CORRECTION

Correction: Lombard effect onset times reveal the speed of vocal plasticity in a songbird

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There was an error published in *J. Exp. Biol.* **220**, 1065-1071.

Eqn 2 was displayed incorrectly. The correct equation appears below.

$$\text{SPL increase} = \sqrt{2^{\left(\frac{\text{dB increase}}{6 \times 2}\right)}}.$$

We apologise to the authors and readers for any inconvenience this may have caused.

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RESEARCH ARTICLE

Lombard effect onset times reveal the speed of vocal plasticity in a songbird

Samuel I. Hardman[1,2,*], Sue Anne Zollinger[2], Klemen Koselj[3], Stefan Leitner[4], Rupert C. Marshall[1] and Henrik Brumm[2]

ABSTRACT

Animals that use vocal signals to communicate often compensate for interference and masking from background noise by raising the amplitude of their vocalisations. This response has been termed the Lombard effect. However, despite more than a century of research, little is known how quickly animals can adjust the amplitude of their vocalisations after the onset of noise. The ability to respond quickly to increases in noise levels would allow animals to avoid signal masking and ensure their calls continue to be heard, even if they are interrupted by sudden bursts of high-amplitude noise. We tested how quickly singing male canaries (*Serinus canaria*) exhibit the Lombard effect by exposing them to short playbacks of white noise and measuring the speed of their responses. We show that canaries exhibit the Lombard effect in as little as 300 ms after the onset of noise and are also able to increase the amplitude of their songs mid-song and mid-phrase without pausing. Our results demonstrate high vocal plasticity in this species and suggest that birds are able to adjust the amplitude of their vocalisations very rapidly to ensure they can still be heard even during sudden changes in background noise levels.

KEY WORDS: **Serinus canaria, Bird song, Amplitude, Noise, Bioacoustics, Response time**

INTRODUCTION

Acoustic communication is often constrained by the effects of background noise, which can mask and degrade acoustic signals, preventing them from being recognised or discriminated by their targeted receivers. To overcome this problem, animals may adjust their acoustic signals in a variety of different ways, including increasing the duration of brief calls (Brumm et al., 2004), increasing the redundancy of their vocalisations by producing longer and more repetitive signal series (Brumm and Slater, 2006; Kaiser and Hammers, 2009), shifting the timing of their vocalisations (Fuller et al., 2007; Vargas-Salinas and Amézquita, 2013) or increasing the pitch of their vocalisations (Slabbekoorn and Peet, 2003; Parks et al., 2007). One of the most efficient and widespread methods by which animals reduce the impact of signal masking is by raising the amplitude of their vocalisations (Brumm and Zollinger, 2011; Hotchkin and Parks, 2013). This phenomenon has been termed

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the Lombard effect in honour of its discoverer, the French otolaryngologist Etienne Lombard (Zollinger and Brumm, 2011), and it has been shown to be much more effective at increasing signal detectability in noise than increasing either the duration or repetition of a vocalisation (Luo et al., 2015). Moreover, although not always true (Brumm and Zollinger, 2013; Hage et al., 2013), it has been suggested that in some cases increases in the pitch of vocalisations in response to noise may in fact be a by-product of calling more loudly (Osmanski and Dooling, 2009; Schuster et al., 2012).

The Lombard effect has now been shown across a diverse range of taxa including mammals, such as humans, monkeys, cetaceans and bats (Hotchkin and Parks, 2013), and numerous species of paleognath and neognath birds (Brumm and Zollinger, 2011). The situation in amphibians is still unresolved, as one study recently found a noise-dependent regulation of call amplitudes in a frog (Halfwerk et al., 2016), whereas a previous study failed to find evidence for the Lombard effect in other anuran species (Love and Bee, 2010). Recently, the presence of the Lombard effect was also reported in a fish (Holt and Johnston, 2014), but the data are difficult to interpret because it is not clear whether and how the noise amplitudes were accounted for in the signal measurements in that study.

The widespread taxonomic distribution of the Lombard effect suggests it is one of the principal mechanisms by which birds and mammals, and perhaps also other vertebrates, improve the detectability of their vocalisations in noise (Brumm and Zollinger, 2011). Furthermore, within these groups it is likely that the Lombard effect occurs independently of learning, as in birds it occurs both in species which acquire their vocalisations through vocal production learning (Cynx et al., 1998; Brumm and Todt, 2002) and in those that do not (Potash, 1972; Leonard and Horn, 2005; Schuster et al., 2012). Studies of humans also suggest that while the Lombard effect usually occurs involuntarily, it may be affected by the social context (Amazi and Garber, 1982; Lu and Cooke, 2008) or linguistic content of the vocalisation (Patel and Schell, 2008), and may also be voluntarily controlled to some extent by cognitive processes (Pick et al., 1989; Tonkinson, 1994).

Evidence for the Lombard effect in animals is extensive. However, with the exception of one study that exposed greater horseshoe bats (*Rhinolophus ferrumequinum*) to short 30 s bursts of white noise (Hage et al., 2013), it has so far only been demonstrated in either wild animals living in continually noisy environments or in captive animals exposed to long periods of synthetic noise (Table 1). Early studies of the Lombard effect often used the term 'Lombard reflex' (e.g. Egan, 1971; Junqua, 1996), possibly hinting that the Lombard effect is typically exhibited very quickly in response to noise. This was shown in humans by Bauer et al. (2006), who found an onset latency of 157 ms when the amplitude of the auditory feedback of a speaker's own voice was increased via headphones. Foery (2008) found a similar onset latency of 127 ms in humans exposed to playbacks of noise.

Table 1. Duration of noise exposure in studies of the Lombard effect in non-human animals

Species	Context	Duration of noise exposure (SPL re. 20 µPa unless otherwise stated)	Reference
Birds			
Japanese quail (*Coturnix coturnix japonica*)	Captive	3 h prior to recordings (48–63 dB)	Potash (1972)
Zebra finch (*Taeniopygia guttata*)	Captive	Until a minimum of five vocalisations had been produced (60–90 dB in 5 dB increments)	Cynx et al. (1998)
Budgerigar (*Melopsittacus undulatus*)	Captive	Until the bird had produced 75 calls; during the first 25 calls, noise was played at 55 dB followed by 70 dB for the second 25 calls and 55 dB again for the last 25 calls	Manabe et al. (1998)
Common nightingale (*Luscinia megarhynchos*)	Captive	20 min or until the bird had sung 27 songs, repeated at 5 dB noise increments between 55 and 75 dB	Brumm and Todt (2002)
Bengalese finch (*Lonchura striata domestica*)	Captive	100 s per noise treatment played successively at 40–70 dB at 10 dB increments	Kobayasi and Okanoya (2003)
Blue-throated hummingbird (*Lampornis clemenciae*)	Wild	Until the bird stopped producing chipping calls (35 and 40 dB)	Pytte et al. (2003)
Common nightingale (*Luscinia megarhynchos*)	Wild	Continuous urban noise (40–64 dB)	Brumm (2004)
Tree swallow (*Tachycineta bicolor*)	Captive and wild	Wild birds – continuous ambient noise (41–67 dB) Captive birds – at least 1 h (55 and 65 dB)	Leonard and Horn (2005)
Domestic fowl (*Gallus gallus*)	Captive	6 min per treatment with four successive noise treatments (60, 67, 75 and 80 dB)	Brumm et al. (2009)
Budgerigar (*Melopsittacus undulatus*)	Captive	Until the bird had produced 60 vocalisations (40–90 dB)	Osmanski and Dooling (2009)
Noisy miner (*Manorina melanocephala*)	Wild	Continuous urban noise (50.83–65.80 dB)	Lowry et al. (2012)
Elegant crested tinamou (*Eudromia elegans*)	Captive	Until the bird had called 12 times (45 and 65 dB in 5 dB increments)	Schuster et al. (2012)
Atlantic canary (*Serinus canaria*)	Captive	20 s bursts of noise introduced mid-song (75 dB)	Present study
Amphibians			
Cope's grey treefrog (*Hyla chrysoscelis*)	Captive	6 min (40, 50, 60, 70 dB), Lombard absent in this species	Love and Bee (2010)
Túngara frog (*Physalaemus pustulosus*)	Captive	1 min (54–94 dB)	Halfwerk et al. (2016)
Mammals			
Crab-eating macaque (*Macaca fascicularis*), southern pig-tailed macaque (*Macaca nemestrina*)	Captive	Until 10 vocalisations had been produced at each of five playback levels in ascending and descending intensity (70, 80, 90, 80, 70 dB)	Sinnott et al. (1975)
Common marmoset (*Callithrix jacchus*)	Captive	30 min per noise treatment played successively in a random order at 40, 50, 60 and 65 dB	Brumm et al. (2004)
Cotton-top tamarin (*Saguinus oedipus*)	Captive	Until 11 calls in both 50 and 70 dB noise had been produced	Egnor and Hauser (2006)
North Atlantic right whales (*Eubalaena glacialis*)	Wild	Continuous exposure to environmental noise (92–143 dB re. 1 µPa)	Parks et al. (2011)
Mexican free-tailed bat (*Tadarida brasiliensis*)	Captive	A minimum of 100 echolocation pulses (55, 65, 75, 85 dB)	Tressler and Smotherman (2009)
Killer whale (*Orcinus orca*)	Wild	Continuous noise exposure from passing ships (~96–118 dB re. 1 µPa)	Holt et al. (2009)
Greater horseshoe bat (*Rhinolophus ferrumequinum*)	Captive	30 s (80, 90, 100 dB)	Hage et al. (2013)
Pale spear-nosed bat (*Phyllostomus discolor*)	Captive	28, 40, 52 dB (6 min)	Luo et al. (2015)
Fish			
Blacktail shiner (*Cyprinella venusta*)	Captive	17 min to 2.5 h; playback noise equivalent to ambient noise levels in nesting sites and 10.2 dB higher than the quiet treatment	Holt and Johnston (2014)

SPL, sound pressure level.

However, despite this and more than a century of research on the subject, only one study has yet directly tested how quickly the Lombard effect can be triggered in a non-human animal. Hage et al. (2013) found that greater horseshoe bats (*Rhinolophus ferrumequinum*), with their highly specialised auditory orientation system, may adjust the amplitude of their echolocation calls as fast as approximately 150 ms, but data on other taxa, and especially on vocalisations used for communication, are still lacking. This omission from the literature is surprising given that many animals are often exposed to sudden changes in noise levels. In undeveloped natural habitats, falling branches, gusts of wind or the calls of conspecifics and heterospecifics are potential sources of intermittent high-amplitude noise bursts (Luther and Gentry, 2013). In urban areas, peaks in traffic during the morning and evening rush hours contribute to a daily fluctuation in noise levels, while short bursts of

often very loud noise from sources such as car alarms, car horns, construction work or passing vehicles are common throughout the day (Warren et al., 2006; Luther and Gentry, 2013).

The ability to rapidly increase the amplitude of their vocalisations would allow animals to avoid signal masking and ensure their calls continue to be heard, even if their vocalisations are interrupted by very sudden bursts of high-amplitude noise. This is likely to be particularly important for species whose vocalisations encode information as complex sequences of different elements because masking of any part of these signals may prevent the correct messages from reaching their targeted receivers. For species whose signals encode warnings about threats or predators (e.g. Seyfarth et al., 1980; Templeton et al., 2005) it may be even more important to ensure that they are successfully transmitted. As animal vocalisations are also often used for territory defence and mate

attraction (Seyfarth and Cheney, 2003), intermittent masking of these vocalisations may lead to reduced breeding success in some species. The ability to avoid signal masking during sudden bursts of noise is therefore likely to be strongly favoured by selection.

On a proximate level, the Lombard effect demonstrates that the auditory system continuously monitors vocal output and uses this feedback to modulate the sound pressure level of vocalisations according to the strength of the background noise (Eliades and Wang, 2012). The speed at which the Lombard effect can act is therefore dependent on the neurons of the auditory system, and the speed with which the muscles of the sound-producing organ can respond to neuronal stimulation (Suthers and Zollinger, 2008). In mammals, the neurons that mediate the Lombard effect seem to be located in the brainstem (Nonaka et al., 1997; Hage et al., 2006), while cortical structures seem to be able to modulate this brainstem-based network (Eliades and Wang, 2012). The ability of humans to voluntarily control the Lombard effect to some degree also points towards some involvement of the motor cortex (Brumm and Zollinger, 2011). These studies suggest that control of the Lombard effect is sophisticated and likely requires well-developed auditory and motor control systems. Understanding how quickly animals are able to exhibit the Lombard effect will provide insight into how these systems function together and may also advance our understanding of the flexibility and plasticity of vocal behaviours in animals.

We addressed this topic in a small passerine bird, the canary [*Serinus canaria* (Linnaeus 1758)]. Male canaries have diverse and well-studied vocal repertoires that are used in mate attraction and stimulation (e.g. Leitner et al., 2001; Voigt and Leitner, 2008; Leboucher et al., 2012). The use of minibreaths during rapid trills, which allows for uninterrupted songs consisting of very long repetitive phrases of the same element type, makes the canary an ideal model to investigate mechanisms of rapid song modulation (Suthers et al., 2012). Furthermore, different aspects of song production and vocal control mechanisms have been studied in this species (e.g. Leitner and Catchpole, 2004; Bolhuis and Gahr, 2006; Suthers et al., 2012), which will eventually allow placing new findings on the mechanisms of vocal plasticity into a broader behavioural physiology context (Elemans et al., 2015). We examined changes in the sound pressure level of the song of canaries during sudden short and sporadic bursts of broadband white noise in order to discover how rapidly they exhibit the Lombard effect.

MATERIALS AND METHODS
Animals
Recordings were taken from seven adult male canaries kept under license (license number: Az.: 311.5-5682.1/1-2014-021). One female canary was used to encourage the males to sing. All birds were bred and raised in aviaries at the Max Planck Institute for Ornithology (Seewiesen, Germany). Experiments were performed under a 14 h:10 h light:dark cycle, and the birds had constant access to *ad libitum* food and water supplemented with fresh vegetables, cuttlebones and grit.

Apparatus
Prior to experiments, all male birds were kept together in an aviary (1.95×1.0×1.8 m). The female was kept in a separate cage (120×80×60 cm). Recordings were made in a separate aviary (1.95×1.0×1.8 m) lined with acoustic tiles which was visually and audibly separated from the other birds. During recordings, individual males were placed inside the recording aviary inside a wire cage (60×40×40 cm) within view of the female (in a separate cage 2 m away). An omni-directional microphone (Sennheiser

ME62) connected to a PC using an external soundcard (Edirol UA-101) and the recording software Sound Analysis Pro (Tchernichovski et al., 2000; version 1.085) were used for all audio recordings. Recordings were made with a sampling rate of 44.1 kHz and 16 bit accuracy. The microphone was placed 60 cm above the centre of the cage, halfway between the two perches, to minimise variation in the recorded sound level caused by lateral movements of the bird's body and head. Recordings were triggered automatically whenever the bird sang using the trigger-record function in Sound Analysis Pro.

To induce the Lombard effect, white noise (0.1–16 kHz) was played towards the birds during singing bouts. The noise was broadcast through a JBL Pro III loudspeaker placed 140 cm away from the cage and connected to a Pioneer A109 stereo amplifier. The sound pressure level of the noise was 75 dB (re. 20 μPa) measured from inside the cage at the position of the perches. A custom MATLAB (version 7.5.0; www.mathworks.com) routine was used to automatically trigger noise playbacks whenever the sound pressure level and duration of a bird's song crossed a pre-defined trigger threshold. The trigger function was controlled using a microphone (Audio-Technica ATR3350) connected to an external soundcard (Edirol UA25) and the Playrec toolkit for MATLAB. Canary song is composed of a succession of phrases which are each formed from long repeats of multiple song elements of the same type (Poulsen, 1959; Nottebohm and Nottebohm, 1978) (Fig. S1). We set white noise playbacks to begin after a random delay of between 1 and 10 s after being triggered to ensure that the noise began during a different phrase and element type in each recording. Playbacks of white noise were always exactly 20 s long.

Analysis
All acoustic analyses were carried out using the software Avisoft-SASLab Pro (version 5.2.09; Avisoft Bioacoustics, Berlin, Germany). Different song element types were identified using spectrograms and each song element type was given a number unique to the element type itself and to the bird it came from. Most of the songs in this study began with a few quieter elements (usually around five to six elements) before the sound pressure level rapidly increased and stabilised. To prevent these initial quieter elements from affecting our results, they were excluded from our analyses and the sound pressure level of song elements was only measured after the point at which the rapid increase in sound pressure level had levelled out. The sound pressure level for each element in both noise (n=2390) and quiet conditions (n=1750) was measured with an averaging time of 10 ms. Per recording, a mean (±s.e.m.) of 11.23±1.69 elements were recorded before the onset of noise and 14.64±2.35 elements after the onset of noise. In our statistical analyses we only included data for element types which occurred both before and during the onset of noise.

The sound pressure level of the background noise was subtracted from these measurements using the following logarithmic computation procedure given by Brumm and Zollinger (2011) in order to calculate the sound pressure level of the song elements alone (L_{signal}):

$$L_{signal} = 10\log_{10}\left(10^{\left(\frac{L_{signal+noise}}{10}\right)} - 10^{\left(\frac{L_{noise}}{10}\right)}\right), \quad (1)$$

where $L_{signal+noise}$ is the sound pressure level of the song element and the background noise and L_{noise} is the sound pressure level of the background noise alone.
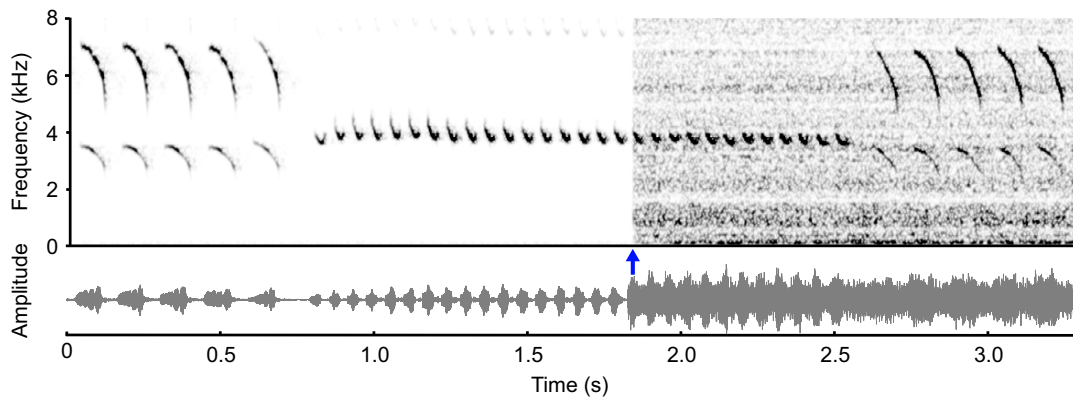
Fig. 1. Spectrogram showing canary song which began during quiet conditions and continued after noise began. The onset of noise is indicated by the blue arrow.

The software was calibrated by recording a sine tone of constant amplitude using the same microphone and software settings as used for the recordings of birdsong. The sound pressure level of this tone (68 dB, 1000 Hz) was directly measured using a sound pressure level meter (Voltcraft SL-400) at the position of the microphone.

The proportional increase in the sound pressure level (SPL increase) of song elements in noise was calculated using the equation:

$$\text{SPL increase} = \sqrt{2\left(\frac{\text{dB increase}}{6 \times 2}\right)}, \qquad (2)$$

where dB increase is the measured increase in dB of a song element.

For every song element sung during noise playbacks we also recorded how many seconds of the noise playback the bird had been exposed to before the element was sung (maximum 20 s). With this information we created a subset from our full dataset for song elements sung during the first 1 s after the onset of noise exposure. These data were used to determine whether canaries exhibit the Lombard effect within 1 s of exposure to noise.

In most of our recordings canaries stopped singing immediately after the noise playback began, before quickly resuming song again in the noise. However, in some cases the canaries continued to sing the same phrase uninterrupted during the quiet period and into the noise (Fig. 1). From these recordings we created a separate dataset of sound pressure level measurements to test whether canaries can adjust the sound pressure level of their songs mid-song and mid-phrase without pausing.

Statistical analyses

To determine whether the Lombard effect occurred within 20 s and within 1 s of the onset of noise, and to determine whether canaries are able to exhibit the Lombard effect mid-phrase without first interrupting their song, we analysed our data using separate generalised linear mixed models (GLMMs) in R (version 3.0.2; http://www.R-project.org/) using the package lme4 (Bates et al., 2015). We modelled the influence of noise exposure (binary fixed factor: present or absent) on the sound pressure level of the song elements (dependent variable) with normally distributed errors and an identity link function. The ID of the birds, the song element code and the recording (the audio file the data were taken from) were all included as random factors. We also included noise as a nested random factor within recordings to account for audio files containing song elements recorded during both quiet and noise exposure. We assessed the effect of noise exposure on the sound

pressure level of canary song by comparing models including noise exposure to null models using likelihood ratio tests with one degree of freedom. Where multiple analyses were carried out on the same dataset, Bonferroni corrections were used to account for multiple comparisons.

To more precisely determine the speed of the onset of the Lombard effect, we further analysed one exemplary element type from one individual (for which the most data were available) using a broken-line regression model fitted using the segmented package in R (Muggeo, 2008; Muggeo and Adelfio, 2010). This allowed us to precisely identify at what time after the onset of noise this element type was sung at a significantly higher sound pressure level than before the noise began.

RESULTS

In total, we measured 4140 song elements from seven birds (n=1750 before the onset of noise, 2390 during white noise exposure, song element types n=31). The sound pressure level of
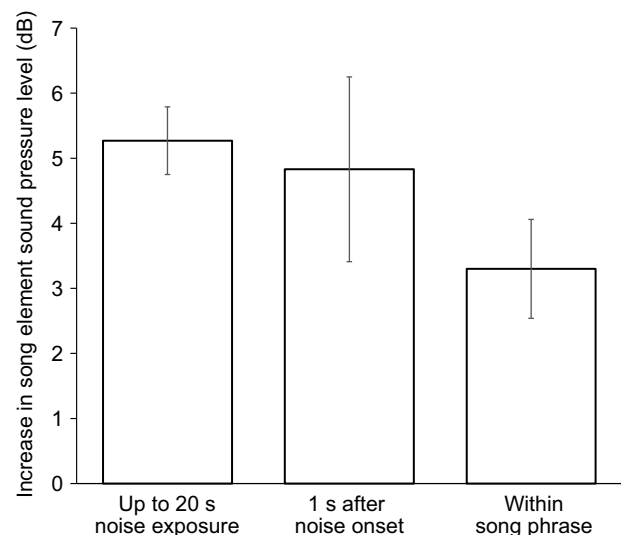

Fig. 2. Mean (±s.e.m.) increases in the sound pressure level of canary song elements sung during noise compared to the period before the onset of noise. Graph shows the increase in the sound pressure level of song elements sung up to 20 s after the onset of noise exposure, during the first 1 s of noise exposure and within song phrases which began during the quiet period and continued uninterrupted into the noise. All bars show a significant increase in the sound pressure level of elements sung after the onset of noise (P<0.05).

song from all seven birds was found to be higher during exposure to white noise and this was significant (Fig. 2). Song elements sung during the full 20 s of exposure to white noise (including elements from songs that were briefly interrupted at the onset of noise before continuing) had a sound pressure level that was on average 5.3 dB (±0.5 s.e.m.) higher than song elements sung before the onset of noise (χ^2=60.166, d.f.=1, Bonferroni-corrected P<0.0001). A subset from this dataset including only song elements sung during the first 1 s of noise exposure (n=636 before the onset of noise, 38 during white noise exposure, birds n=3, song element types n=5) showed that song elements sung during the first 1 s of noise exposure had a sound pressure level that was on average 4.8 dB (±1.4 s.e.m.) higher than song elements sung before the onset of noise (χ^2=9.430, d.f.=1, Bonferroni-corrected P=0.004). These results represent increases in sound pressure level of 84% and 75%, respectively (Fig. 2).

In many of our recordings we found that canaries often briefly stopped singing immediately after the noise playback began. However, in some of our recordings the birds began to sing a song phrase during quiet conditions and continued to sing the same phrase uninterrupted as a noise playback began (Fig. 1; n=159 elements sung before the onset of noise, 80 during white noise exposure, birds n=4, mean phrase length 1.5 s). From these recordings we created a separate dataset that allowed us to test whether canaries are able to exhibit the Lombard effect mid-song and mid-phrase without pausing. In these recordings, the sound pressure level of song elements sung during noise exposure was on average 3.3 dB (±0.8 s.e.m.) higher than song elements sung before the onset of noise (χ^2=8940, d.f.=1, P=0.002), representing a 46% increase in the sound pressure level (Fig. 2).

For the element types analysed using the broken-line regression model, we found that the sound pressure level of song elements sung more than 0.318 s after the onset of noise was significantly higher (P<0.05) than song elements sung before the onset of noise (Fig. 3).

DISCUSSION

This is the first study, to our knowledge, to directly test the onset latency of the Lombard effect in a bird, and the first to show that the Lombard effect is exhibited by canaries. Our results show that male canaries exhibit the Lombard effect approximately 300 ms after the onset of noise, and are able to do this mid-song and mid-phrase without pausing. Thus, we show that canaries possess a remarkably fast vocal plasticity which allows them to adjust their vocalisations in real time to mitigate the masking effects of sudden bursts of noise. Given that the Lombard effect is an ancient trait that is likely shared by all extant bird species (Brumm and Zollinger, 2011), it is possible that all vocalising birds exhibit similarly rapid response times. Moreover, while humans exhibit the Lombard effect within 150 ms (Bauer et al., 2006; Foery, 2008), greater horseshoe bats (*Rhinolophus ferrumequinum*) have been shown to increase the sound pressure level of their echolocation calls almost instantaneously when exposed to noise (Hage et al., 2013). These studies suggest that similarly rapid Lombard responses may be present in taxa other than birds.

In recent years, numerous studies have identified adjustments to the vocalisations of animals living in noisy environments which help them to mitigate the problem of signal masking (reviewed in Brumm, 2013). Most of these studies contrast the vocalisations of populations living in noisy and quiet environments and identify differences that may be adaptive. However, it is still not fully understood exactly how these differences arise. Several hypotheses have been proposed, including short-term vocal plasticity,
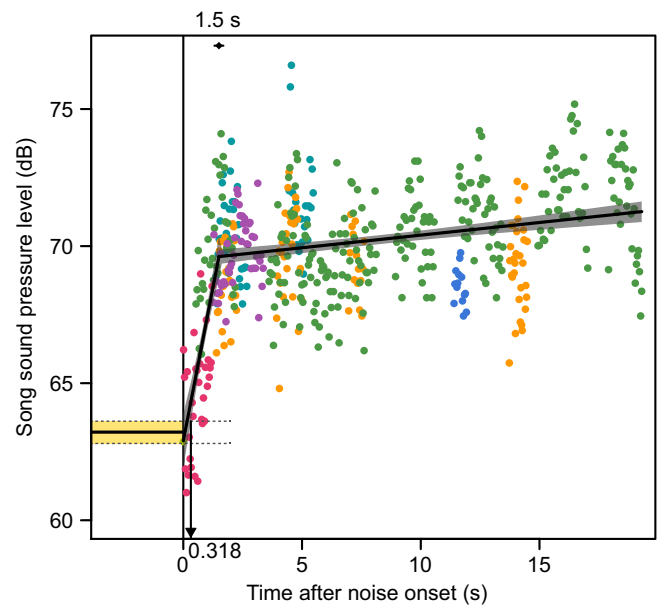
Fig. 3. Sound pressure level of individual canary song elements before and after the onset of noise. Data are depicted for one element type (from one individual), which had a high enough repetition rate to give a sufficient sampling size (n=251 elements measured before the onset of noise; 209 elements during white noise). Each point depicts a single vocalisation after the onset of noise, and different colours show measurements taken from different recordings. The mean sound pressure level of song elements sung in the 5 s before the onset of noise is depicted by a thick line with yellow bands and dashed lines showing the 95% confidence intervals for the mean. The thick line after the onset of noise was fitted using a broken-line regression model and shows the mean sound pressure level of song elements with grey bands showing the 95% confidence intervals. The onset of the Lombard effect is defined as the point at which the 95% confidence intervals for elements sung before and after noise no longer overlap, and occurred here at 0.318 s after the onset of noise (arrow). Elements sung after this time point were significantly louder (P<0.05) than elements sung before the noise began. The initial rapid increase in the sound pressure level of song elements stopped at 1.5 s, as shown at the top of the panel. After this point the increase in the sound pressure level slowed markedly.

long-term ontogenetic vocal adjustments, selective attrition of vocalisations that transmit poorly in noise, passive acquisition of vocalisations that transmit well in noise, and microevolutionary change (Patricelli and Blickley, 2006). The Lombard effect is a clear example of short-term vocal plasticity. The very fast reaction times demonstrated in our study suggest, in combination with the evidence for the perceptual efficiency of the Lombard effect for signal detection in noise (Nemeth and Brumm, 2010; Luo et al., 2015), that vocal plasticity is the key factor allowing animals to cope with the problem of signal masking in environments with unpredictable and fluctuating noise levels.

Studies on the speed of behavioural song plasticity in birds are rare, but the available evidence suggests that other song parameters can be modified in response to changes in the environment on time scales different or similar to the one we found for the Lombard effect. House finches (*Carpodacus mexicanus*) have been shown to shift the frequency of their song notes within the time it takes them to sing three songs to avoid masking by high-amplitude noise playbacks (Bermúdez-Cuamatzin et al., 2010), while black-capped chickadees (*Poecile attricapillus*) increase the frequency of their song notes on average after more than 1 min to avoid spectral overlap with lower frequency masking tones played back to them (Goodwin and Podos, 2013). In contrast, the onset of singing

activity in nightingales (*Luscinia megarhynchos*) can be triggered on average within 0.9 s to reduce temporal overlap by heterospecific songs (Brumm, 2006). In duetting bird species, the two partners of a pair may react even faster to integrate their duet parts into one coherent song (Hall, 2009; Templeton et al., 2013). Thus, the regulation of vocal onset in birds operates on a similar time scale as the Lombard effect, suggesting a similar role for the fast adjustment of signalling in fluctuating environments.

The rapid onset of the Lombard effect also indicates how quickly the auditory system can be integrated with the different motor systems to enable fast vocal plasticity (Bauer et al., 2006). For the Lombard effect to occur, a singing bird first needs to detect an increase in noise and then increase the contraction of abdominal and intercostal muscles to increase bronchial pressure, which eventually leads to an increase in song amplitude (Plummer and Goller, 2008). To stay on pitch during Lombard vocalisations (Templeton et al., 2016), birds need to decouple amplitude from frequency during vocal production, which could be achieved by a reduction of labial tension via the syringeal muscles or a reduction of air pressure in the interclavicular air sac via the respiratory muscles (Elemans et al., 2015).

Our study also sheds light on the question of what is the smallest unit of vocal production in birds. Cynx (1990) approached this question by interrupting the song of zebra finches (*Taeniopygia guttata*) and observing at what point in their songs the birds stopped singing. He found that zebra finches always stopped singing at discrete locations between song elements and never stopped halfway through a song element. That song elements in zebra finches may represent the smallest motor unit of song production was further supported by the discovery of Yu and Margoliash (1996) of precisely timed temporal correlations between discharge patterns in the vocal motor nucleus RA (robust nucleus of the archopallium) and individual song elements in singing birds. Franz and Goller (2002) later confirmed this result in the peripheral vocal production system, by showing that each song element sung by zebra finches corresponds to a single expiratory pressure pulse from the lungs. Evidence from measurements of peripheral vocal motor patterns in canaries also supports the hypothesis that individual elements represent the smallest units of song production in this species. Even in trills with a repetition rate of up to 30 elements per second, canaries take a rapid minibreath between each element, demonstrating that each element in these trills represents a discrete production unit (Suthers and Zollinger, 2008). However, unlike in zebra finches (Yu and Margoliash, 1996), almost nothing is known about the smallest motor units of song production in the canary brain. However, it has been shown that for the majority of canary element types there is a one-to-one relationship between air-sac compression and the production of individual notes (Hartley, 1990). In our study, canaries often interrupted their song almost immediately in response to the sudden onset of noise. As in zebra finches, we observed that canaries always stopped singing at discrete intervals between elements. Our recordings therefore suggest that song elements are also the smallest units of sound production in this species. Furthermore, as repeats of individual syllables in canary song are controlled by the HVC and the song pattern by the RA (Halle et al., 2003), the ability to stop singing so quickly after the onset of noise suggests extremely rapid modulation of this pathway.

In conclusion, our study adds to the growing number of studies that show that animals use the Lombard effect to improve communication efficacy in noise. Furthermore, we show for the first time that the Lombard effect can be exhibited extremely rapidly in response to sudden bursts of noise in birds. For animals that live in environments with highly variable and unpredictable background noise, this ability is likely to be of particular importance as it would allow them to maintain signal transmission despite sudden changes in noise levels.

Supplementary information
Supplementary information available online at
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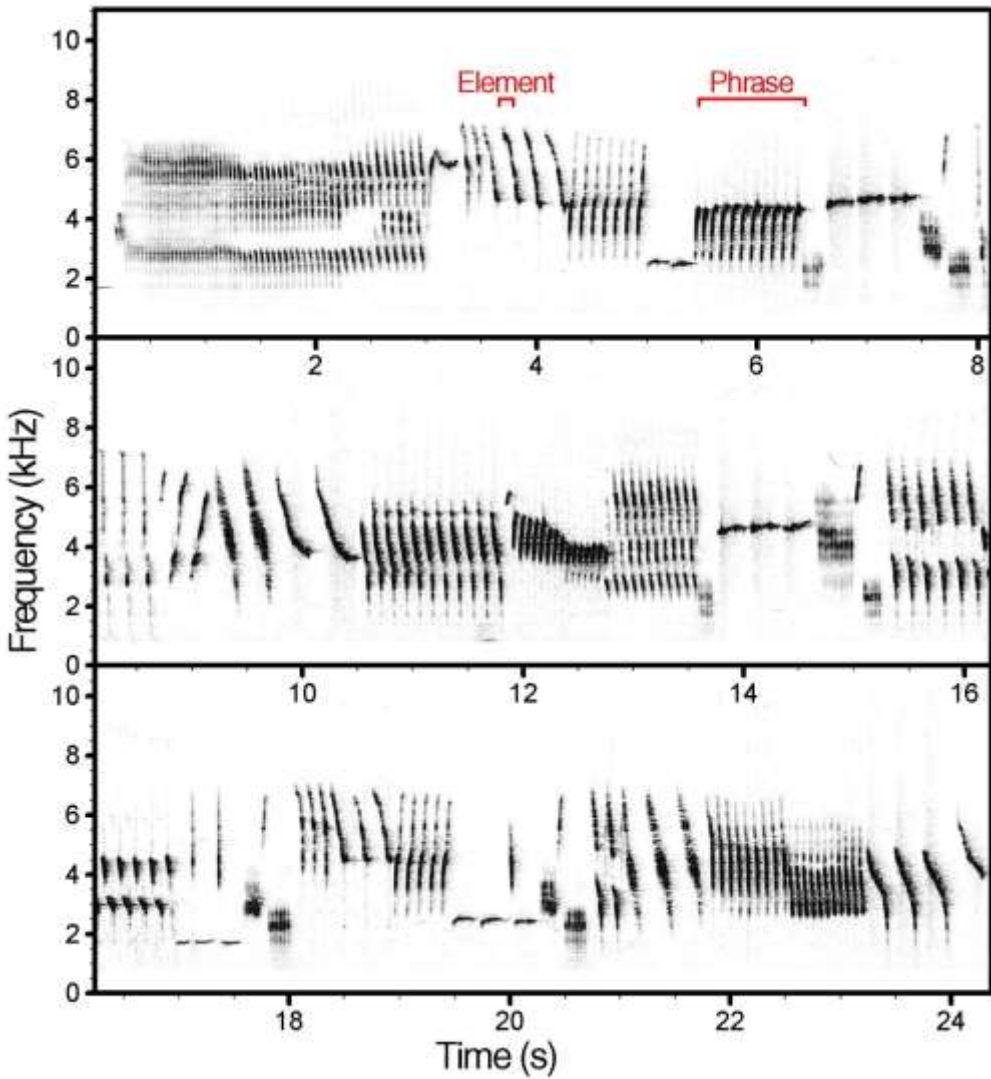
Fig. S1. Spectrogram showing a complete canary song with song element and song phrase labelled.

Table S1. Number of song elements and element types recorded per bird.

Bird ID	Number of song elements recorded	Number of different element types recorded
1	851	1
2	765	5
3	798	8
4	259	6
5	335	4
6	361	1
7	771	6
	Mean: 591.43 **Range: 592**	**Mean: 4.43** **Range: 7**