alarm calls) using the discriminant functions created for the training set, classified calls to individuals with an accuracy of 64.24% (Table 3).

Classifying accuracy to individuals did not differ between the three DFAs: between DFA1 and DFA2 (repeated measures ANOVA, $F_{1,49} = 0.67$, $P = 0.42$); between DFA1 and DFA3 ($F_{1,49} = 1.08$, $P = 0.30$); and between DFA2 and DFA3 ($F_{1,49} = 0.08$, $P = 0.77$). These results confirm the uniformity of acoustic structures within individuals, providing the basis for the individual-specific alarm calls in Brandt's vole.

Alarm calls of all 50 animals were classified significantly above chance. Both DFA1 on the total sample of calls and cross-validating DFA3 classified less than 40% of alarm calls to correct callers (min $= 24\%$) only in 7 individuals; in all remaining cases, 40 to 100% of calls were classified to correct callers (Table 3). The first two discriminant functions accounted for 73.72% of the variation, four functions had eigenvalues over 1, and for complete discrimination, 7 discriminant functions were necessary (Table 4). The Wilks' Lambda values revealed that, in order of decreasing importance, f0end, f0max and duration were mainly responsible for the discrimination of individuals (Table 4). Canonical correlation analysis in DFA revealed that different acoustic variables of alarm calls were correlated with different discriminant functions (Table 4). This result indicates that all measured acoustic variables of alarm calls were necessary for the reliable discrimination of individuals.

Based on the values of correct assignment of alarm calls to individuals in the three DFAs, we assigned individuals to

Table 2 Mean $\pm$ SD values for acoustic variables of alarm calls with four different contours of f0 (basic, middle-peak upstretch, high-peak upstretch, low-frequency) and ANOVA results for their comparison (for the exception of calls with the low-frequency contours, because of insufficient sample size). Vole ID is included as a random factor. Designations of acoustic variables are as in Table 1. Different superscripts indicate statistically different values (Unequal N HSD, $P < 0.05$), N number of calls

Acoustic variable	Basic contour (N = 458)	Middle-peak upstretch contour (N = 676)	High-peak upstretch contour (N = 1350)	Low-frequency contour (N = 16)	ANOVA
duration (ms)	29.95 ± 6.74 [a]	29.02 ± 5.48 [b]	31.43 ± 5.12 [c]	17.46 ± 1.70	$F_{2,2432} = 338.9, P < 0.001$
f0beg (kHz)	4.64 ± 1.32 [a]	5.08 ± 1.28 [b]	5.16 ± 1.01 [c]	4.04 ± 1.06	$F_{2,2432} = 10.26\ P < 0.001$
f0max (kHz)	9.71 ± 1.10 [a]	12.19 ± 1.20 [b]	12.43 ± 1.10 [c]	7.15 ± 0.81	$F_{2,2432} = 2024.9, P < 0.001$
f0end (kHz)	4.77 ± 1.59 [a]	5.00 ± 1.73 [b]	4.96 ± 1.36 [b]	3.84 ± 1.04	$F_{2,2432} = 32.60, P < 0.001$
fpeak (kHz)	9.73 ± 1.40 [a]	9.16 ± 0.67 [b]	12.14 ± 1.11 [c]	7.57 ± 1.63	$F_{2,2432} = 3373.6, P < 0.001$
bnd (kHz)	1.21 ± 0.51 [a]	2.66 ± 1.05 [b]	2.08 ± 1.12 [c]	1.87 ± 1.34	$F_{2,2432} = 208.0, P < 0.001$
interval (ms)	158.6 ± 56.2	174.0 ± 58.1	169.6 ± 49.3	161.4 ± 44.0	$F_{2,2432} = 2.43, P = 0.09$

Table 3 Individual-based DFA results for acoustic analyses, numbers of alarm calls with different contours and index of qualitative variation (*IQV*) of the contours for each of the 50 individual Brandt's voles. The *IQV* can vary from 0 to 1, where 0 indicates no variability and 1 indicates maximum variability. Designations: *DFA1*, DFA on the total sample of 2500 alarm calls; *DFA2*, DFA on the training set of 1250 alarm calls, odd in series; *DFA3*, DFA on the test set of 1250 alarm calls, even in series; *High-Low*, labels of individuals with high or low individuality of alarm calls

Vole ID	Percent of alarm calls correct assignment				Alarm call contour				IQV
	DFA1	DFA2	DFA3	High-Low	Basic	Middle-peak upstretch	High-peak upstretch	Low-frequency	
1	70	64	76		0	11	39	0	0.458
2	80	84	88	High	48	0	0	2	0.102
3	92	92	84	High	50	0	0	0	0
4	26	32	40	Low	33	14	3	0	0.643
5	96	76	68		3	4	43	0	0.334
6	94	88	100	High	3	33	14	0	0.643
7	82	84	72		3	7	40	0	0.449
8	92	92	88	High	26	4	20	0	0.751
9	100	100	100	High	2	9	39	0	0.477
10	24	16	32	Low	0	9	41	0	0.394
11	98	96	100	High	2	24	24	0	0.717
12	54	52	48		0	36	14	0	0.538
13	72	80	72		2	38	8	2	0.525
14	68	64	60		0	3	47	0	0.150
15	32	20	24	Low	2	28	19	1	0.720
16	44	48	44	Low	25	5	17	3	0.828
17	36	48	32	Low	0	5	45	0	0.240
18	90	88	96	High	2	14	32	2	0.678
19	84	76	92		48	2	0	0	0.102
20	66	64	60		8	38	4	0	0.521
21	70	72	68		0	6	44	0	0.282
22	30	24	28	Low	0	17	33	0	0.598
23	92	92	96	High	1	47	2	0	0.153
24	36	36	44	Low	5	15	29	1	0.751
25	34	44	24	Low	12	18	20	0	0.870
26	50	48	44	Low	49	0	0	1	0.052
27	84	80	84	High	24	19	5	2	0.818
28	54	52	68		0	19	31	0	0.628
29	58	48	60		2	33	15	0	0.630
30	42	52	40		9	20	21	0	0.842
31	90	92	88	High	39	7	4	0	0.487
32	50	56	48		6	28	16	0	0.759
33	58	72	56		0	1	49	0	0.052
34	82	88	72		0	6	44	0	0.282
35	88	80	88	High	1	2	47	0	0.153
36	56	60	48		0	30	20	0	0.640
37	46	52	44		11	35	4	0	0.607
38	90	88	92	High	0	0	50	0	0
39	68	72	68		0	1	49	0	0.052
40	60	48	80		2	0	48	0	0.102
41	48	40	52		3	4	42	1	0.379
42	70	64	60		1	5	44	0	0.287
43	46	52	36		0	17	33	0	0.598
44	60	52	60		0	1	49	0	0.052
45	74	80	64		2	5	42	1	0.377
46	62	56	72		31	16	3	0	0.679
47	56	56	32		0	2	48	0	0.102
48	56	48	60		0	0	50	0	0

Table 3 (continued)

Vole ID	Percent of alarm calls correct assignment				Alarm call contour				IQV
	DFA1	DFA2	DFA3	High-Low	Basic	Middle-peak upstretch	High-peak upstretch	Low-frequency	
49	78	84	68		0	21	29	0	0.650
50	88	84	92	High	3	17	30	0	0.694
Total	65.52	64.72	64.24		458	676	1350	16	

Table 4 Results of DFA1 analysis on the total sample of alarm calls and the values of correlation between acoustic variables and the seven discriminant functions; the eigenvalues, and percentage of variance, described by each function. Values of correlation exceeding 0.5 are labeled in bold. Designations of acoustic variables are as in Table 1

Acoustic variable	Wilks' Lambda	F-to-remove	Canonical correlation analysis with DFA						
			Function 1	Function 2	Function 3	Function 4	Function 5	Function 6	Function 7
duration	0.002165	$F_{49,2444} = 151.9$; $P < 0.001$	0.169	0.232	−0.472	**0.755**	0.138	−0.288	−0.152
f0beg	0.001747	$F_{49,2444} = 113.0$; $P < 0.001$	−0.423	−0.473	**−0.692**	−0.338	0.0247	0.009	0.061
f0max	0.002678	$F_{49,2444} = 199.8$; $P < 0.001$	0.211	**−0.569**	−0.275	**0.654**	0.075	−0.351	0.016
f0end	0.003061	$F_{49,2444} = 235.5$; $P < 0.001$	**−0.846**	−0.475	−0.012	0.217	0.060	−0.092	0.010
fpeak	0.000859	$F_{49,2444} = 30.23$; $P < 0.001$	0.095	−0.184	−0.144	0.300	**0.915**	−0.047	−0.08
bdw	0.000844	$F_{49,2444} = 28.78$; $P < 0.001$	0.065	−0.065	0.108	−0.242	−0.085	**−0.939**	0.181
interval	0.000755	$F_{49,2444} = 20.53$; $P < 0.001$	0.001	−0.099	−0.008	−0.137	−0.089	−0.075	**−0.979**
Eigenvalue			9.16	6.01	2.86	1.11	0.58	0.49	0.37
Percent of variance			44.5%	29.21%	13.88%	5.40%	2.83%	2.38%	1.78%

two groups: with high and with low individuality of alarm calls (Table 3). The group of individuals with the high individuality (13 individuals) had high (over or equal to 80%) values of correct assignment of alarm calls to individuals in all three DFAs. The group of individuals with low individuality (9 individuals) had low (below or equal to 50%) values of correct assignment of alarm calls to individuals in all three DFAs (Table 3).

Individual variation of acoustic variables

For estimating the degree of individualization of alarm calls, we used DFA-based values of correct assignment of calls to individuals (Table 3). A within-individual series of 50 successive alarm calls could either contain acoustically similar calls or consist of calls strongly differing in contours and acoustic variables (Fig. 3). For estimating variation in amounts of calls with different contours for each individual, we calculated amounts of calls with particular contours (Table 3). Using these data, we further calculated the index

of qualitative variation (*IQV*), which can vary from 0 to 1, where 0 indicates no variation and 1 indicates maximum variation (Table 3). Only in 3 of 50 callers all alarm calls had the same contour within their series of 50 successive alarm calls ($IQV = 0$).

We did not find correlations between percentages of alarm calls correctly assigned to individuals with three DFAs and *IQV* coefficients (DFA1: $r = -0.22$, $P = 0.12$; DFA2: $r = -0.17$, $P = 0.24$; DFA3: $r = -0.19$, $P = 0.19$; $n = 50$ in all cases). One-way ANOVA did not reveal differences ($F_{1,20} = 1.01$, $P = 0.33$) in *IQV* coefficients between callers with high ($IQV = 0.436 \pm 0.309$) and low ($IQV = 0.566 \pm 0.280$) individuality of alarm calls (Table 3).

For estimating the individual variation of acoustic variables for each caller, we calculated the coefficient of variation (*CV*) for each acoustic variable (Supplementary information Table S3). We found a negative correlation between the values of the correct assignment of calls to individuals with DFA2 and *CV* of f0end and also a negative correlation between the values of the correct
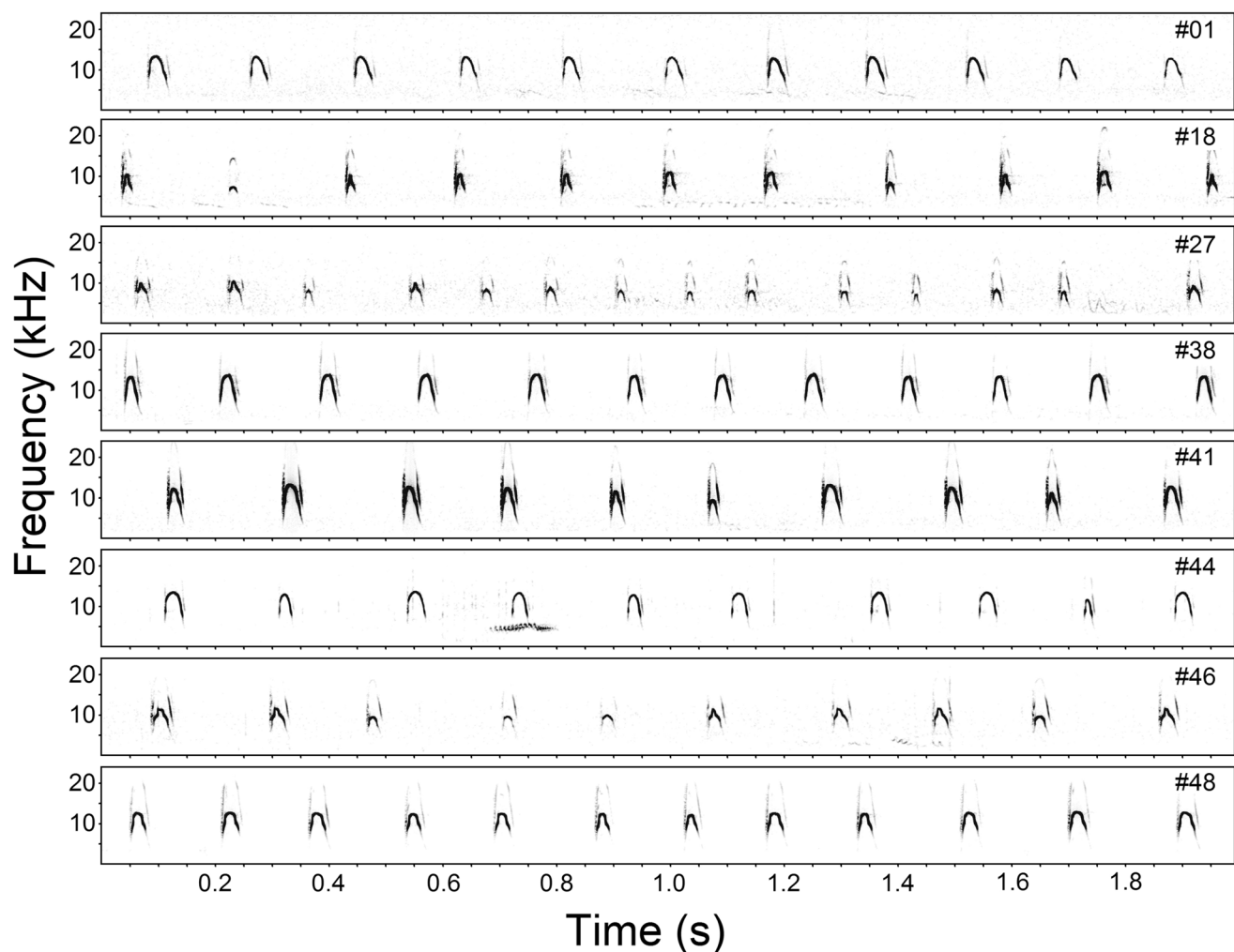
Fig. 3 Spectrograms illustrating the within and between-individual acoustic variation of alarm calls in natural 2-s long segments of alarm calling toward a human researcher in 8 individual wild Brandt's voles. Vole ID numbers are indicated in the upper right corners. The indexes of qualitative variation (*IQVs*) of the contours for individual Brandt's voles are given in Table 3. The spectrograms were created at 48 kHz sampling frequency, FFT length 512, Hamming window, frame 50%, overlap 93.75%. An audio file with these calls is provided in Supplementary information Audio S2

assignment of the calls to individuals with DFA3 and *CV* of bandwidth (Table 5). The *CV*s of f0end, bandwidth and fpeak showed marginally significant negative correlations with the values of correct assignment of alarm calls to individuals with all three DFAs (Table 5). We did not find correlations between percent of calls correctly assigned to individuals with three DFAs and the mean value of any acoustic variable (Table 5, Supplementary information Table S4). One-way ANOVA revealed that *CV*s of f0end, bandwidth and fpeak were lower in callers with high individuality of their alarm calls, whereas the mean values of acoustic variables did not differ between groups of callers with high alarm call individuality, with the exception of marginally significant differences in fpeak (Table 6).

Discussion

In this study, we described, for the first time, that antipredator vocalizations of Brandt's voles represent a very long series of short alarm calls with four different contour shapes, emitted with short inter-call intervals (Tables 1 and 2). The production of a long series of alarm calls might help receivers perceive the auditory signal as belonging to the same individual when noise masks or degrades part of the signal, e.g., strong wind, as is typical in Brandt's vole habitat. Call receivers experiencing an auditory continuity illusion, extracting call rate information where syllables within repeated calls are obscured by noise, have been

Table 5 Pearson's correlation coefficients between coefficients of variation (*CV*) and the mean values of acoustic variables of alarm calls and percent of alarm calls correctly assigned to individuals with three DFAs (DFA1 based on the total sample of 2500 alarm calls; DFA2 based on the training set of 1250 alarm calls, odd in series; and DFA3 based on the test set of 1250 alarm calls, even in series). Significant correlations after Bonferroni correction ($P=0.05/7=0.0071$) are labeled in bold, $N=50$ individuals. Designations of acoustic variables are as in Table 1

Acoustic variable	Coefficients of variation (*CV*s)			Means		
	DFA1	DFA2	DFA3	DFA1	DFA2	DFA3
duration	$r=-0.13, P=0.35$	$r=-0.19, P=0.18$	$r=-0.12, P=0.42$	$r=0.02, P=0.87$	$r=-0.02, P=0.87$	$r=-0.04, P=0.77$
f0beg	$r=-0.06, P=0.67$	$r=-0.04, P=0.77$	$r=-0.01, P=0.97$	$r=0.08, P=0.60$	$r=-0.02, P=0.90$	$r=0.07, P=0.64$
f0max	$r=-0.14, P=0.32$	$r=-0.13, P=0.38$	$r=-0.09, P=0.53$	$r=-0.25, P=0.09$	$r=-0.26, P=0.06$	$r=-0.28, P=0.049$
f0end	$r=-0.36, P=0.010$	$\boldsymbol{r=-0.41, P=0.003}$	$r=-0.34, P=0.016$	$r=0.15, P=0.29$	$r=0.11, P=0.46$	$r=0.17, P=0.23$
fpeak	$r=-0.29, P=0.037$	$r=-0.30, P=0.037$	$r=-0.35, P=0.013$	$r=-0.20, P=0.16$	$r=-0.24, P=0.09$	$r=-0.25, P=0.08$
bdw	$r=-0.35, P=0.013$	$r=-0.29, P=0.039$	$\boldsymbol{r=-0.41, P=0.003}$	$r=-0.04, P=0.80$	$r=0.06, P=0.67$	$r=-0.07, P=0.61$
interval	$r=-0.14, P=0.35$	$r=-0.16, P=0.27$	$r=-0.15, P=0.30$	$r=-0.15, P=0.31$	$r=-0.24, P=0.09$	$r=-0.12, P=0.39$

Table 6 Mean $\pm SD$ values of coefficients of variation (*CV*s), mean $\pm SD$ values of the means for acoustic variables of callers with high or low individuality of alarm calls and ANOVA results for differences between the groups of individuals with highly and lowly individualistic alarms. *N* number of individuals. Designations of acoustic variables are as in Table 1

Acoustic variable	Coefficients of variation		ANOVA	Means		ANOVA
	High ($N=13$)	Low ($N=9$)		High ($N=13$)	Low ($N=9$)	
duration	$10.38\pm3.81\%$	$12.50\pm3.75\%$	$F_{1,20}=1.77, P=0.20$	29.84 ± 7.24 ms	29.77 ± 3.02 ms	$F_{1,20}=0.01, P=0.98$
f0beg	$9.90\pm2.80\%$	$10.92\pm3.57\%$	$F_{1,20}=0.57, P=0.46$	4.92 ± 1.47 kHz	4.70 ± 0.95 kHz	$F_{1,20}=0.16, P=0.69$
f0max	$5.92\pm3.04\%$	$7.42\pm4.12\%$	$F_{1,20}=0.98, P=0.33$	10.94 ± 1.43 kHz	11.71 ± 1.73 kHz	$F_{1,20}=1.30, P=0.27$
f0end	$7.89\pm1.81\%$	$11.22\pm3.67\%$	$F_{1,20}=8.02, P=0.01$	5.11 ± 2.36 kHz	4.60 ± 0.60 kHz	$F_{1,20}=0.39, P=0.54$
fpeak	$8.33\pm3.87\%$	$14.56\pm7.62\%$	$F_{1,20}=6.40, P=0.02$	10.07 ± 1.33 kHz	11.05 ± 0.61 kHz	$F_{1,20}=4.30, P=0.051$
bdw	$32.25\pm14.68\%$	$47.03\pm13.03\%$	$F_{1,20}=5.89, P=0.025$	1.84 ± 0.79 kHz	1.96 ± 0.45 kHz	$F_{1,20}=0.17, P=0.67$
interval	$19.63\pm8.51\%$	$21.65\pm8.14\%$	$F_{1,20}=0.31, P=0.58$	159 ± 27 ms	172 ± 28 ms	$F_{1,20}=1.30, P=0.27$

documented for Richardson's ground squirrels *Urocitellus richardsonii* (Enright et al. 2020).

Our data on alarm call duration and parameters of the fundamental and peak frequencies in Brandt's voles (Table 1) coincide well with published data on another wild population of Brandt's voles (Nikolskii and Suchanova 1992; Rutovskaya 2012). In our study, we confirmed that fpeak of alarm calls of Brandt's voles is very high, on average, 10.17–11.57 kHz (Rutovskaya 2012) and 10.87 kHz (this study) and even higher than the fpeak of discomfort calls of Brandt's voles (8.33 kHz, Dymskaya et al. 2022).

Our results are consistent with previous reports of vocal individuality of alarm calls in the Brandt's vole (Rutovskaya 2012). In our study, the value of the correct assignment of alarm calls to 50 individuals with DFA was 65.52% (15 times higher than the value expected by chance) and did not decrease after cross-validation. Parameters primarily contributing to the discrimination of individuals by their alarm calls were the values of f0end and call duration (Table 4). Similar results were obtained by Rutovskaya (2012) for the alarm calls of Brandt's voles recorded in another wild population. However, in contrast to our data, where f0end was the most important acoustic parameter for discriminating individuals, the most important parameters in the study by Rutovskaya (2012) were f0max and the difference between f0max and f0min.

The difference in the results between our study and those of Rutovskaya (2012) may be a product of the differences in the analytic approaches. In our study, callers were individually identified and all calls included in the analysis belonged to known individuals. In the study by Rutovskaya (2012), the animals were recorded without individual identification, and the recorded calls were attributed to different individuals based on similarity/dissimilarity of their acoustic structure, that is, blind to the actual identity of the callers. However, our study showed that one caller can change the acoustic structure of its alarm calls even within a series of 50 successive calls (Fig. 3). So, our study suggests that calls cannot be attributed to different individuals based solely on differences in their acoustic structure, because pronounced variation was evident in calls with different contour shapes produced by the same individuals (Table 3).

The high individuality of alarm calls in Brandt's voles was expected based on previous reports of the individual-specific nature of alarm calls produced by different species of burrowing rodents and lagomorphs (Conner 1985; Hare and Atkins 2001; McCowan and Hooper 2002; Blumstein and Munos 2005; Matrosova et al. 2009, 2010a, 2010b, 2011; Pollard and Blumstein 2011; Schneiderová et al. 2017; Loughry et al. 2019; Volodin et al. 2018, 2021a; Goncharov et al. 2021). While individualistic alarm calls were previously reported for many other species of rodents, the impact of individual variation for each acoustic variable of each caller (Table 5) and for callers with high versus low individuality of alarm calls (Table 6) is novel to this study. In spite of their strong individual distinctiveness, the alarm calls of particular Brandt's vole callers were non-uniform in their acoustic structure within a series of 50 successive alarm calls; the primary differences were in call contour shapes. However, we did not find a relationship between the degree of individualization (calculated with DFA) and the index of qualitative variation of the contours in particular Brandt's vole callers (Table 3). We did, however, find that variation of acoustic variables expressed via a coefficient of variation (CV) displayed a relationship with individuality (Tables 5 and 6).

The unpredictable occurrence of different call contours within repetitive alarm calls of Brandt's voles might make them less monotonous, in agreement with the unpredictability hypothesis suggesting the adaptive value of vocal uncertainty in preventing habituation and supporting alertness to antipredator vocalizations (Blumstein and Récapet 2009; Townsend and Manser 2011). Unpredictability may act as a potential adaptation against habituation to maintain conspecific recipients in an alert state during the monotonous emissions of antipredator vocalizations (Fitch et al. 2002). Playbacks of modified alarm calls enriched with nonlinear phenomena to yellow-bellied marmots *Marmota flaviventer* and to meerkats *Suricata suricatta* showed that vocal unpredictability prevents habituation of conspecific recipients (Blumstein and Récapet 2009; Townsend and Manser 2011; Karp et al. 2014). Marmots and meerkats displayed more vigilance in response to calls with modified acoustic structure than after control calls (Blumstein and Récapet 2009; Townsend and Manser 2011; Karp et al. 2014).

The adaptive value of individually distinct Brandt's vole alarm calls is presently unclear. In all likelihood, individual distinctiveness is a by-product of differences in individual vocal morphology, but theoretically, it can be used for different communicative purposes, which can be proposed based on available data on alarm calling of other colonial species of rodents and lagomorphs. Potential adaptive benefits from using the individualistic alarm calls by the voles are distinguishing reliable from unreliable callers (Hare and Atkins 2001; Blumstein et al. 2004; Sloan and Hare 2006);

estimating the degree of threat urgency based on the number of simultaneously vocalizing callers (Weary and Kramer 1995; Sloan and Hare 2008); estimating the direction of the movements of a predator over the colony and modifying the anti-predator behaviour accordingly (Thompson and Hare 2010); and/or discrimination between members of one's own family and intruders (Conner 1984). These potential adaptive benefits have yet to be tested for Brandt's voles in either the field or in semi-captive conditions.

Our estimations of Brandt's vole vocal tract lengths by the acoustic parameters also provide some approximation regarding formant frequencies in this species' alarm calls. Our observations show that Brandt's voles produce their alarm calls through a widely open mouth (Supplementary information Movie S1). In rodents, the larynx position is immediately behind the skull at the level of the 1st–2nd neck vertebrae (Fitch 2000; Pasch et al. 2017; Riede et al. 2017). So, based on the available measurements of skulls (Allen 1940) and heads of live Brandt's voles (Dymskaya et al. 2022), we could estimate the oral vocal tract length for adults of this species as about 26–27 mm long. For a uniform tube with one end closed, formant frequencies are described by the formula:

$$F_i = (2i - 1) \times c/4VTL$$

where i is the formant number, c is the speed of sound (350 m/s), VTL is vocal tract length (in m) and F_i is the frequency (in Hz) of ith formant (Riede and Fitch 1999). Based on this formula, the first formant of the orally produced alarm calls of Brandt's voles should have the central frequency of 3.24–3.37 kHz, and the second formant should have the central frequency of 9.72–10.01 kHz. The calculated central frequency of the second formant is very close to the average value of fpeak (10.87 kHz) in the Brandt's vole, measured in this study.

We propose that the shift of essential energy of alarm calls toward higher frequencies in Brandt's voles, supported by resonances of the second formant, could evolve for better propagation of alarm calls in open landscapes, characteristic of Brandt's vole habitat. Brandt's voles produce their anti-predator vocalizations near ground level, often from their burrow entrances (Supplementary information Movie S1). Thus, their alarm calls are often in shadow zones arising from temperature and/or wind gradients and are affected by ground attenuation (Wiley and Richards 1978). In the open habitats of Brandt's voles, ground attenuation is higher for calls lower than 2 kHz and lower for calls higher than 6 kHz (Marten and Marler 1977). So, the frequency range of alarm calls of Brandt's voles is well suited for propagation through the environment in their steppe habitats (Nikolskii and Sukhanova 1992; Rutovskaya 2012; this study). Consistent with this interpretation, other vole species inhabiting open

landscapes produce alarm calls having frequency ranges similar to those of Brandt's voles (Pandourski 2011; Rutovskaya and Nikolskii 2014; Volodin et al. 2024b). Although hearing sensitivity has yet to be studied specifically for the Brandt's vole, another vole species of open landscapes, the prairie vole *Microtus ochrogaster* has a hearing range with the greatest sensitivity between 8 and 16 kHz (New et al. 2024).

While different contour shapes contribute substantially to complicating the acoustic structure of individualistic alarm calls in Brandt's voles, nonlinear phenomena were rare in the alarm calls of this species. The rarity of nonlinear vocal phenomena was also previously reported in discomfort-related calls of Brandt's voles in captivity (Dymskaya et al. 2022). Nonlinear phenomena (biphonations and subharmonics) detected in some alarm calls of Brandt's voles in this study were very similar in the acoustic structure to the nonlinear phenomena found previously in the alarm calls of Altai pikas *Ochotona alpina*, a species which also displays a rapid increase in f0 at call onset and rapid fall of f0 after the f0 maximum, although pika alarm calls are longer (Volodin et al. 2018). In contrast to Brandt's voles, in the alarm calls of Altai pikas, nonlinear phenomena are much more abundant and constitute an important contributor to individual variation of alarm calls (Volodin et al. 2018).

Nonlinear phenomena increase the complexity of the acoustic structure of animal calls, contributing to their individuality and allowing reliable discrimination of individuals by their calls (Fitch et al. 2002; Volodina et al. 2006). Also, in many species of mammals, including people, the inclusion of nonlinear phenomena can enhance the perception of alertness and convey distress in vocalizations (Blumstein and Récapet 2009; Townsend and Manser 2011; Karp et al. 2014; Marx et al. 2021; Massenet et al. 2022). However, nonlinear phenomena did not constitute a substantial part of the acoustic variation in the alarm calls of Brandt's voles. The rarity of nonlinear phenomena in alarm calls was also reported for three species of ground squirrels (Matrosova et al. 2012) and in the Harting's vole *Microtus hartingi* (Volodin et al. 2024b). It is possible that some species of rodents avoid producing nonlinear phenomena in their alarm calls for optimization of their intensity and better propagation through the environment via concentration of energy of the fundamental frequency on the formant frequencies of the vocal tract, similar to primates (Joliveau et al. 2004; Koda et al. 2012).

We could not estimate the effects of sex and age on the structure of alarm calls in Brandt's voles. However, some species of rodents produce alarm calls that are indistinguishable between adults and pups, in spite of the large differences in body size (Matrosova et al. 2007; Swan and Hare 2008; Volodina et al. 2010). Furthermore, the influence of sex and age on the structure of alarm calls in rodents is much weaker than the influence of individuality (Blumstein and Munos 2005; Matrosova et al. 2011; Goncharov et al. 2021).

We consistently recorded alarm calls of Brandt's voles in the same context, towards a standing immobile researcher. Recording alarm calls in the presence of a human is a widespread method for studying alarm communication in ground-dwelling mammals (McCowan and Hooper 2002; Matrosova et al. 2009, 2010a, 2010b, 2011; Schneiderová and Policht 2010, 2012; Volodina et al. 2010; Schneiderová 2012; Schneiderová et al. 2017, 2020; Volodin et al. 2018, 2021a, 2024a; Goncharov et al. 2021). In some species of ground squirrels, alarm calls differ according to the type of predator (terrestrial or aerial) (Melchior 1971; Owings and Virginia 1978; Davis 1984; Leger et al. 1984; Goncharov et al. 2021), whereas in other species of rodents, such differences were not found (Matrosova et al. 2012; Schneiderová and Policht 2012; Loughry et al. 2019). To our knowledge, there is no study on voles in which differences in the alarm calls toward different types of predators were found.

Recordings of each individual vole in a single session in our study may promote overestimation of the individuality of calls, though this approach is widely applied in studying individuality in alarm calls of ground-dwelling mammals because it imitates one episode of predator appearance before the caller (McCowan and Hooper 2002; Matrosova et al. 2009, 2011; Schneiderová and Policht 2010; Volodina et al. 2010; Volodin et al. 2018, 2021a). However, the large sample size (50 calls per individual) and high variability of acoustic structure even among calls following each other in series argue against the overestimation of individuality in the Brandt's vole. Furthermore, our study provides the evidence that successive alarm calls in a monotonous series can be substantially dissimilar and investigates the degrees of similarity between them. It remains unclear, however, how variable alarm calls of Brandt's vole individuals are over different time spans, as studies paralleling those performed with ground squirrels (Matrosova et al. 2009, 2010a, 2010b; Schneiderová et al. 2017) have yet to be conducted.

Declarations

Conflict of interest The authors declare no competing interests.

References

Allen GM (1940) The mammals of China and Mongolia. Natural history of Central Asia, v. 11. part 2. New York, American Museum of Natural History. https://doi.org/10.5962/bhl.title.12195

Bai D, Wan X, Li G, Wan X, Guo Y, Shi D, Zhang Z (2022) Factors influencing range contraction of a rodent herbivore in a steppe grassland over the past decades. Ecol Evol 12:e8546. https://doi.org/10.1002/ece3.8546

Batsuren E, Zhang X, Song M, Wan X, Li G, Liu J, Huang S, Zhang Z (2022) Density-dependent changes of mating system and family structure in Brandt's voles (*Lasiopodomys brandtii*). Ecol Evol 12:e9199. https://doi.org/10.1002/ece3.9199

Blumstein DT (2007) The evolution, function, and meaning of marmot alarm communication. Adv Study Behav 37:371–401. https://doi.org/10.1016/S0065-3454(07)37008-3

Blumstein DT, Munos O (2005) Individual, age and sex-specific information is contained in yellow-bellied marmot alarm calls. Anim Behav 69:353–361. https://doi.org/10.1016/j.anbehav.2004.10.001

Blumstein DT, Récapet C (2009) The sound of arousal: the addition of novel non-linearities increases responsiveness in marmot alarm calls. Ethology 115:1074–1081. https://doi.org/10.1111/j.1439-0310.2009.01691.x

Blumstein DT, Verneyre L, Daniel JC (2004) Reliability and the adaptive utility of discrimination among alarm callers. Proc R Soc B 271:1851–1857. https://doi.org/10.1098/rspb.2004.2808

Briefer E, Vannoni E, McElligott AG (2010) Quality prevails over identity in the sexually selected vocalisations of an ageing mammal. BMC Biol 8:35. https://doi.org/10.1186/1741-7007-8-35

Chelysheva EV, Klenova AV, Volodin IA, Volodina EV (2023) Advertising sex and individual identity by long-distance chirps in wild-living mature cheetahs (*Acinonyx jubatus*). Ethology 129:288–300. https://doi.org/10.1111/eth.13366

Chrdileli M, Borchia D (2024) Index of Qualitative Variation Calculator. Available at: https://www.omnicalculator.com/statistics/index-of-qualitative-variation. Accessed 17 Dec 2024

Conner DA (1984) The role of an acoustic display in territorial maintenance in the pika. Can J Zool 62:1906–1909. https://doi.org/10.1139/z84-280

Conner DA (1985) The function of the pika short call in individual recognition. Z Tierpsychol 67:131–143. https://doi.org/10.1111/j.1439-0310.1985.tb01382.x

Cui C, Xie Y, Hua Y, Yang S, Yin B, Wei W (2020) Brandt's vole (*Lasiopodomys brandtii*) affects its habitat quality by altering plant community composition. Biologia 75:1097–1104. https://doi.org/10.2478/s11756-020-00469-z

Da Silva KB, Kramer DL, Weary DM (1994) Context-specific alarm calls of the eastern chipmunk, *Tamias striatus*. Can J Zool 72:1087–1092. https://doi.org/10.1139/z94-146

Davis LS (1984) Alarm calling in Richardson's ground squirrels (*Spermophilus richardsonii*). Z Tierpsychol 66:152–164. https://doi.org/10.1111/j.1439-0310.1984.tb01362.x

Dymskaya MM, Volodin IA, Smorkatcheva AV, Vasilieva NA, Volodina EV (2022) Audible, but not ultrasonic, calls reflect surface-dwelling or subterranean specialization in pup and adult Brandt's and mandarin voles. Behav Ecol Sociobiol 76:106. https://doi.org/10.1007/s00265-022-03213-6

Dymskaya MM, Volodin IA, Smorkatcheva AV, Rudyk A, Volodina EV (2024) Field tests reveal acoustic variation of call types in a subterranean rodent, the Northern Mole Vole *Ellobius talpinus*. J Mammal gyae123. https://doi.org/10.1093/jmammal/gyae123

Eddington VM, Nichols HK, Calistri-Yeh A, Hilliard Young VK, Kloepper LN (2024) Graded alarm call behavior in wild fox squirrels (*Sciurus niger*). J Acoust Soc Am 155:1308–1314. https://doi.org/10.1121/10.0024771

Enkhbold N, Dazhao S, Batsaikhan N, Wang D, Tseveendorj D, Erdenetuya B (2014) Study on the distribution and habitat characteristics of Brandt's vole. Mongol J Agricul Sci 13:88–95. https://doi.org/10.5564/mjas.v13i2.524

Enright DT, Leboe-McGowan LC, Hare JF (2020) Richardson's ground squirrel (*Urocitellus richardsonii*) alarm call receivers experience an auditory continuity illusion. Can J Exp Psychol 74:193–200. https://doi.org/10.1037/cep0000215

Fitch WT (2000) Skull dimensions in relation to body size in nonhumans mammals: the causal bases for acoustic allometry. Zoology - Anal Complex Syst 103:40–58

Fitch WT, Neubauer J, Herzel H (2002) Calls out of chaos: the adaptive significance of nonlinear phenomena in mammalian vocal production. Anim Behav 63:407–418. https://doi.org/10.1006/anbe.2001.1912

Goncharov D, Policht R, Hambálková L, Salovarov V, Hart V (2021) Individual-based acoustic variation of the alarm calls in the long-tailed ground squirrel. R Soc Open Sci 8:200147. https://doi.org/10.1098/rsos.200147

Gromov VS (2023) Ecology and social behavior of the Brandt's vole: a generalized review. J Arid Environ 214:104994. https://doi.org/10.1016/j.jaridenv.2023.104994

Guo Y, Li Z, Dong S, Si X, Ta N, Liang H, Xu L (2023) Multiple infections of zoonotic pathogens in wild Brandt's voles (*Lasiopodomys brandtii*). Vet Med Sci 9:2201–2211. https://doi.org/10.1002/vms3.1214

Hare JF (1998) Juvenile Richardson's ground squirrels, *Spermophilus richardsonii*, discriminate among individual alarm callers. Anim Behav 55:451–460. https://doi.org/10.1006/anbe.1997.0613

Hare JF, Atkins BA (2001) The squirrel that cried wolf: reliability detection by juvenile Richardson's ground squirrels (*Spermophilus richardsonii*). Behav Ecol Sociobiol 51:108–112. https://doi.org/10.1007/s002650100414

Joliveau E, Smith J, Wolfe J (2004) Tuning of vocal tract resonance by sopranos. Nature 427:116–116. https://doi.org/10.1038/427116a

Karp D, Manser MB, Wiley EM, Townsend SW (2014) Nonlinearities in meerkat alarm calls prevent receivers from habituating. Ethology 120:189–196. https://doi.org/10.1111/eth.12195

Kirilyuk O, Kirilyuk V, Maksakovsky N, Kobyakova S, Goroshko O, Tkachuk T, Butorin A, Namkhai A, Urtnasan N, Tseveenmyadag N, Oyungerel B, Dashdorj K, Knapp HD, Golde A (2013) Landscapes of Dauria. ANNIE, Moscow

Koda H, Nishimura T, Tokuda IT, Oyakawa C, Nihonmatsu T, Masataka N (2012) Soprano singing in gibbons. Am J Phys Anthropol 149:347–355. https://doi.org/10.1002/ajpa.22124

Leger DW, Berney-Key SD, Sherman PW (1984) Vocalizations of Belding's ground squirrels (*Spermophilus beldingi*). Anim Behav 32:753–764. https://doi.org/10.1016/S0003-3472(84)80151-7

Li K, Kohn MH, Zhang S, Wan X, Shi D, Wang D (2017) The colonization and divergence patterns of Brandt's vole (*Lasiopodomys brandtii*) populations reveal evidence of genetic surfing. BMC Evol Biol 17:145. https://doi.org/10.1186/s12862-017-0995-y

Loughry WJ, Oeser M, Anderson CD, Hoogland JL (2019) The importance of individual variation in the alarm calls of Gunnison's prairie dogs. Anim Behav 150:59–68. https://doi.org/10.1016/j.anbehav.2019.01.019

Marten K, Marler P (1977) Sound transmission and its significance for animal vocalization. I Temperate Habitats Behav Ecol Sociobiol 2:271–290. https://doi.org/10.1007/BF00299740

Marx A, Lenkei R, Pérez Fraga P, Bakos V, Kubinyi E, Faragó T (2021) Occurrences of non-linear phenomena and vocal harshness in dog whines as indicators of stress and ageing. Sci Rep 11:4468. https://doi.org/10.1038/s41598-021-83614-1

Massenet M, Anikin A, Pisanski K, Reynaud K, Mathevon N, Reby D (2022) Nonlinear vocal phenomena affect human perceptions of distress, size and dominance in puppy whines. Proc R Soc B 289:20220429. https://doi.org/10.1098/rspb.2022.0429

Matrosova VA, Volodin IA, Volodina EV, Babitsky AF (2007) Pups crying bass: vocal adaptation for avoidance of age-dependent predation risk in ground squirrels? Behav Ecol Sociobiol 62:181–191. https://doi.org/10.1007/s00265-007-0452-9

Matrosova VA, Volodin IA, Volodina EV (2009) The short-term and long-term individuality in speckled ground squirrel alarm calls. J Mammal 90:158–166. https://doi.org/10.1644/08-MAMM-A-032.1

Matrosova VA, Volodin IA, Volodina EV, Vasilieva NA, Kochetkova AA (2010a) Between-year stability of individual alarm calls in the yellow ground squirrel Spermophilus fulvus. J Mammal 91:620–627. https://doi.org/10.1644/09-MAMM-A-143.1

Matrosova VA, Volodin IA, Volodina EV, Vasilieva NA (2010b) Stability of acoustic individuality in the alarm calls of wild yellow ground squirrels Spermophilus fulvus and contrasting calls from trapped and free-ranging callers. Naturwissenschaften 97:707–715. https://doi.org/10.1007/s00114-010-0686-7

Matrosova VA, Blumstein DT, Volodin IA, Volodina EV (2011) The potential to encode sex, age, and individual identity in the alarm calls of three species of Marmotinae. Naturwissenschaften 98:181–192. https://doi.org/10.1007/s00114-010-0757-9

Matrosova VA, Schneiderová I, Volodin IA, Volodina EV (2012) Species-specific and shared features in vocal repertoires of three Eurasian ground squirrels (genus Spermophilus). Acta Theriol 57:65–78. https://doi.org/10.1007/s13364-011-0046-9

McCowan B, Hooper SL (2002) Individual acoustic variation in Belding's ground squirrel alarm chirps in the High Sierra Nevada. J Acoust Soc Am 111:1157–1160. https://doi.org/10.1121/1.1446048

Melchior HR (1971) Characteristics of arctic ground squirrel alarm calls. Oecologia 7:184–190. https://doi.org/10.1007/BF00346360

Mundry R, Sommer C (2007) Discriminant function analysis with non-independent data: consequences and an alternative. Anim Behav 74:965–976. https://doi.org/10.1016/j.anbehav.2006.12.028

New EM, Hurd JA, Alarcon GA, Miller CS, Williams PA, Greene NT, Sergott CE, Li BZ, Lei TC, McCullagh EA (2024) Hearing ability of prairie voles (Microtus ochrogaster). J Acoust Soc Am 155:555–567. https://doi.org/10.1121/10.0024357

Nikolskii AA, Sukhanova MV (1992) Situation dependent variations of a call emitted by great gerbil, Rhombomys opimus and by Brandt's vole, Microtus brandtii, retreating to burrows. Zool Zh 71(12):125–132 [in Russian]

Nikonova VR, Naumova AE, Bergaliev AM, Dymskaya MM, Rudyk AI, Volodina EV, Smorkatcheva AV (2024) Dental radiography as a low-invasive field technique to estimate age in small rodents, with the mole voles (Ellobius) as an example. Eur J Wildlife Res 70:46. https://doi.org/10.1007/s10344-024-01802-6

Obyazov VA, Kirilyuk VE, Kirilyuk AV (2021) Torey lakes as an indicator of moisture long-term changes in Southeastern Transbaikalia and Northeastern Mongolia. Hydrosphere 3:204–232 [in Russian]. https://doi.org/10.34753/HS.2021.3.3.204

Owings DH, Virginia RA (1978) Alarm calls of California ground squirrels (Spermophilus beecheyi). Z Tierpsychol 46:58–70. https://doi.org/10.1111/j.1439-0310.1978.tb01438.x

Pandourski I (2011) Case of alarm vocalization in a colony of Microtus guentheri (Danford & Alston, 1880) (Mammalia, Rodentia, Arvicolidae) from Southern Bulgaria. ZooNotes 20:1–3

Pasch B, Tokuda IT, Riede T (2017) Grasshopper mice employ distinct vocal production mechanisms in different social contexts. Proc R Soc B 284:20171158. https://doi.org/10.1098/rspb.2017.1158

Pollard KA (2011) Making the most of alarm signals: the adaptive value of individual discrimination in an alarm context. Behav Ecol 22:93–100. https://doi.org/10.1093/beheco/arq179

Pollard KA, Blumstein DT (2011) Social group size predicts the evolution of individuality. Curr Biol 21:413–417. https://doi.org/10.1016/j.cub.2011.01.051

R Core Team (2022) R: a language and environment for statistical computing. Vienna, R Foundation for Statistical Computing. https://www.R-project.org/. Accessed 17 Dec 2024

Riede T, Fitch T (1999) Vocal tract length and acoustics of vocalization in the domestic dog (Canis familiaris). J Exp Biol 202:2859–2867. https://doi.org/10.1242/jeb.202.20.2859

Riede T, Borgard HL, Pasch B (2017) Laryngeal airway reconstruction indicates that rodent ultrasonic vocalizations are produced by an edge-tone mechanism. R Soc Open Sci 4:170976. https://doi.org/10.1098/rsos.170976

Rutovskaya MV, Volodin IA, Feoktistova NY, Surov AV, Gureeva AV, Volodina EV (2024) Acoustic complexity of pup isolation calls in Mongolian hamsters: 3-frequency phenomena and chaos. Curr Zool 70:559–574. https://doi.org/10.1093/cz/zoad036

Rutovskaya MV, Nikolskii AA (2014) The sound signals of the narrow-skulled vole (Microtus gregalis Pall.). Sens Systems 28:76–83 [In Russian].

Rutovskaya MV (2012) The sound signals of Brandt's vole (Lasiopodomys brandti). Sens Systems 26:31–38 [In Russian]

Samjaa R, Zophel U, Peterson J (2000) The impact of the vole Microtus brandti on Mongolian steppe ecosystems. Marburger Geographische Schriften 135:346–360

Schibler F, Manser MB (2007) The irrelevance of individual discrimination in meerkat alarm calls. Anim Behav 74:1259–1268. https://doi.org/10.1016/j.anbehav.2007.02.026

Schneiderová I (2012) Frequency-modulated second elements of two-element alarm calls do not enhance discrimination of callers in three Eurasian ground squirrels. Curr Zool 58:749–757. https://doi.org/10.1093/czoolo/58.5.749

Schneiderová I, Policht R (2010) Alarm calls of the European ground squirrel Spermophilus citellus and the Taurus ground squirrel S. taurensis encode information about caller identity. Bioacoustics 20:29–43. https://doi.org/10.1080/09524622.2011.9753631

Schneiderová I, Policht R (2012) Acoustic analysis of the alarm call of the Anatolian ground squirrel Spermophilus xanthoprymnus: a description and comparison with alarm calls of the Taurus S. taurensis and European S. citellus ground squirrels. Naturwissenschaften 99:55–64. https://doi.org/10.1007/s00114-011-0870-4

Schneiderová I, Volodina EV, Matrosova VA, Volodin IA (2017) One plus one: binary alarm calls retain individual signature for longer periods than single-note alarms in the European ground squirrel (Spermophilus citellus). Behav Process 138:73–81. https://doi.org/10.1016/j.beproc.2017.02.014

Schneiderová I, Štefanská L, Kratochvíl L (2020) Geographic variability in the alarm calls of the European ground squirrel. Curr Zool 66:407–415. https://doi.org/10.1093/cz/zoz055

Sedova LM, Volodin IA, Erofeeva MN, Alekseeva GS, Naidenko SV, Volodina EV (2023) Meows of captive feral domestic cats (Felis silvestris catus) in the mating season: acoustic correlates of caller identity and sex. Behaviour 160:869–888. https://doi.org/10.1163/1568539X-bja10239

Sloan JL, Hare JF (2006) Adult Richardson's ground squirrels (Spermophilus richardsonii) ignore rate changes in juvenile alarm calls:

age-differential response urgency perception? Ethology 112:896–902. https://doi.org/10.1111/j.1439-0310.2006.01235.x

Sloan JL, Hare JF (2008) The more the scarier: adult Richardson's ground squirrels (*Spermophilus richardsonii*) assess response urgency via the number of alarm signalers. Ethology 114:436–443. https://doi.org/10.1111/j.1439-0310.2006.01235.x

Solow AR (1990) A randomization test for misclassification probability in discriminant analysis. Ecology 71:2379–2382. https://doi.org/10.2307/1938650

Swan DC, Hare JF (2008) Signaler and receiver ages do not affect responses to Richardson's ground squirrel alarm calls. J Mammal 89:889–894. https://doi.org/10.1644/07-MAMM-A-228.1

Thompson AB, Hare JF (2010) Neighbourhood watch: multiple alarm callers communicate directional predator movement in Richardson's ground squirrels, *Spermophilus richardsonii*. Anim Behav 80:269–275. https://doi.org/10.1016/j.anbehav.2010.04.028

Townsend SW, Manser MB (2011) The function of nonlinear phenomena in meerkat alarm calls. Biol Lett 7:47–49. https://doi.org/10.1098/rsbl.2010.0537

Volodin IA, Matrosova VA, Frey R, Kozhevnikova JD, Isaeva IL, Volodina EV (2018) Altai pika (*Ochotona alpina*) alarm calls: individual acoustic variation and the phenomenon of call-synchronous ear folding behavior. Sci Nat 105:40. https://doi.org/10.1007/s00114-018-1567-8

Volodin IA, Volodina EV, Frey R, Karaseva KD, Kirilyuk VE (2021a) Daurian pika (*Ochotona dauurica*) alarm calls: individual acoustic variation in a lagomorph with audible through ultrasonic vocalizations. J Mammal 102:947–959. https://doi.org/10.1093/jmammal/gyab048

Volodin IA, Yurlova DD, Ilchenko OG, Volodina EV (2021b) Ontogeny of audible squeaks in yellow steppe lemming *Eolagurus luteus*: trend towards shorter and low-frequency calls is reminiscent of those in ultrasonic vocalization. BMC Zoology 6:27. https://doi.org/10.1186/s40850-021-00092-8

Volodin IA, Klenova AV, Kirilyuk VE, Ilchenko OG, Volodina EV (2024a) Ultrasonic alarm call of Mongolian gerbils (*Meriones ungiuculatus*) in the wild and in captivity: a potential tool for detecting inhabited colonies during population depression. Mamm Biol 104:407–416. https://doi.org/10.1007/s42991-024-00416-4

Volodin IA, Rutovskaya MV, Golenishchev FN, Volodina EV (2024b) Startle together, shout in chorus: collective bursts of alarm calls in a social rodent, the Harting's vole (*Microtus hartingi*). Behaviour 161:587–611. https://doi.org/10.1163/1568539X-bja10274

Volodina EV, Volodin IA, Isaeva IV, Unck C (2006) Biphonation may function to enhance individual recognition in the dhole, *Cuon alpinus*. Ethology 112:815–825. https://doi.org/10.1111/j.1439-0310.2006.01231.x

Volodina EV, Matrosova VA, Volodin IA (2010) An unusual effect of maturation on the alarm call fundamental frequency in two species of ground squirrels. Bioacoustics 20:87–98. https://doi.org/10.1080/09524622.2011.9753634

Weary DM, Kramer DL (1995) Response of eastern chipmunks to conspecific alarm calls. Anim Behav 49:81–93. https://doi.org/10.1016/0003-3472(95)80156-1

Wilcox AR (1973) Indices of qualitative variation and political measurement. The Western Political Quarterly 26:325–343. https://doi.org/10.1177/106591297302600209

Wilden I, Herzel H, Peters G, Tembrock G (1998) Subharmonics, biphonation, and deterministic chaos in mammal vocalization. Bioacoustics 9:171–196. https://doi.org/10.1080/09524622.1998.9753394

Wiley RH, Richards DG (1978) Physical constraints on acoustic communication in the atmosphere: implications for the evolution of animal vocalizations. Behav Ecol Sociobiol 3:69–94. https://doi.org/10.1007/BF00300047

Wilson DR, Hare JF (2003) Richardson's ground squirrels (*Spermophilus richardsonii*) do not communicate predator movements via changes in call rate. Can J Zool 81:2026–2031. https://doi.org/10.1139/z03-225

Wilson-Henjum GE, Job JR, McKenna MF, Shannon G, Wittemyer G (2019) Alarm call modification by prairie dogs in the presence of juveniles. J Ethol 37:167–174. https://doi.org/10.1007/s10164-018-0582-8

Xu L, Schmid BV, Liu J, Si XY, Stenseth NC, Zhang Z (2015) The trophic responses of two different rodent-vector-plague systems to climate change. Proc R Soc B 282:20141846. https://doi.org/10.1098/rspb.2014.1846

Zahler P, Dolgormaa L, Hinds LA, Smith AT (2004) The management of Brandt's vole in Mongolia: toward an ecologically based means of control. Mongolian J Biol Sci 2:51–55. https://doi.org/10.22353/mjbs.2004.02.17

Zhong W, Wang G, Zhou Q, Wang G (2007) Communal food caches and social groups of Brandt's voles in the typical steppes of Inner Mongolia, China. J Arid Environ 68:398–407. https://doi.org/10.1016/j.jaridenv.2006.06.008