### Chapter 4

The effects of anthropogenic noise on pygmy marmoset calls on a gradient of exposure

### 4.0 Abstract

The environment is a known pressure driving change in vocal communication structure but as anthropogenic noise infiltrates wild areas it is important to understand how this novel stress further impacts vocalisations. Gradients of exposure are a key knowledge gap in our understanding of the impacts of anthropogenic noise on wildlife. This chapter investigates the association between exposure to a gradient of anthropogenic noise and changes in pygmy marmoset (*Cebuella niveiventris*) calls. I placed an autonomous sound recorder (Audiomoth) near the feeding trees of twenty-three pygmy marmoset groups in the Área de Conservación Regional Comunal Tamshiyacu Tahuayo reserve in north-eastern Peru. The Audiomoths were left up for 2 days and 12 hours (5:30-17:30) of recordings for each day were analysed for anthropogenic noise and four pygmy marmoset calls. I found significant differences in both spectral and temporal call characteristics on an anthropogenic noise gradient for three of the four calls. Filling the knowledge gap of changes in vocalisation characteristics on a gradient of exposure is essential as it reveals if animals are able to adapt to varying levels of exposure.

### 4.1 Introduction

Human generated noise, henceforth referred to as anthropogenic noise, is causing lasting change to the environment and is now recognised as a major pollutant (Radford *et al.*, 2014). Noise-generating human activities differ from biotic and abiotic sounds in their acoustic characteristics such as cycle, impulsiveness, rise time, and constancy (Popper and Hastings, 2009; Radford *et al.*, 2014). Therefore, anthropogenic noise presents a genuine and, in some cases, novel challenge to animals in their environment.

Communication is the means of sending information via a signal produced by a transmitter which is broadcasted through an environment and received and processed by a receptor (Simmons, 2003; Gomes *et al.*, 2022). These communication signals include information about the identity, sexual state and location of the one vocalising (Gerhardt and Huber, 2003). Signal degradation caused by the environments acts as a selecting pressure shaping how animals communicate (Hardt and Benedict, 2021). Closed habitats (like dense rainforests) are thought to

inflict stronger selection pressures than open ones because in these environments the auditory and visual communication channels cannot complement one another (Ey and Fischer, 2009). The acoustic adaptation hypothesis postulates that acoustic signals should transmit better in the habitat in which they evolved (Morton, 1975). This hypothesis theorises that vocalisations are more stereotyped in closed verses open habitats to allow for an increase in detection rates. Animals are expected to increase the rate of their vocalisations in closed versus open habitats, this also helps with the limited use of visual communication possible in denser vegetation (Ey and Fischer, 2009). However, a key gap in our current understanding is how this selection pressure factors in novel changes in environments with anthropogenic noise becoming ever more prevalent.

It is known that changes in sound levels can affect an animal's behaviour in three major ways: by masking acoustic cues or signals (Radford et al., 2014), as a distracting stimulus (Chan and Blumstein, 2011), and as a stressor (Knight and Swaddle, 2011). If the listener is unable to detect a signal then masking is complete, if they can still detect the signal but cannot understand its content, there is partial masking (Clark et al, 2009; Radford et al., 2014). Anthropogenic masking affects a wide variety of vertebrates (primates, birds, fish, amphibians, and marine mammals) with an increase in background noise making communication more challenging for those vertebrates (Radford et al., 2014). Which indicates that not only is vegetation and other elements of the environment negatively impacting communication propagation so too is anthropogenic noise. Anthropogenic noise also distracts prey species, the Caribbean hermit crab (*Coenobita clypeatus*) have a reduced ability to make thorough predation risk assessments when subjected to loud motor boat sounds (Chan et al., 2010). This reduction in an individual's ability to make risk assessments can lead to a decrease in fitness, especially if the signals that are being interpreted are ones that indicate predation threats (Lowry et al., 2012; Radford et al., 2014). Noise pollution can also serve as a stressor for example by impacting reproductive success and disrupting signals between offspring and parents (Leonard and Horn, 2012) as well as reducing the ability to correctly assess the quality of potential mates (Halfwerk *et al.*, 2011).

Animals mitigate some of these negative impacts of anthropogenic noise on communication by adjusting the vocal structure of their calls (Hu and Cardoso, 2010). These vocal adjustments include increasing the frequency of their call (Nemeth and Brumm, 2009), increasing the

amplitude (Hage et al., 2013), call rate (Sun and Narins, 2005), shifting the time of day for their singing (Fuller et al., 2007), number of notes (Slabbekoorn and Boer-Visser, 2006) and increasing the duration (Brumm et al., 2004). Most research in this field has examined how anthropogenic noise affects acoustic signalling in marine mammals and birds (Radford, et al., 2014). South Atlantic right whale (Eubalaena australis) calls have higher peak frequencies and call at lower rates when in the presence of an increase in low-frequency anthropogenic noise, these changes are occurring during an individual's lifetime (Parks et al., 2007). Great tits (Parus *major*) sing with a higher minimum frequency at noisier locations to prevent masking, demonstrating a behavioural plasticity driven by urbanisation (Slabbekoorn and Peet, 2003). Although the literature is more limited in scope there is evidence of primates responding to abiotic changes in their environments. Cotton-top tamarins (Saguinus oedipus) increase their call amplitude and the syllable duration when exposed to an increase in background noise amplitude (Egnor and Hauser, 2006). Black tufted-ear marmosets (Callithrix penicillata) make phee vocalisations (long distance contact call) at lower low, high and dominant frequencies and the calls have longer durations in nosier environments than quiet ones (Santos et al., 2017). However, not all species show an ability to adapt to these changes in the soundscape. Duarte et al. (2018) compared the long-distance vocal communications of the black-fronted titi monkey (Callicebus nigrifrons) at a site with loud mining and one without, and found that the monkeys by the mining site had a lower rate of long communication calls.

One of the main knowledge gaps in the current literature investigating the impacts of anthropogenic noise on wildlife is the lack of studies measuring behavioural responses on a gradient of noise levels (Shannon *et al.*, 2016). Most studies focus on quiet versus loud treatments (as in Santos *et al.*, 2017) rather than on a more nuanced look at what thresholds of exposure these changes are occurring. The only study to date to incorporate the use of a gradient of noise exposure to measure the difference in wild primates' behavioural responses is Lineros *et al.* (2020), who investigated the hormonal and behavioural responses in the Bolivian grey titi monkey (*Plecturocebus donacophilus*) on an exposure gradient. They found that when the titi groups located further away from a highway were presented with a human mannequin they produced more alarm calls, which they postulate is an indication that the groups closer to the highway are becoming more habituated to human presence. However, this study did not

investigate the impacts on the structure of titi monkey calls. Gómez-Espinosa *et al.* (2022) investigated the impacts of anthropogenic noise on male mantled howler monkey (*Aloutta palliate*) behaviour and also catalogued the anthropogenic noises present that the monkeys are exposed to, with a focus on vigilance behaviours and the changes in the frequency of calling but not any structural changes that might also be occurring. In their study male howler monkeys called more frequently and displayed more vigilance behaviours in response to higher sound pressure audios, finding theses behavioural differences on the group level. The authors postulate that the frequency of exposure to these different anthropogenic sounds is the likely explanation for the group level behavioural differences found. Both studies provide evidence that primates are exhibiting behavioural changes on a gradient of exposure to humans and anthropogenic noise.

However, none of the studies mentioned considered the impacts these anthropogenic sounds have on the vocal structure of the study species, leaving a hole in the understanding of how a gradient of exposure impacts primate vocalisation. Filling the knowledge gap of changes in vocalisation characteristics on a gradient of exposure is essential as it reveals if animals are able to adapt to varying levels of exposure. It is even more pertinent to understand the potentiality negative impacts that anthropogenic noise is having on species, like primates, who rely heavily on vocal communication to survive. It also allows for further investigation into the correlation between the level and duration of anthropogenic noise exposure and potential habituation to these sounds in a wide variety of species and habitats (Shannon *et al.*, 2016).

This study aims to investigate the association between exposure to a gradient of anthropogenic noise and the changes in the characteristics of the eastern pygmy marmoset (*Cebuella niveiventris*) calls. If anthropogenic noise impacts calls, then I expect these changes would be greater for long distance calls. I investigate how this change is occurring on a gradient scale in a more remote setting rather than a strict comparison of an urban population versus a rural one as seen in Santos *et al.* (2017), providing more insight and filling the knowledge gap on where these vocal changes are potentially occurring. Considering previous literature, I predict that all the contact calls made by pygmy marmosets will be influenced by the presence of anthropogenic noise and that calls will be longer and at lower frequency with greater exposure to anthropogenic

noise. This study can be used to further our understanding of how anthropogenic noise disturbance is affecting primate behaviour allowing for a more thoughtful discussion of what mitigation measures can be taken to limit these effects, especially those driven by an increase in ecotourism.

## 4.2 Methods

#### 4.2.1 Study site

The research was conducted in Área de Conservación Regional Comunal Tamshiyacu Tahuayo a reserve located in north-eastern Peru which was established as a conservation protection area in 1991 (Newing and Bodmer, 2003). It is located in the Amazon flood basin and undergoes monomodal flooding which is denoted by a dry season during the equatorial winter and a flooded season (when the river is at its highest) in during the equatorial summer. The reserve has three zones: a permanent settlement zone, where people live; a subsistence use zone, designated for the sustainable use of natural resources; a fully protected zone, where hunting and logging are prohibited and the hunting of primates is illegal throughout all zones (Hurtado-Gonzales and Bodmer, 2004). There are several different habitat types found throughout the reserve ranging from aquatic (rivers, canals, lakes, oxbow lakes) to terrestrial including varying types of lowland and upland forests including terra firme, igapo and varzea (Bodmer, 1989).

There is only one tour operator with accommodation built inside the reserve limits, run by Amazonia Expeditions, it is also the only manmade structure found inside the reserve. Amazonia Expeditions main lodge, where most of their clients stay, is based outside of the reserve limits and close to the El Chino community (locations shown in Figure 4.2). They experience the highest volume of tourists during their peak occupancy season of July-August which falls in the dry season. Chapter 3 detailed that there was a higher variety and amount of anthropogenic noise outside of the reserve boundary. With the location that had the highest total anthropogenic noise catalogued located in El Chino behind a person's home and the group with the lowest being located inside the reserve boundary on a trail to an oxbow lake. The most common anthropogenic sound encountered across the reserve and outside of it was low motor and the least was high motor.

#### 4.2.2 Study species

The eastern pygmy marmoset, *Cebuella niveiventris*, is a Platyrrhine species found in the Amazon rainforest in Bolivia, Brazil, and Peru (de la Torre *et al.*, 2021). They are habitat specialists and are found in forests along river-edges, with small home ranges of 0.1-0.5 ha, which feature 1-6 feeding trees (Soini, 1988). Adult males weigh 110g and females weigh 122g on average (Soini, 1982; de la Torre and Rylands, 2008). They are a highly vocal arboreal species and are gum-feeding specialists, however they do also eat insects and fruits (Soini, 1982; de la Torre and Rylands, 2-9 individuals (de la Torre *et al.*, 2000), groups are comprised of a dominant breeding pair and their successive litters of offspring (de la Torre and Rylands, 2008).

### 4.2.2.1 Pygmy marmoset calls

Pygmy marmosets, much like other arboreal primates, are extremely reliant on vocal communications. Pygmy marmosets and other family-living/ pair-bonded primates (titi monkeys, tamarins, night monkeys, gibbons, other species of marmosets) have different ecological and social challenges than other primate species which is why their vocal communication is different to others (Snowdon, 2017). The vocal communication that takes place in these species serve a role in developing and sustaining the pair bond as well as helping partners reduce stress (Snowdon, 2017).

The principle frequency of pygmy marmoset calls (both *C. niveiventris* and *C. pygmaea*) is above the spectral range of most ambient noises (like insects, birds and frogs) and is even higher than the hearing range of many birds of prey, which suggests that this species is very well vocally adapted for their environment (de la Torre and Snowdon, 2002; Snowdon, 2017). The vocalisations made by both species of pygmy marmoset are similar to other marmoset species (Epple, 1968; Snowdon and Elowson, 1999). Calls have been well documented in captive colonies (Pola and Snowdon, 1975; Elowson *et al.*, 1992; Elowson and Snowdown, 1994; Snowdon and Elowson, 1999) which serve as the backbone to the field research later conducted (de la Torre and Snowdon, 2002; de la Torre and Snowdon, 2009). Two calls (j calls and trills) of wild populations in Ecuador (western pygmy marmoset, *Cebuella pygmaea*) have been studied by de la Torre and Snowdon (2009). They found consistent structural differences at the population level, with both trills and j calls showing significant variation across all the parameters studied (duration, notes/sec, maximum frequency, minimum frequency and bandwidth).

This study will be focusing on four pygmy marmoset calls. Three contact-location calls; trills, j calls and long calls, and an alarm call known as a tsik call. These calls are the focus in this study as contact calls serve as an important means of communication within pygmy marmoset groups and need to be able to travel effectively through the environment in order to deliver the information needed to the receiving individuals. The tsik call is included as alarm calls are needed in order to successfully pass information regarding predator presence to other group members.

**Trills**. Trill calls are used to provide short-range contact, when group members are up to 9 meters apart, as well as settle group interactions (de la Torre and Snowdon, 2002). There are both open mouthed and closed trills (Pola and Snowdon, 1975). Open-mouth trills are used most in conjunction with agonistic threat displays, and two types of genital presentation (Pola and Snowdon, 1975). Closed-mouth trills are used in situations of low-level arousal and when the marmosets were moving around calmly (Pola and Snowdon, 1975). In captivity, both trills are used after warning calls were emitted, and when fresh food was presented (Pola and Snowdon, 1975). They have a pulsatile temporal structure of 32-38 cycles and of a sinusoidally frequency modulated tones of 7-12 kHz (Figure 4.1a, spectrogram of the call).

**J calls**. J calls (so called because of the shape of the call on a spectrogram, Figure 4.1b) are also short-range contact calls that can also be used in group interactions, used when group members were more than 20 meters apart (de la Torre and Snowdon, 2002). They are often followed by an alarm call (Pola and Snowdon, 1975). J calls were also emitted as a distress call in a captive colony studied by Snowdon and Elowson (1999). They have a pulsating temporal structure of 17-23 cycles and a sinusoidally frequency modulated tones of 7-12 kHz (de la Torre and Snowdon, 2002).

**Long calls**. Long calls are a contact call used for long distances over 20 meters (de la Torre and Snowdon, 2002). Long calls have a frequency range of 8-11 kHz and 3 cycles (de la Torre and

Snowdon, 2002; See Figure 4.1c for a spectrogram). They are also often used by lone males in the hopes of attracting a female to join them to start a new group.

**Tsik calls.** These calls are not well studied in pygmy marmosets but have been studied in common marmoset (*Callithrix jacchus*) family groups (Epple, 1968; Bezerra and Souto, 2008; Kato *et al.*, 2014) as most pygmy marmoset call research has been focused on contact calls. These calls are used when an individual is alarmed and are also heard during aggressive encounters (Bezerra and Souto, 2008). Tsik calls in rapid succession are known as mobbing calls and they are given in response to the presence of a potential predator where members of the group join in (Bezerra and Souto, 2008). The following information is on the tsik call emitted by the common marmoset, they have a frequency range of 5-14 kHz (Epple, 1968), which is a lower frequency than those emitted by the pygmy marmosets in this study (Figure 4.1d)



Figure 4.1 Spectrograms of pygmy marmoset calls created in Raven Pro a) one trill call b) three j calls c) long call with three notes d) two tsik calls. The frequency in kHz displayed on the y axis.

### 4.2.3 Experimental procedure

One autonomous sound recorder (Audiomoth, Open Acoustics) was placed near the main feeding tree of 23 groups (see Table 4.7 in the appendix for locations and descriptions for the groups) in the dry season. Fourteen of the groups were outside of the reserve and nine were inside the reserve (Figure 4.2). The Audiomoths were programmed to record continuously for 24 hours over a 2-day period in August-September 2019. They were configured to record WAV files at a sampling rate of 48 (kHz) with a medium gain. This sampling rate was chosen as it encompassed the frequency range for most anthropogenic noise and pygmy marmoset calls.

The experiment underwent the Royal Holloway's ethical review process as some marmoset groups are located in a local community and therefore the Audiomoths will pick up human conversations, the experiment was approved by the college. The recordings are kept on password-protected hard drives and permission was given by the landowners before the Audiomoths were placed near their homes.



Figure 4.2. A map of the locations of the marmoset groups where the Audiomoths were placed in August 2019 with green diamonds denoting a group located inside the reserve and a purple diamond representing a group located outside of the reserve boundary.

## 4.2.4 Data extraction

## 4.2.4.1 Anthropogenic Noise

Once the data was collected the WAV files were processed in Adobe Audition. 12 hours (from 05:30 to 17:30) was analysed for anthropogenic noise and calls for each day of recordings, with the total recording time analysed for each group being 24 hours. The WAV files were thirty minutes in length and were viewed in Audition, cataloguing each call as well as any

anthropogenic noise recorded (in number of seconds heard, see Chapter 3 for the breakdown of the categories of anthropogenic noise). The total time heard of all the anthropogenic noise was summed for each group (see Figure 4.7 in the appendix for the total hours heard of anthropogenic noise per group). When heavy rain was encountered in the recording the files were not added to the analysis (37 files across all groups) however because the recordings were conducted in the dry season the bouts of rain would only last 1-2 hours at a maximum so it was not a significant detractor in the amount of anthropogenic noise being recorded.

#### 4.2.4.2 Call Data Extraction

Once all calls were documented in the sound files, fifteen calls for each call type (trill, j call, long call, and tsik) for each group were randomly selected using random integer set generator (https://www.random.org/integer-sets/). The ten calls which were the best quality (e.g. no other overlapping noises) were selected and were then analysed in Raven Pro 1.6. The following was recorded for each call; number of subsequent calls in the call bout (15 seconds without a call ended the bout), duration of the call (ms), minimum frequency (kHz), maximum frequency (kHz), peak frequency (kHz) and for long calls the cycle i.e. the number of notes in the call. Where a call is defined as the vocalisation overall, a bout is a series of the same call over a period, notes are the number of syllables that make up a call sequence.

#### 4.2.5 Statistical Analysis

#### Call Data

The statistical program RStudio version 1.1.456 (R Core Team, 2020) was used to run a MANOVA on each call type to examine which call characteristics were significantly different across groups or with total hours of anthropogenic noise heard. This was done as the total hours of anthropogenic noise heard analysis was testing our 'hypothesis' variable and group served as our 'control'. I chose to use a MANOVA for these tests as it allows for the test of the effects of an independent categorical variable on multiple dependent variables, in this case it allowed me to have group and number of hours of exposure as the independent variables and the minimum, maximum, peak frequency, the number of calls and duration as the dependent variables. An ANOVA was run to see which of the call characteristics frequency differed significantly across hours of anthropogenic noise heard and group.

# 4.3 Results

# 4.3.1 Call Data

A total of 920 calls were analysed across 23 groups and 4 call types (see Table 4.1 for a breakdown of call characteristics).

Table 4.1.	The averages a	and standard	deviations f	for all of t	the call char	acteristics of	of the 4 of	call
types, N=9	20 calls.							

Call Type	Minimum Frequency (kHz)	Maximum Frequency (kHz)	Peak Frequency (kHz)	Duration (ms)	Number of calls in the bout	Cycle (Number of notes)
Trill	$8.23 \pm 0.77$	$12.35 \pm 0.88$	$10.21 \pm 1.26$	$417.48 \pm 108.37$	1.9 ± 1.81	N/A
J Call	$11.47 \pm 5.53$	$15.6 \pm 2.32$	$11.82 \pm 2.21$	$178.28 \pm 66.33$	$4.18 \pm 8.71$	N/A
Long Call	9.71 ± 6.54	$10.96 \pm 1.07$	10.09 ±0.71	$1649.34 \pm 644.43$	$1.69 \pm 1.57$	3.56 ± 1.34
Tsik Call	$14.04 \pm 2.92$	21.13 ± 1.89	$19.86 \pm 2.47$	$167.7 \pm 81.84$	3.58 ± 3.92	N/A

# Trills

Across all call characteristics, there was no effect of number of hours of anthropogenic noise (p = 0.118, N=230 calls), but there were significant differences between groups (MANOVA, F= 4.971, p<0.001). Specifically, groups differed in the minimum, maximum and peak frequency, number of notes and duration (ANOVA, Table 4.2, Figure 4,3).

Table 4.2. Results for the ANOVA on trill call characteristics showing a strong effect of group across all call characteristics, N=230 calls.

Call Characteristic	Hours	Group
Minimum Frequency	F=1.185 p=0.278	F=16.530 p < 2 x e <sup>-16</sup>
Maximum Frequency	F=1.986 p=0.160	F=7.183 p= 1.514 x e <sup>-15</sup>
Peak Frequency	F=0.875 p=0.351	F=3.526 p=1.456 x e <sup>-06</sup>
Number	F=1.503 p=0.222	F=1.966 p=0.009
Duration	F=2.757 p=0.098	$F = \overline{5.892}$ p=1.748 x e <sup>-12</sup>



## J calls

Across all call characteristics, there was an effect of number of hours of anthropogenic noise (MANOVA, F=4.8, p<0.001, N=230 calls) and there were significant differences between groups (MANOVA, F= 3.535, p<0.001, N=230 calls). Specifically, maximum frequency decreased with increasing hours exposed to anthropogenic noise and duration increased with more hours of anthropogenic noise (ANOVA, Table 4.3, Figure 4.4).

Table 4.3. Results for the ANOVA on j call characteristics showing a strong effect of group on all call characteristics, N=230 calls.

Call Characteristic	Hours	Group
Minimum	F=2.166	F= 3.865
Frequency	P=0.143	$p= 2.01 \text{ x e}^{-07}$
Maximum	F=5.133	F=5.858
Frequency	p=0.025	p=2.11 x e <sup>-12</sup>
Peak	F=2.955	F=3.014
Frequency	p= 0.087	p= 2.843 x e <sup>-05</sup>
Number	F=3.96 p=0.067	F=3.960 p=1.152 x e <sup>-07</sup>
Duration	F=18.093 p=3.186 x e <sup>-05</sup>	F=7.290 $p=8.541 \text{ x e}^{-16}$



# Long Calls

Across all call characteristics, there was an effect of number of hours of anthropogenic noise (MANOVA, F=10.103, p<0.001, N=230 calls) and there were significant differences between groups (MANOVA, F= 4.59, p<0.001, N=230 calls). Specifically, minimum frequency, peak frequency and the number of notes (cycle) decreased with increasing hours exposed to anthropogenic noise. Duration increased with more hours of anthropogenic noise (ANOVA, Table 4.4, Figure 4.5).

Table 4.4 Results for the ANOVA on long call characteristics showing the strong effect of group on all call characteristics, N=230 calls.

Call Characteristic	Hours	Group
Minimum	F= 28.625	F=9.504
Frequency	$p=2.782 \text{ x e}^{-09}$	$p < 2.2 x e^{-16}$
Maximum	F= 3.001	F= 11.516
Frequency	p=0.0847	$p < 2 x e^{-16}$
Peak	F= 26.013	F=9.352
Frequency	$p=7.644 \text{ x } \text{e}^{-07}$	$p < 2.2 x e^{-16}$
Number	F=0.176	F=1.856
	p= 0.675	p=0.016
Duration	F=5.134	F=3.525
	p= 0.025	$p=1.469 \text{ x e}^{-06}$
Cycle (number of	F= 4.343	F=6.651
notes)	p= 0.038	$p=2.652 \text{ x e}^{-14}$



Tsiks

Across all call characteristics, there was an effect of number of hours of anthropogenic noise (MANOVA, F=2.967, p<0.05, N=230 calls) and there were significant differences between groups (MANOVA, F= 5.367, p<0.001, N=230 calls). Specifically, maximum frequency and peak frequency both increasing in frequency with increasing hours exposed to anthropogenic noise (ANOVA, Table 4.5, Figure 4.6).

Table 4.5 Results for the ANOVA on tsik call characteristics showing the strong effect of group on all call characteristics expect the number of calls in the bout, N=230 calls.

Call Characteristic	Hours	Group
Minimum	F=1.345	F= 4.886
Frequency	p=0.248	$p=5.302 \text{ x e}^{-10}$
Maximum	F= 6.441	F= 19.280
Frequency	p=0.012	$p < 2 x e^{-16}$
Peak	F=4.400	F=18.545
Frequency	p=0.037	$p < 2 x e^{-16}$
Number	F=0.159	F= 0.971
	p=0.690	p=0.501
Duration	F=3.536	F= 9.460
	p=0.0615	$p < 2 x e^{-16}$



#### 4.4 Discussion

I found differences in spectral and temporal call characteristics on a gradient of exposure to anthropogenic noise for three of the four call types (expect trill calls). Groups exposed to more anthropogenic noise had long calls with a lower low and peak frequency, fewer notes and the calls were significantly longer. J calls had the same pattern with the maximum frequency becoming significantly lower the higher the exposure and the duration of the calls becoming significantly longer with more exposure. This is consistent with other studies of marmoset species which increase the duration of their call syllables with an increase in background noise levels (common marmoset; Brumm et al., 2004) and in 'nosier' urban areas (black tufted-ear marmoset; Santos et al., 2017). These differences could arise as higher sound frequencies are less effective at travelling longer distances (Wiley and Richards, 1978) and as the long call is a long-distance contact call it is essential that it can permeate further across landscapes. Tsik calls were also found to vary with hours of anthropogenic noise heard, except that the maximum and peak frequencies were higher with more exposure. This pattern is a more common finding in cetaceans (Parks et al., 2007) and birds (Slabbekoorn and Peet, 2003) than in primates, primates have been found to shift their calls to lower frequencies in nosier environments. As this is an alarm call it is more important that it escapes the effects of potential masking in a shorter radius rather than being able to carry over long distances, which could be why we see this pattern. These results indicate that for at least three calls there is an impact of anthropogenic noise on a gradient of number of hours exposed to anthropogenic noise, which provides the first evidence for these impacts on wild primates found on an exposure gradient.

I found differences between groups across almost all call characteristics and call types. This variation at the group level is expected as group-specific variations/dialects are found in both monkeys and apes (Fedurek and Slocombe, 2011). Between group differences have also already been shown in the western pygmy marmoset, de la Torre and Snowdon (2009) showed groups within a wild population of western pygmy marmosets in Ecuador have consistent structural differences in their trill and j call structures between the populations studied. Snowdon postulates that the dialects are most likely due to social learning or social plasticity (Snowdon, 2017). Red-bellied tamarins (*Saguinus labiatus*) in Bolivia also have dialects in their long calls, with differences in the frequency range and the peak frequencies between two populations (Maeda

#### and Masataka, 1987).

Using evidence provided by previous studies conducted with marmosets and other primate species I postulate that the driving selection pressure on eastern pygmy marmoset call shifts is mix of both familial/social pressure and environmental. As previously discussed the western pygmy marmoset can develop dialects due to social induced plasticity and habitat acoustics did not predict the call structures of the populations studied (de la Torre and Snowdon, 2009). The socially plastic nature of the pygmy marmoset calls was also shown in a study by Snowdon et al. (1999), where individuals would modify the structure of trill calls when paired with a new mate. Their results suggest that nonhuman primates, specifically pygmy marmosets, will modify the vocal structure of their calls in responses to social pressure. The fact that social pressure is a driving pressure for nonhuman primates shows the multifaceted nature of what drives vocal structure change. Another pressure is the environment, pressures like habitat type can also cause animals to have to adapt their vocalisations as theorised by the acoustic adaptation hypothesis (Ey and Fisher, 2009). Brown et al. (1995) shows that the vocalisations of two rain forest monkeys (grey-cheeked managabeys, Cercocebus albigena, and blue monkeys, Ceropithecus *mitis*) have undergone a stronger selection for reduced distortion in the physical form of their calls then that of two savanna species (yellow baboons, Papio cynocephalus, and vervet monkeys, Cercopithecus aethiops). This change occurs in the same species located in different habitats, with the black tufted-ear marmoset phee vocalisation characteristics changing when in a 'nosier' urban area compared to a rural national park (Santos et al., 2017). This mix in vocal selection pressures also occurs in frogs, with Goutte et al. (2018) finding some evidence to support the acoustic adaptation hypothesis, however, they did not find the differences in temporal call features to be driven by the frog's habitat. This shows that the mechanisms by which call structure has evolved is complicated and is not driven by one singular force.

This study is not without its limitations. I was not able to collect habitat and ambient sound data due to Covid-19 and travel restrictions. With both habitat and ambient sound data the influence of other environmental pressures could have also been explored in the analyses to have a clearer understanding of what specific change anthropogenic noise is driving. I also was not able to distinguish which individual in the group is calling so therefore there could be pseudo-replication in the individual calls studied. In a future study it would be ideal if the anthropogenic noise,

ambient noise level and habitat data for each group is recorded and then investigating how each of these factors impacts the differences in these vocalisations of the eastern pygmy marmoset.

Primates are inherently social animals and communication plays a role in all aspects of life (Fedurek and Slocombe, 2011), which is why their ability to display vocal plasticity plays a large role in their ability to adapt to changing environments and is found in a wide variety of primate species. Female olive baboons (*Papio Anubis*) modify their grunt calls by making them longer and at a lower frequency to better communicate in varying habitats (Ey et al. 2009). Pygmy marmosets (*Cebuella pygmaea*) alter the structure of their contact calls when placed with an unfamiliar group to make them sound more acoustically similar, showing the ability of vocal learning (Elowson and Snowdown, 1994). Japanese macaques (Macaca fuscata) reply to coo calls played to them at that same frequency that they were called at, showing that these changes can occur on even shorter time scales (Sugiura, 1998). The fact that these animals can be so vocally plastic may also be factoring into their responses to abiotic external stimuli. Suggesting that with the plastic nature of pygmy marmoset calls, as shown in previous work and in this chapter, that they could be able to shift their calls in the short term when actively being exposed to anthropogenic noise or in periods of time when the exposure to anthropogenic noise is higher on average. Addressing this in future work would fill the current knowledge gap of the shortterm vocalisation changes wild primates are making to adapt to environments with higher levels of anthropogenic noise disturbance.

The environment is a pressure driving change in vocal communication structure but as anthropogenic noise infiltrates these formally wild areas it is important to understand how this added environmental pressure further impacts how animals are shifting their calls. This study adds to the current literature that suggests that anthropogenic noise is now another factor that must be considered when looking at changes in vocal structure. Underscoring the importance of understanding this impact as human expansion and urbanisation increases and noise pollution reaches even further into formally wild spaces. This chapter also reinforces the importance of studying these disturbances on a gradient of exposure not just as site (high-low) comparisons as the implementation of a gradient provides a more nuanced analyses of where vocalisation shifts are occurring. In sites like the Área de Conservación Regional Comunal Tamshiyacu Tahuayo a mitigation plan to reduce the amount of noise disturbance the pygmy marmosets are exposed to through ecotourism, could be to only approach the groups with motors turned off, lowing speeds while driving past group locations, and limiting talking while observing these groups.

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# 4.6 Appendix



Figure 4.7 Visual breakdown of how many hours of anthropogenic noise each group was exposed to over the 24-hour period analysed. The groups above the black line are the groups outside of the reserve boundary and the ones below are located inside the reserve.

Pygmy Marmoset	Location Description	Reserve Location
TL2	Along the river edge close to the main lodge.	Out
TL5	Downriver from the main lodge close to an oxbow lake.	In
TL6	In a forested area between the main lodge and El Chino	Out
TL7	Close to some houses upriver from El Chino.	Out
TL8	On a short cut taken during the rainy season on the Rio Blanco.	Out
TL10	Very close to the main lodge behind one of the tourist rooms.	Out
TL11	Further in the forest behind the main lodge.	Out
CV1	Close to the river from El Chino.	Out
CV2	Behind an occupied house in the middle of Chino.	Out
CV3	On the river turnoff into El Chino from the main lodge close to an abandoned house.	Out
CV4	Behind an occupied house on the outskirts of Chino.	Out
CV5	Located close to a stream on the outskirts of El Chino.	Out
CV6	Located in the forest behind El Chino.	Out
RC1	Across the river from the research centre.	In
RC2	Close to the river's edge by the turn off for the trail to Yarina Lake.	In
RC3	On the trail to a highly tourist visited oxbow lake (Yarina).	In
RC4	Located in the grid system behind the research system.	In
RC6	Close to the river's edge by the turn off for the trail to Yarina Lake.	In
RC7	On the river's edge downriver from the research centre.	In
RC8	Located in the grid system behind the research system.	In
RC9	Far upriver from the research centre on the river bank.	In
RC10	Far upriver from the research centre on the river bank.	In
RC11	Located in the grid system behind the research centre.	In

Table 4.7 List of the marmoset groups and a description of their locations.