

Remote and automated detection of Asian hornets (*Vespa velutina nigrithorax*) at an apiary, using spectral features of their hovering flight sounds

Harriet Hall^a, Martin Bencsik^{a,*}, Nuno Capela^b, José Paulo Sousa^b, Dirk C. de Graaf^c

^a Physics and Mathematics, School of Science and Technology, Nottingham Trent University, Clifton Lane, Nottingham NG11 8NS, United Kingdom

^b Centre for Functional Ecology, Associate Laboratory TERRA, Department of Life Sciences, University of Coimbra, Calçada Martim de Freitas, 3003-531 Coimbra, Portugal

^c Department of Biochemistry and Microbiology, Ghent Universiteit, Sint-Pietersnieuwstraat 33, 9000 Gent, Belgium

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ABSTRACT

Asian hornets (*Vespa velutina nigrithorax*) are an invasive species that have spread across Europe since 2004. As *V. velutina* largely predate on honeybees, assessing their presence at apiaries would be useful for invasive species control programmes and beekeepers to help protect their hives. At present, hornet monitoring techniques are both costly and time consuming. A promising alternative is a remote detection strategy at apiaries, which would promote straightforward, non-invasive data acquisition. The remote capture of flight acoustics should benefit hornet detection as wingbeat frequencies have previously been described as ‘the fingerprint’ of some flying invertebrate species. We here demonstrate a non-invasive method of *V. velutina* detection using their hovering flight sounds, captured by microphones that can be left at an apiary over the long-term. Paired with a training algorithm (principal component analysis and discriminant function analysis) that discriminates between hornet flight and other external noises (honeybee flight sounds and general background noise), we demonstrate that hornet hovering acoustics exhibit specific spectral features that promote the detection of individuals at an apiary. The training algorithm in our study was highly accurate (98.7 %) when testing just under 1-hour of apiary recordings. Utilising two-dimensional-Fourier-transforms has also benefited this algorithm, as the analysis technique is ideal for identifying repeating features in sound/vibrational data, which are an inherent consequence of hovering hornet sounds. The experimental design and training algorithm used in this study have demonstrated excellent potential for hornet detection in the field, and are now ready to be tested on long-term, continuous data to further assess their success.

1. Introduction

The Asian hornet, or yellow-legged hornet (*Vespa velutina nigrithorax*), is native to Asia, but has been spreading across Europe and the UK as a result of accidental importation to France in 2004, followed by natural colonisation of the species (Lioy et al., 2022). For the European honeybee (*Apis mellifera*), this invasion poses a huge threat. Honeybees are the preferred food source of this hornet, whose predation technique involves ‘hawking’ (hovering) around the honeybee colony entrance for prolonged periods until a bee is caught, dismembered, and eaten (Laurino et al., 2019). The presence of hornets at an apiary can lead to a reduction in fitness (Dong et al., 2023), as well as cause ‘colony foraging paralysis’, where honeybees reduce their engagement in flight activities

around the hive entrance, therefore impacting upon foraging success (Requier et al., 2019). This paralysis has a negative effect on honey stores over the critical winter months and can lead to low overwintering survival in colonies (Requier et al., 2019). There is also an impact on egg laying and the number of workers produced in colonies of *A. mellifera* that are stressed from hornet attacks which has been shown to lead to colony death (Dong et al., 2023). Unfortunately, European honeybee defence strategies in response to hornet attacks are not as efficient as those of the Asian honeybee, which evolved in sympatry with the predator (Lioy et al., 2022). Reports show that European apiaries can be severely damaged by hornet presence, demonstrated through the weakening or destruction of colonies in high numbers (Requier et al., 2019; Laurino et al., 2019).

* Corresponding author.

E-mail address: martin.bencsik@ntu.ac.uk (M. Bencsik).

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In addition to predation, the negative impacts associated with the presence of an invasive species like *V. velutina* can be numerous, including ecosystem modification, resource competition with native species, and the introduction of new diseases (Simberloff, 2010). Aside from the obvious (to prevent invasions in the first place), the best course of action against such species is to monitor and control (or where possible, eradicate) their populations (Simberloff, 2010; Laurino et al., 2019).

Currently, hornet population tracking and control appears to be mostly limited to expensive and complex nest identification techniques (Laurino et al., 2019; Liroy et al., 2022), such as harmonic radar (Milanesio et al., 2017; Maggiora et al., 2019; Liroy et al., 2021a), and radio tracking (Kennedy et al., 2018; Kim et al., 2019), both of which have also been paired with thermal imaging (Liroy et al., 2021b), and the use of unmanned aerial vehicles (UAV) (Reynaud & Guérin-Lassous, 2016; Kim et al., 2019; Kim et al., 2022). A more simplistic approach to hornet monitoring (already employed in some countries) is the involvement of citizen science, where reports of hornet sightings can be logged by the public, promoting trapping and destruction of nests, as well as knowledge on the species' distribution (Leza et al., 2021). Identifying hornet presence in new areas early on is considered an ideal strategy for slowing the spread of the species and enabling nest destruction/population control (Robinet et al., 2017). It is suggested that involving beekeepers in this type of hornet monitoring would be advantageous because of the predator's preference for bees (Simberloff, 2010; Laurino et al., 2019). A surveillance programme has previously benefited from this strategy (Leza et al., 2021).

At present, hornet detection strategies, as far as we know, are all carried out in person, not remotely. Employing an automatic, remote hornet monitoring procedure at apiaries could therefore be advantageous for quicker species recognition and faster action to prevent their spread. Rather than relying on in-person inspections to establish hornet presence (by the time the threat has been identified, the hornets may have already been preying on colonies for hours or days), a remote system could alert a beekeeper immediately via their phone or other device. Population control measures would benefit from swift information sharing as a result of this automatic detection. Nest identification/control is considered superior to other hornet control techniques (the fitting of beehive muzzles (mesh, protective cages) to colony entrances that allow bees to pass through, but not hornets (Requier et al., 2019; Bonneford et al., 2021), or using traps or poisoned bait to control hornet numbers (Turchi & Derijard, 2018; Liroy et al., 2020; Rojas-Nossa et al., 2022) that can be employed at apiaries (Robinet et al., 2017), and as a result, beekeepers would also benefit from automatic monitoring as hornet populations would be reduced more quickly, lessening their negative impacts on colonies.

Remote monitoring techniques are already employed for a range of honeybee/apiary parameters that can be continuously measured, such as colony temperature, colony weight, and the number of bees entering and exiting the hive (Zacepins et al., 2015). This allows beekeepers to keep up to date with changes that occur in their colonies without needing to be present or involved in physical inspections (Zacepins et al., 2015). Extending the repertoire of parameters that can be monitored by these systems to predator recognition should therefore be advantageous for informing apiary management decisions and improving colony health.

Non-invasive, acoustic monitoring has recently been successfully researched at apiaries, revealing important information on bee behaviour and activities such as swarming events and queen presence (Terenzi et al., 2020; Abdollahi et al., 2022; Uthoff et al., 2023). Sound analysis has good potential for its application in hornet detection in this environment as well. Several studies have confirmed that an insects' wing-beat frequency can be used for its successful identification, with this particular parameter even being referred to as the 'fingerprint' of flying invertebrates (Moore et al., 1986; Batista et al., 2011; van Roy et al., 2014; Potamis & Rigakis, 2015; Kawakita & Ichikawa, 2019; Parmezan

et al., 2021; Parmezan et al., 2022; Herrera et al., 2023). Two types of technology have so far been used to identify the characteristics of hornet flight sound, (i) microphones, and (ii) optical sensors (which gather acoustic frequency data via lasers), both paired with machine learning and feature extraction analyses for species classification (Kawakita & Ichikawa, 2019; Herrera et al., 2023). Both studies demonstrate that hornet flight sounds can be successfully discriminated from those of other hymenopteran species and background noise, but neither have been tested at an apiary setting for remote hornet detection purposes (Kawakita & Ichikawa, 2019; Herrera et al., 2023).

In this study, we have chosen to use microphones paired with machine learning to discriminate *V. velutina* flight sounds from those of honeybees in an apiary setting. Optical sensors are considered to be more accurate than microphones as they avoid capturing background noise, providing clean data for only those signals of interest (Potamis & Rigakis, 2015). However, for our study, we have concluded that microphones are the better choice, for reasons that will now be discussed. Regarding hornet species in general, microphones have so far only been used to detect the acoustic features of *Vespa simillima xanthoptera* (yellow hornet) (Kawakita & Ichikawa, 2019), but we seem to provide the first careful study regarding *V. velutina*. Microphones are easy to maintain in an apiary environment and offer a cheaper monitoring system than those already commercialised (such as hive scales for measuring weight changes) (Zacepins et al., 2015). Additionally, microphones demonstrate advantages over the use of optical sensors when considering placement in an apiary setting, because although optical sensors can be inexpensive (Batista et al., 2011; Parmezan et al., 2021; Parmezan et al., 2022), they require insects to fly directly through the light sensor for acoustic frequency data to be captured. In previous studies, optical sensors have either been implemented in the lab, e.g., using flight tents or boxes (Batista et al., 2011; van Roy et al., 2014; Potamis & Rigakis, 2015; Kalfas et al., 2022; Chatzaki et al., 2023; Herrera et al., 2023), or placed over the colony entrances of the wasp/hornet/bee species to ensure overlap with the light beams (Parmezan et al., 2021; Parmezan et al., 2022). It has been suggested that under natural field conditions, there would be a need for attractants e.g., pheromones or protein bait, to lure hornets past the sensor (Herrera et al., 2023). Microphones, on the other hand, can easily be clipped to a colony landing board, to record audio data within the vicinity where hornets engage in hawking behaviour, without this requirement.

We here explore a remote, *V. velutina* (henceforth 'hornet' in this manuscript) monitoring system for use at apiaries that can discriminate between hornet and honeybee flight sounds. We establish that, as a consequence of hovering, the sound spectra of both species oscillate, not in magnitude (vertical axis), but in frequency (horizontal axis), although this occurs less commonly for the honeybee than for the hornet. This frequency modulation feature is here characterised in detail using two-dimensional-Fourier-transform (2DFT) images and is a component of hornet and honeybee acoustics that benefits feature extraction for discrimination of the two species. This is the first study that we are aware of, that (i) identifies and characterises the resonant frequency features of hornet and honeybee flight sounds, and (ii) utilises 2DFT imaging for hornet/bee/background discrimination. We demonstrate that this method, which is in its pilot stage, can currently reliably detect hornets at an apiary, and further discuss that this technique may have future potential as a remote 'alert-system' to inform of hornet presence in this environment.

2. Methods

2.1. Data collection

Data was collected at an apiary in Portugal between October 2022 and September 2023 (40°10'55.4"N 8°24'51.5"W). The Asian hornet arrived in this region in 2014 and numbers of reported nests have now stabilised. In the last five years, nest density has fluctuated between 1.5

and 3 nest/km². Although the city of Coimbra hosting the apiary is neither coastal or mountainous, it has the Mondego river providing the water known to promote *Vespa Velutina*. The 2023 yearly and October monthly temperatures are, respectively, mean = 16.8 °C and 18.5 °C, min = -1.2 °C and 9.4 °C, max = 41.5 °C and 33.3 °C.

To capture video and audio data for building and testing the hornet discrimination training database, a microphone (BEEP, Netherlands) was attached to the landing board of three hives (one hive at a time on different days) which connected to a camera (Panasonic DMC-FZ300). This camera was set-up on a tripod facing the hive, to capture video data of the landing board and surrounding areas. The microphone was plugged into the camera mic input slot to enable synchronous video and audio recording (see Fig. 1). Microphone data was recorded at a sampling rate of 48000 Hz and video data was captured at 25 FPS.

2.2. Building the training database

Code written specifically for this study (Matlab, 2020a), at Nottingham Trent University, was used for all analyses. The audio data from five videos was used to build the training database. The videos amounted to 85 min and 47 s, with 301 s contributing to training database creation (hornet = 88 s from 3 videos, honeybee = 112 s from three videos, winter background = 81 s from two videos, summer background = 20 s from one video). Two of the videos were filmed on 13.10.22, two on the 15.12.22, and one on the 16.05.23. Hornets were visibly and audibly present in 4/5 of these videos (intermittently in 2/5, and continuously 2/5). Honeybees were present in all five videos and regularly flew in and out of the hive throughout the three videos where hornets were seen intermittently or not at all. In the other two videos, where hornets were continuously present, honeybees were not seen to fly often and were mostly observed gathered around the hive entrance. When honeybee or hornet sounds did not occur, background noise was detected.

Each video was exhaustively, critically assessed for hornet, honeybee, and background presence, to ensure that timings were selected from the recordings that contained only the acoustic data of interest, i.e., just hornet flight, just honeybee flight, or just background noise. The number of high-quality extractions was limited to those portions of video where two signal types did not overlap, as there were time periods where, for example, hornets and honeybees could be heard flying simultaneously, or where multiple honeybees or multiple hornets could be seen and heard flying at the same time. That said, only instances where individuals were flying alone in the videos were chosen for the database, to ensure that the sounds heard could be attributed to those honeybees and hornets. All time periods in the videos that did not contain overlapping signals and were therefore clearly attributed to a single honeybee or hornet individual were included in the training database. All periods of background noise were included that contained only general background acoustics i.e., without instances of other, identifiable

sounds such as vehicle noise or bird calls. Each second of audio data that contributed to the training database was transformed into a two-dimensional-Fourier-transform (or 2DFT, see Fig. 2) with a multiplication factor (a tool for visualising the spectral details of a waveform over the x axis) of 4 and a temporal resolution of 0.04 s. These parameters were found to provide the clearest assessment of acoustic features. 2DFTs are a visual tool used to identify the repetition frequency of a waveform over a selected time-period. This repetitive element appears to be typical of insect hovering and as such, is captured by this analysis. A detailed explanation of the 2DFT process and uses is outlined in the thesis of Hall (2023).

To ensure that only the features of interest were kept in the 2DFTs, a selection of 2DFT images were assessed for frequency harmonic presence ($n = 34$ for hornet, $n = 47$ for honeybee). Originally, the bandwidth of the vertical axis of the 2DFT was 0 – 24000 Hz, but these were cropped to 1500 Hz because on closer inspection of the images, there were no frequency features of interest beyond this value. Although some harmonics beyond 1500 Hz were present in some of the honeybee 2DFTs (30 %), these were faint traces, and it was expected that the clear frequency differences seen below 1500 Hz for the two species would be the most relevant ones for discrimination. A low frequency band of 0 – 75 Hz was also cropped from the vertical axis of the 2DFT images to reduce the effect of strong background frequencies on the clarity of the 2DFT features. All 2DFTs were scaled identically (by the maximum) to remove the flight sound signal strength from the discrimination exercise.

The training database used in this study is based on several rounds of machine learning algorithm improvements, as is typical with this type of analysis. The fourth training database was the final one, and yielded the best outcome achieved so far, following improvements and the addition of extra data to promote discrimination success. Although the total time duration of our training database perhaps appears small, our work provides convincing results within the context of out pilot study, and uses an algorithm which (unlike deep learning) does not necessitate a very large training database.

It was important to define ‘winter background noise’ from ‘summer background noise’ as the two were very different, with ‘winter’ being quieter, and ‘summer’ containing loud, generic buzzing from inside the colony as well as on the landing board due to honeybees fanning at the colony entrance.

2.3. Training database analysis

Once built, the training database underwent principal component analysis (PCA) to determine which features in the database exhibited high variance, and then discriminant function analysis (DFA), where the features with the highest variance were used as a reduced dataset for supervised classification (Ramsey et al., 2018; Ramsey et al., 2020; Hall, 2022; Hall et al., 2023). From these analyses, a scatterplot and two discriminant images were created. The scatterplot demonstrates the

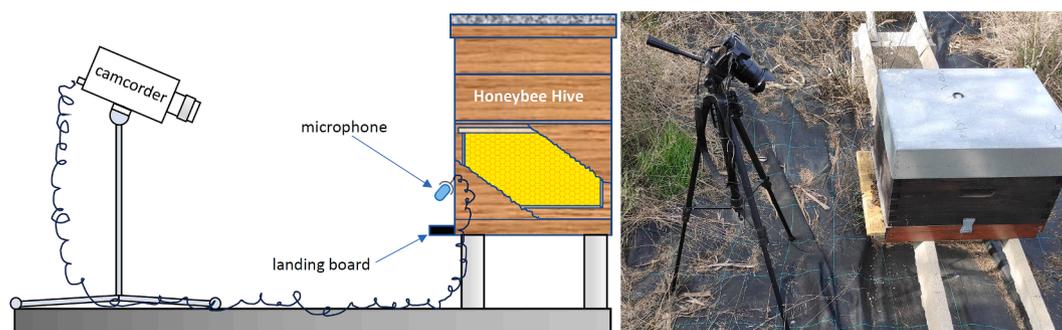


Fig. 1. Sketch and photo showing the experimental setup. The microphone at the landing board captures the flight sound of bees landing and taking off, and hornets hovering nearby. The camcorder records the signal from said microphone (instead of using its own built-in microphone) and provides video footage used to corroborate the presence, or absence of hornets.

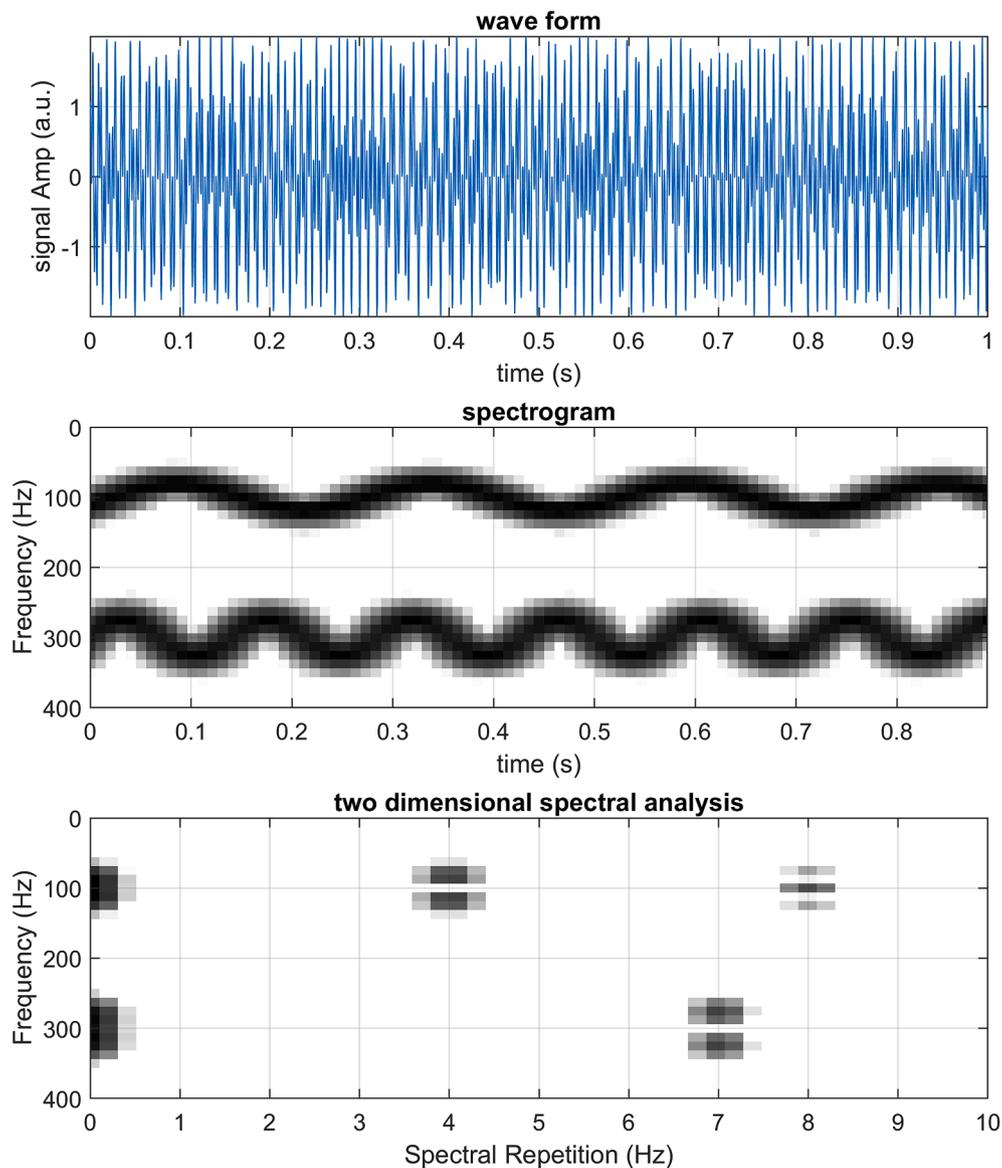


Fig. 2. Feature extraction by means of 2DFT. The figure shows the time course (**top**) of an imaginary signal, highly representative of the flight sounds that we have processed in our study. Here, instead of flight sounds, we show two sinusoidal oscillations, respectively at 100 and 300 Hz, regularly frequency-modulated respectively 4 times and 7 times per second, by ± 20 Hz. Said frequency modulation is clearly seen by displaying the stacked short-duration individual spectra of the signal in the form of a spectrogram (**middle**). The faster modulation of the 300 Hz component is clearly seen. By further computing the frequency spectrum of each horizontal line of the spectrogram, the 2DFT is obtained (**bottom**). Two bright signals are seen at vertical coordinates 100 Hz and 300 Hz, on the extreme left-hand side, as the spectrogram is made of power spectra with only positive (or zero) magnitude, necessarily resulting in a non-zero averaged spectrum at the zero spectral repetition frequency horizontal coordinate. However, the vertical coordinates 100 Hz and 300 Hz are also seen to have substantial signal at respective horizontal coordinates 4 Hz and 7 Hz, due to the frequency-modulation of the original signal. The oscillations picked up by the spectra undertaken on the horizontal lines of the spectrogram are not necessarily sinusoidal, and therefore result in smaller harmonic peaks, such as that seen for the 8 Hz spectral repetition of the 100 Hz component. Although here, and in our data, spectral repetition signals appear in the 2DFT due only to the frequency modulation of the original signals, they may also appear due to other mechanisms, such as the rapid repetition of a given short-lived signal, for instance.

clustering of each group of signals (hornet, honeybee, summer background, winter background) in DF space. The two discriminant images, transformed into 2DFTs, enabled visualisation of the feature variation that allowed the high-quality clustering between the categories of signal. Optimum clustering of the groups and clarity of the discriminant 2DFT images was achieved using 31 % of the total deviations in the dataset.

The peripheral points of each cluster in the DF space scatterplot were identified as a polygon and used to determine the boundary of each group. The areas within these boundaries are here referred to as ‘masks’ and are representative of the location that each group inhabits in DF space.

New data points from the recordings that contributed to the training database, as well as from novel recordings that did not contribute, could then be projected onto the DF space scatterplot to determine if they fell into the correct area, e.g., if hornets were present in the tested recording, we expected the corresponding datapoints to fall into the hornet mask area, if successful. This exploration allowed us to establish if independent hornet, honeybee, and background sounds could be correctly classified using the training data.

Initially, to determine if the classification algorithm was working as expected, the recordings that contributed to the training database were tested. As each recording lasted from 2 min to 28 min, they were composed of many time durations that contained hornet, honeybee, and

background sounds that did not contribute to the training data (301 out of 85 min and 47 s used for training). Specific periods of recording that contained the acoustics for each type of signal (hornet, honeybee, summer, and winter background) were identified by critical listening and video observation. These time periods were then prepared in the same way as the training data (transformed into 1-second-long 2DFTs, scaled, cropped).

Cross-correlation product analysis was then implemented between the first 1-second-long 2DFT of the selected time period of the recording and the two discriminant 2DFTs that resulted from PCA/DFA. This produced the two DF space scores necessary for projection of the data onto DF space. This analysis was then repeated on the next 1-second-long extract of the selected time period, by moving on in a step of one second. Co-ordinates were then obtained for this extract as well. This was repeated until the end of the time selection. The acquired set of co-ordinates for the entire tested period could then be plotted on the DF space to identify if the data had been correctly classified or not, dependent on the position of the projected datapoints in relation to the signal masks. By using recordings containing known sounds, correct discrimination could be established with high certainty, further supported by critical listening and visualisation of the plotted data. Of the 85 min 47 s of video, 57 min 40 s were tested.

If any datapoints of known acoustic features fell outside of the expected mask, this data could then be added to the training database for the purpose of improving the training, as is typical with machine learning techniques. As previously mentioned, the database underwent four such rounds of training improvement.

The training database was then tested for its ability to correctly classify data using recordings that did not contribute at all to its

creation. Two videos were tested, amounting to 54 min 28 s. Of this time duration, 52 min 25 s were tested, omitting periods where human noise interfered in the recording and prioritising periods where hornets were both present and absent.

Overall, including recordings that contributed to the training database and those that did not 110 min 5 s of recording were tested. Of these, 17 time periods were tested from the data that partially contributed to the training database (12/17 = hornets present, 5/17 = hornets absent) and 7 time periods were tested from the novel recordings (3/7 = hornets present, 4/7 = hornets absent).

3. Results

3.1. Characterisation of hornet and honeybee flight sounds

Hornets were typically seen to hawk for periods of several seconds to several minutes around the hive landing boards and produced uninterrupted hovering sounds, but they were also seen to fly more quickly in and out of the area in some cases, which created shorter bursts of sound (see [Video 1](#)). For the purpose of hornet flight characterisation and detection, in this study we chose to focus on the more common and acoustically prominent hovering flight sounds. Honeybees, in contrast to hornets, flew quickly in and out of the hive entrance on a regular basis and hovered less commonly (see [Video 1](#)).

Video 1: Hornet and honeybee flight sounds

A video to demonstrate hornet hovering flight (0 to 5 s), an example of brief hornet flight past the recording area (5 to 7 s), and honeybee colony enter/exit sounds (8 to 13 s).

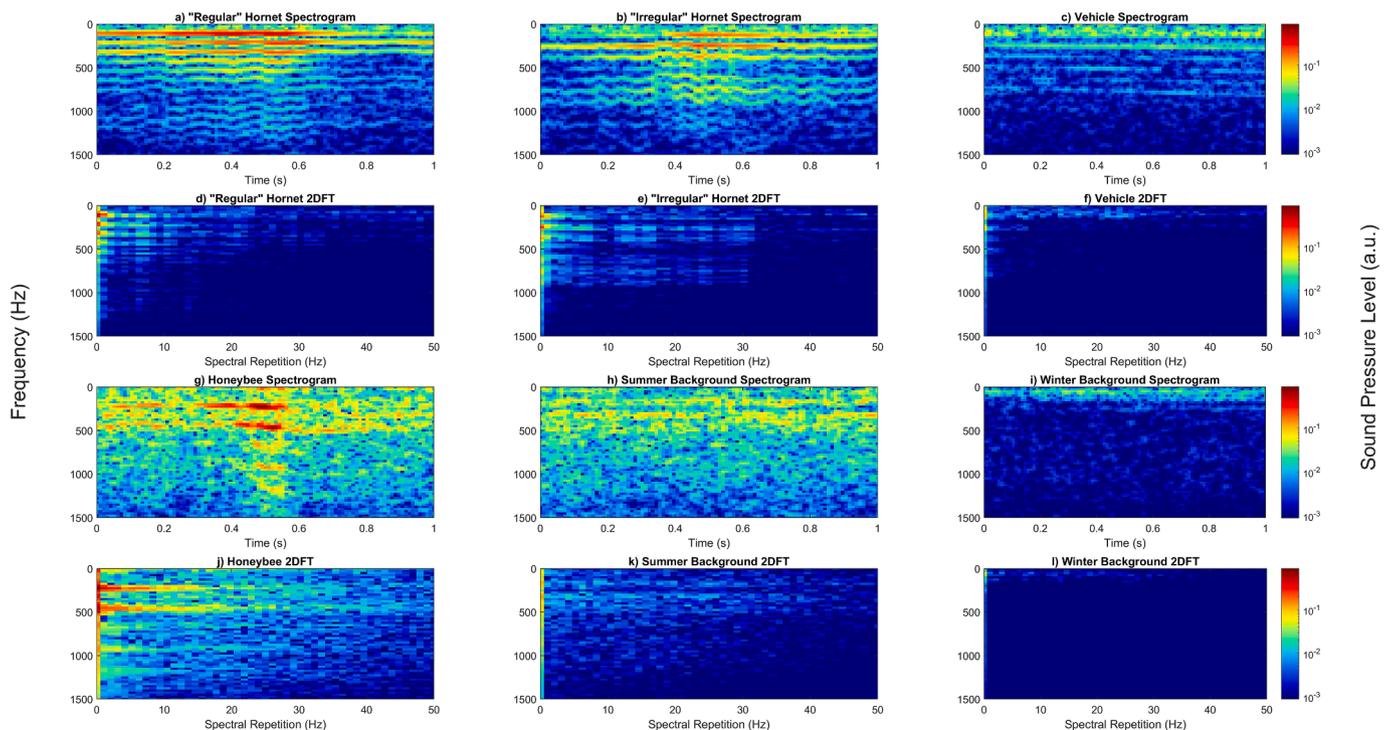


Fig. 3. Spectrogram and 2DFT images representative of each of the noise categories of interest. Representative spectrograms and 2DFT images for all acoustic extracts of interest. Each image comprises of one second of sound data for typical ('regular') hornet, honeybee, summer background, winter background (all contributing to the TDB), 'irregular' hornet (differing in acoustic features to other observed hornets, but also included in the 'hornet' category of the TDB), and vehicle noise (not included in the TDB). The 'regular' hornet (a, d), 'irregular' hornet (b, e), and vehicle (c, f) are placed side-by-side for ease of feature comparison, with honeybee (g, j), summer background (h, k), and winter background (i, l) placed side-by-side below these. Each of the six spectrogram panels (a, b, c, g, h, i) exhibit the frequency components of the sound data (y axis) over time (x axis). Each of the six 2DFT panels (d, e, f, j, k, l) exhibit both data against frequency data (y axis) and spectral repetition rate (x axis). Data has been cropped to 1500 Hz along the y axis to better showcase the details of interest. All panels are scaled identically for ease of comparison. Sound amplitude is logarithmic (a.u.) where the highest magnitude is dark red (8.8×10^{-1} a.u.) and the lowest magnitude, dark blue, is forced to be 1/1000 of the maximum.

This is the first detailed description of the acoustic features of *V. velutina nigrithorax* hornet flight sounds (see Fig. 3) in the vicinity of a honeybee hive entrance. Hovering flight frequencies in hornets were highly consistent (number of one second extracts inspected as spectrogram and 2DFT images = 50, from 8 hornet individuals), exhibiting a lowest fundamental frequency of 100 to 120 Hz and a second harmonic of 200 to 250 Hz (96 %) (see Fig. 3). The remaining 4 % lacked the 100 to 120 Hz frequency band, instead showcasing the 200 to 250 Hz band as the fundamental frequency. A third harmonic at 320 to 350 Hz was seen in 76 % of extracts, accompanied by a range of higher frequency harmonics of weaker strength (see Fig. 3). Strong harmonics beyond 250 Hz were not seen in the other 24 % of extracts.

Viewing the one second extracts as 2DFT images revealed repeating frequencies, also known as the spectral repetition rate, in the audio data along the x axis (Hz). Any vertical band appearing on the right-hand side of the 0 Hz line of the 2DFT provides a highly specific feature of any sound that regularly repeats over time, such as hornet hovering noise. There are two separate sound features that can result in a clear vertical trace on a 2DFT, (i) sound magnitude which may increase and decrease regularly, and (ii) the sound spectrum which may expand and shrink regularly. In the case of insect hovering in these results, the 2DFT feature is a consequence of the sound spectrum. Of the 50 hornet extracts, 76 % demonstrated a clear vertical trace at a non-zero spectral repetition rate. The most common repeating frequencies observed in hornet hovering flight were found between 2 and 8 Hz, with 79 % between 3 and 5 Hz (see Fig. 3). Of the 2DFTs that exhibited spectral repetition frequencies, 14 % showcased an additional, weaker trace at 9 to 16 Hz alongside the lower one.

In contrast to hornets, hovering behaviour was less common in honeybees. Occasionally, individuals hovered up to a few seconds before entering the hive, but most often they entered and exited their hives quickly, with corresponding short bursts of sound (see Fig. 3 and Supplementary Video 1). The frequency features of honeybee enter/exit sounds were highly uniform. Of 53 one-second-long extracts, 100 % exhibited a fundamental frequency of 200 to 250 Hz. A second main harmonic of 400 to 480 Hz was seen in 87 % of extracts, whereas the remaining 13 % showcased a slightly higher bandwidth of 500 to 520 Hz. All 53 honeybee extracts displayed a range of weaker, higher frequency harmonics (most commonly 600 to 650 Hz, 700 to 750 Hz, 850 to 900 Hz, 1000 Hz, 1100 Hz) (see Fig. 3). Clear vertical traces at non-zero spectral repetition rates were observed in 35 of the honeybee 2DFTs (66 %) residing between 2 and 8 Hz, occasionally (11 % of those with spectral repetition) with a higher frequency component of 9 to 11 Hz. The vertical traces found at non-zero repeating frequencies in honeybee 2DFTs were less clear than those observed for hornets (see Supplementary Fig. 1).

A single, specific hornet individual was observed to hover with a different spectral repetition rate, and frequency harmonics than seen in the other eight hornets. We here refer to this individual as an ‘irregular’ hornet, in comparison to the eight ‘regular’ hornets discussed above. For this individual, 38 one second extracts were inspected. The fundamental frequency seen was 200 to 280 Hz, with an additional strong harmonic at 350 to 380 Hz (see Fig. 3). In 34 % of extracts, a lower frequency harmonic of 50 to 120 Hz, of similar amplitude to the 200 to 280 Hz band, could be seen (see Fig. 3). Clear vertical traces at non-zero spectral repetition frequencies were seen in 76 % ($n = 29$) of the inspected extracts when viewed as 2DFTs at 4 to 8 Hz, often accompanied by a higher frequency of 17 to 21 Hz (39 % of those with spectral repetition) (see Fig. 3).

Background noise was characterised for both summer and winter recordings. Summer recordings contained loud colony buzzing/wing fanning sounds that contributed to the background soundscape, in comparison to winter recordings where the colony was quieter. No significant traces of interest were observed in any background extracts that contributed to the training database (see Fig. 3). In rare instances, vehicle noise could be heard in the background of the recordings (see

Fig. 3) (the effects of external noise on hornet discrimination and the percentage of time this was heard in recordings is discussed later).

3.2. Discrimination of hornet, honeybee, and background sounds

A training database (TDB) was created that contained 301 one-second-long audio extracts for (i) hornet hovering flight sounds (both ‘regular’ ($n = 50$ from winter recordings) and ‘irregular’ individuals ($n = 38$ from winter recordings)), (ii) honeybee flight sounds ($n = 54$ (from winter recordings), $n = 58$ (from summer recordings)), (iii) summer background noise ($n = 20$), and (iv) winter background noise ($n = 81$) (see Supplementary Fig. 2). These waveforms were transformed into 2DFT images. A simple machine learning algorithm (principal component analysis (PCA) and discriminant function analysis (DFA)) was used to search the TDB for features that exhibited high variation between the four groups. When using 31 % of all deviations, clear clustering of the hornet group in DF space was achieved (see Fig. 4). The honeybee and summer/winter background groups clustered together with heavy overlap (see Fig. 4); however, the purpose of this study was to achieve hornet detection, and as a result, the poor clustering of the non-hornet groups was inconsequential and expected to have little effect on the identification of hornet flight sounds in future tested data.

Two discriminant function images were also obtained as a result of PCA/DFA, and monitored to enable visualisation of the features that contributed to the discrimination outcome (see Fig. 4). Variation linked to the horizontal and vertical axes of DF score space can be seen in discriminant 2DFT no.1 and no.2, respectively (see Subplot b and c, Fig. 4). The colour-coding of the two images represents the level of influence that each feature had on this outcome, where dark blue is indicative of features that had little to no impact on the clustering outcome, and dark red is indicative of features that strongly impacted the discrimination outcome (see Subplot b and c, Fig. 4). Both discriminant images showcase a frequency harmonic between 150 and 200 Hz, seen in hornet flight data, but not that of honeybees, and another between 400 and 520 Hz, seen in honeybee data but not that of hornets (see Subplot b and c, Fig. 4). This demonstrates that frequency differences were a strong discriminatory feature along both axes. Spectral repetition rate also somewhat influenced the discrimination outcome. Discriminant 2DFT no.2 exhibits a trace at 9 Hz, a repeating frequency seen in some hornet and honeybee flight sounds (see Subplot b and c, Fig. 4). Both images also showcase traces at 19 Hz, indicating that the inclusion of ‘irregular’ hornet flight data benefited the discrimination outcome (see Subplot c, Fig. 4).

3.3. Classification of apiary recording data that contributed to the TDB

When establishing the accuracy of the training algorithm, it was first necessary to test new sections of the same recordings that had contributed to the building of the TDB. The full recordings ($n = 5$) from which the TDB was built ranged from 2 min to 30 min in length, and as such, not all the acoustic data within each recording was utilised ($n = 301$ s) (see Supplementary Fig. 2). This is because the TDB was built from specifically selected, clear extracts that corresponded to each category. Time periods within each recording were critically assessed and identified for hornet presence or absence, and the discrimination outcome for each was then projected onto DF space for categorisation purposes (see Fig. 5). Overall, 3460 s of data were tested, comprising of 17 separate, homogenous time periods (hornet present 12/17, hornet absent 5/17) (see Table 1).

The performance of the classification analysis was very high, with the datapoints most often falling into the correct cluster area based on the known acoustics occurring in each selected period (see Fig. 5). A negligible number of datapoints fell into the incorrect cluster area in 2/17 of the tested time periods (see Table 1). For these two periods, hornets were detected when there were no individuals present in the frames (see Table 1). Upon closer inspection of the data, i.e., through critical

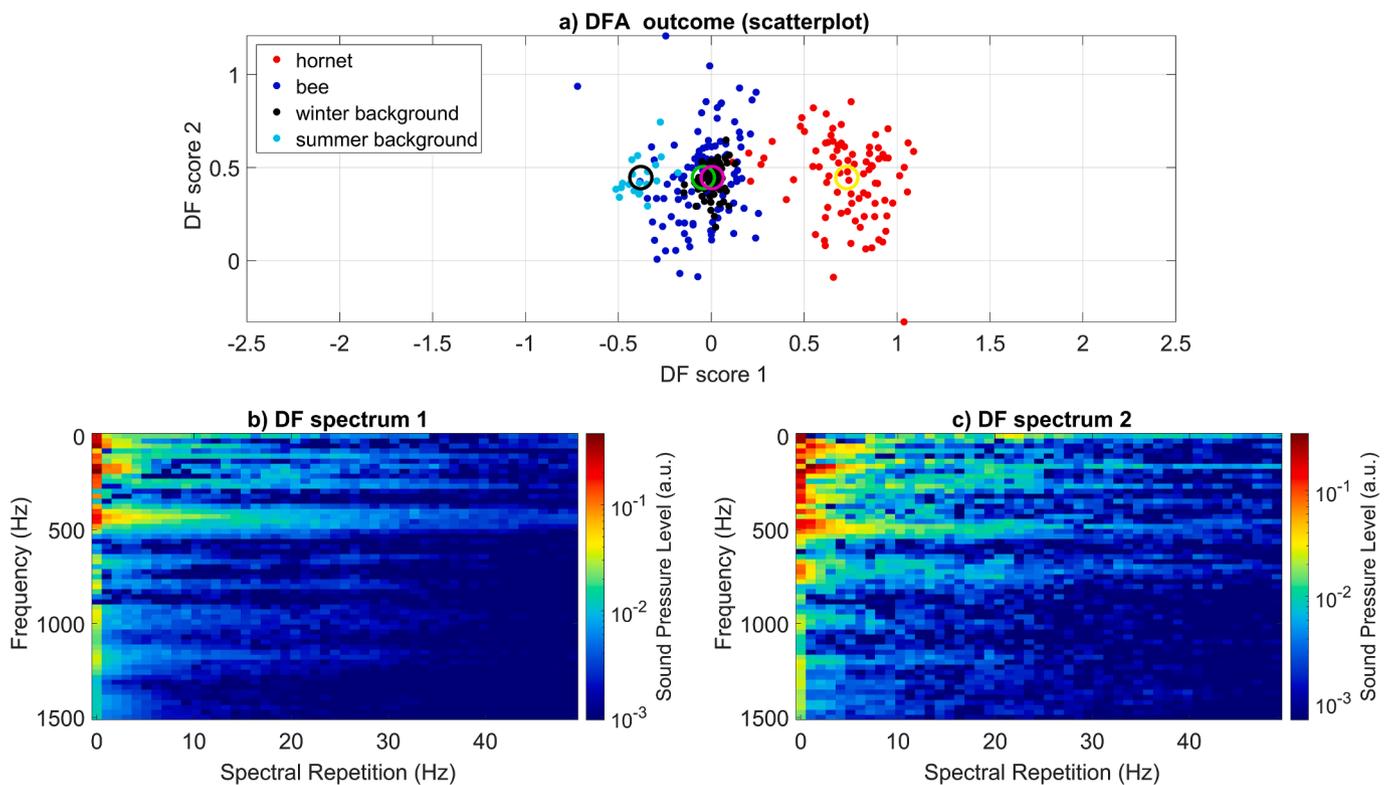


Fig. 4. The outcome of the PCA/DFA exploration. The results gathered from the PCA/DFA exploration of the training database. Panel ‘a’ exhibits the scatterplot outcome of the four groups (hornet = red points, honeybee = black points, winter background = blue points, summer background = cyan points). The centroid for each dataset is also shown to help clarify where the clusters fall in relation to one another (hornet = yellow, honeybee = green, winter background = magenta, summer background = black). Panels ‘b’ and ‘c’ show the discriminant 2DFT images for the horizontal (‘b’) and vertical (‘c’) axes of DF space. In these panels, sound amplitude is logarithmic (a.u.), where dark red represents the maximum (4.7×10^{-1} a.u.) and dark blue is the minimum (forced to be $1/500$ of the maximum). Both panels are scaled identically for ease of comparison. Areas of dark blue represent the features in the 2DFTs of the four groups that had low influence on the discrimination outcome, whereas areas of dark red are features that had the highest influence.

assessment of both the video/audio recording and the 2DFT images that corresponded to the timestamps of the specific datapoints that fell into the incorrect area, it was confirmed that 18 s related to loud vehicle noise (see Fig. 3) and 13 s corresponded to honeybee flight sounds. No behavioural differences were identified in these honeybees that could explain why they were miscategorised. The vehicle sounds exhibited two main frequency bands at 200 to 250 Hz and 300 to 380 Hz (see Fig. 3), which overlap with those of both the ‘regular’ and ‘irregular’ hornet, and may explain why the training algorithm incorrectly categorised these extracts. Overall, mis-categorisation accounted for just 0.9 % of the tested data.

Hornets were detected by the algorithm even in instances where flight sounds were audibly faint, or the hornets were only present in the recording briefly (4/17 selected time periods) (see Table 1, see Supplementary Fig. 3). The 2DFT likely detected the spectral repetition feature when hornets flew quickly through the recording area because even when flying rapidly (flight sounds of less than 1 s), the repeating frequency elements could still be observed in the spectrograms (see Supplementary Fig. 3).

3.4. Classification of apiary recording data that did not contribute to the TDB

Following the success of the algorithm using known data, recordings that did not contribute to the TDB were then further tested. Two videos (each approximately 30 min in length) were critically (visually and acoustically) assessed in the same way as previous recordings to determine periods of time that did or did not contain hornet presence. Seven specific, homogenous periods were selected, amounting to 3145 s.

Hornets were present in three of the selected time periods and absent in the other four. The data were correctly classified in 4/7 of the periods (see Table 2). In the remaining three, hornets were detected when none were present. Following the same assessments as recordings that contributed to the TDB, it was determined that the mis-categorised datapoints related to loud vehicle and human related noise (41 s, 1.3 % of the tested data).

As with the recordings that partially contributed to TDB creation, in the novel recordings, the training algorithm still correctly detected the presence of hornets that were audibly faint or flew loudly, but briefly past the microphone (3/7 of the selected time period (see Table 2, see Supplementary Fig. 3).

4. Discussion

Excellent *V. velutina* discrimination has been achieved using the training database and algorithm in this study. When testing data that partially contributed to the database, hornet presence/absence was correctly identified in 99.1 % of the recordings. When testing novel data, the accuracy was 98.7 %.

4.1. The advantages of utilising 2DFT analysis for species discrimination

Both our discrimination outcome and exploration into the features of *V. velutina nigrithorax* flight sounds benefitted from 2DFT analysis. Prior to this investigation, Japanese yellow hornet (*Vespa simillima xanthoptera*) flight sounds had been characterised in terms of frequency and amplitude (Kawakita & Ichikawa, 2019), and the species in question, *V. velutina nigrithorax*, only had the fundamental frequency of its

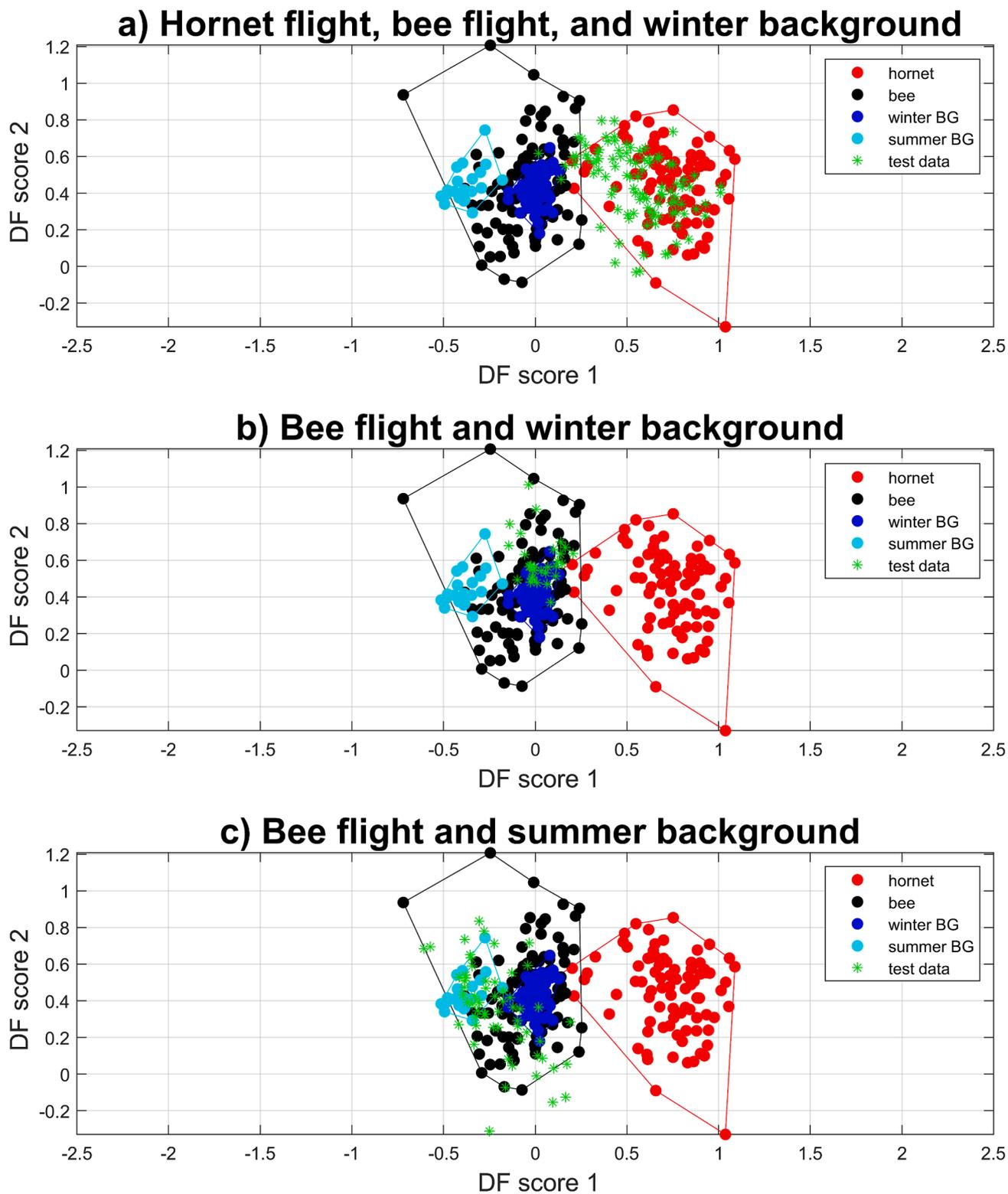


Fig. 5. The outcome of the training algorithm testing phase. This figure shows the mask areas for each of the four categories and the outcome of projecting data onto the same DF space plot. Each panel shows the scatterplot that is the outcome of PCA and DFA exploration (hornet = red line and red points, honeybee = black line and black points, winter background = blue line and blue points, summer background = cyan line and cyan points). The peripheral datapoints for each cluster area were used to define the masks. Data taken from recordings that contributed to the training database is projected onto the scatterplot to demonstrate the success of the algorithm in discriminating hornet, honeybee, and background as the projected points fall into the same area of DF space as the corresponding mask (this projected data is shown as green stars, each star is representative of 1 second of data). Panel 'a' shows 90 seconds of projected data where hornets were present alongside some honeybee activity and background noise. Panel 'b' shows 30 seconds of projected data where honeybee activity occurred during the winter period. Panel 'c' shows 60 seconds of projected data where honeybee activity occurred during the summer period, with background noise containing colony buzzing and fanning activity.

Table 1
Tested time periods for data that partially contributed to the TDB.

Tested Time Period (seconds)	Hornets present in recording?	Hornets detected by algorithm?	Matching outcome?
Video 1.1 (60)	Yes (multiple, loud)	Yes	✓
Video 1.2 (110)	Yes (multiple, loud)	Yes	✓
Video 2 (100)	Yes (multiple, loud)	Yes	✓
Video 3.1 (80)	Yes (singular, faint/brief)	Yes	✓
Video 3.2 (180)	No	Yes	×
Video 3.3 (100)	Yes (singular, faint/brief)	Yes	✓
Video 3.4 (50)	No	No	✓
Video 3.5 (650)	Yes (multiple, loud)	Yes	✓
Video 3.6 (100)	Yes (singular, faint/brief)	Yes	✓
Video 4.1 (89)	Yes (multiple, loud)	Yes	✓
Video 4.2 (37)	Yes (singular, faint/brief)	Yes	✓
Video 4.3 (110)	Yes (singular, loud)	Yes	✓
Video 4.4 (30)	No	No	✓
Video 4.5 (30)	No	No	✓
Video 4.6 (30)	No	No	✓
Video 4.7 (50)	Yes (singular, loud)	Yes	✓
Video 5 (1654)	No	Yes	×

A table to demonstrate the number of time periods that were tested using the training algorithm per recording that partially contributed to the TDB. Whether hornets were present or absent in each time period, and if the algorithm correctly categorised the data, is listed here. The table highlights whether hornets were present in multiple or singular numbers, as well as whether the hornet flight sounds were audibly loud, faint, or brief.

Table 2
Tested time periods for data that did not contribute to the TDB.

Tested Time Period (seconds)	Hornets present in recording?	Hornets detected by algorithm?	Matching outcome?
Video 1.1 (45)	No	No	✓
Video 1.2 (55)	Yes (singular, faint/brief)	Yes	✓
Video 1.3 (163)	Yes (singular, faint/brief)	Yes	✓
Video 1.4 (115)	Yes (singular, faint/brief)	Yes	✓
Video 1.5 (310)	No	Yes	×
Video 1.6 (890)	No	Yes	×
Video 2 (1567)	No	Yes	×

A table to demonstrate the number of time periods that were tested using the training algorithm per novel recording. Whether hornets were present or absent in each time period, and if the algorithm correctly categorised the data, is listed here. The table highlights whether hornets were present in multiple or singular numbers, as well as whether the hornet flight sounds were audibly loud, faint, or brief.

wingbeat pattern described (Herrera et al., 2023). In our study, the 2DFT revealed the additional feature of spectral repetition frequency, which can be observed in any waveform that regularly repeats over time. This explorative exercise has determined that the hovering flight of a hornet results in a systematically recurring waveform.

Our results have additionally shown that in 7 of the tested time periods (novel and known), hornets were detected even if their flight sounds were quiet or very brief. This is the same for the honeybee flight

sounds that were attained in our study. Even though the majority of honeybee enter/exit traces had a short time duration, the 2DFT still detected the elements of the sounds that repeated (albeit with less clarity than for consistent hornet hovering). These findings highlight the strength of the 2DFT as a feature in determining this specific, repeating acoustic feature; even without prolonged or strong hovering flight traces in the analysed time window, the 2DFT can establish the repeatability of a waveform. This has also previously been demonstrated in our study investigating Varroa gait patterns (Hall et al., 2023). Not all Varroa walking vibrational traces were detected by vibration sensors, yet the 2DFT successfully determined the repeating elements by ‘filling in the data blanks’ (Hall et al., 2023).

As an additional bonus of using the 2DFT, the discriminant 2DFT images in our study emphasise the importance of repeating frequency components in the machine learning exercise for discriminating between hornet, honeybee, and background. Despite both hornet and honeybee exhibiting similar spectral repetition frequencies, there were small, observable differences between the two species that have clearly been detected by the algorithm for clustering purposes. Hornet and honeybee spectral repetition traces overlapped in frequency at 2 to 8 Hz, yet for hornets these most prominently occurred at 3 to 4 Hz and above. A more apparent feature was the presence of a second repeating component in some hornet 2DFTs, at a higher frequency than that of honeybees (9 to 16 Hz (‘regular’ hornet), 17 to 21 Hz (‘irregular’ hornet), compared to 9 to 11 Hz (honeybee)). Alongside the strong frequency harmonic differences between the two species, these diminutive features also played a role in identifying variation between the groups.

This result additionally emphasises the value of including the ‘irregular’ hornet in the training data. This individual exhibited the highest spectral repetition rate (17 to 21 Hz), featured in the discriminant 2DFT images (at approximately 19 Hz). This benefitted the discrimination of hornets from honeybees and furthermore did not adversely affect the detection of any hornet during the algorithm testing phase (individuals were correctly detected regardless of ‘regular’ or ‘irregular’ features). The fact that the two hornet categories formed a single, clear cluster in DF space (see Fig. 5) further strengthens the claim that the training algorithm can discriminate hornets from other noise without affecting hornet classification.

The inclusion of both hornet ‘types’ will benefit detection at apiaries, regarding those whose wingbeat frequencies do not conform to the ‘norm’. *V. velutina* individuals vary in body size and weight, as well as wing size and shape, dependent on caste, gender, season, and individual differences (Monceau et al., 2013; Pérez-de-Heredia et al., 2017). Size, body mass, and wing morphology are factors known to affect wingbeat frequency and natural frequency in insects (San Ha et al., 2013). Although we could not visibly identify any specific morphological differences between the hornets in our recordings (we did not catch the hornets seen at the apiary to assess the effects of these variables), it is likely that the detected acoustic properties varied based upon individual size, weight, and wing characteristics. It is also possible that environmental factors affected the frequencies of hornet flight, as hymenopteran flight performance, speed, and frequency can be influenced by abiotic conditions such as temperature, humidity, and illuminance (Spiewok & Schmolz, 2006; Parmezan et al., 2021). This may also explain the differences observed in the small number ($n = 13$) of honeybee flight sounds that were mis-categorised as hornets in our study – these variables may have influenced honeybee acoustics and caused variation that more closely resembled that of a hornet. However, it is worth noting that hornets were correctly identified in both summer and winter recordings, despite the hornet category in the training database being built only from winter recordings. This demonstrates that hornet flight sounds can be successfully detected by our algorithm regardless of potential seasonal/temperature effects, and implies that the ‘irregular’ hornet flight sounds may have differed due to morphological rather than abiotic factors. Overall, our training algorithm has demonstrated strong robustness when faced with acoustic variation, which may have been

affected by morphological and environmental variables. However, in future long-term recordings, previously un-encountered ‘irregular’ hornet individuals may be present, which could affect the success of the algorithm (should these new individuals have wing-beat frequencies that vary in comparison to those already included in the training database). Under such circumstances, the training database would benefit from being updated to include instances of new hornet hovering sounds, or instances of honeybee and/or hornet spectral changes due to local variations in the species, resulting in moderately different flight sound frequency spectra.

4.2. The advantages of microphone implementation for hornet data capture at an apiary

As highlighted above, the training algorithm demonstrated strength in its discrimination ability and detection of specific acoustic features for hornet identification. Using a microphone to capture acoustic data has also benefited the results in our study (advantages discussed below), supporting our choice to use this form of recording equipment over the alternative optical sensor approach, which is the only other technology that has so far been employed to capture hornet flight sounds (Kawakita & Ichikawa, 2019; Herrera et al., 2023). Other methods of measuring wing-beat frequency are the use of a stroboscope or a radar, but these were unsuitable for the purpose of our study as they require flying insects to be caught and tethered, or fitted with measuring devices (Wang et al., 2017; Long et al., 2020).

Microphones may be considered less accurate than optical sensors, as they capture many external sounds alongside those of interest, whereas the second type of sensors are free from such interference (Potamis & Rigakis, 2015). In our study, vehicle sounds were captured, as was bird song, dog barking, and human noise. However, as a result of our successful training algorithm, the percentage of external noises that actually interfered with hornet classification was extremely low, and only in the form of some vehicle and human noise (0.9 % of recordings that contributed to TDB creation; 1.3 % of novel recordings). These results are similar to those of a study that successfully detected and classified *V. similima xanthoptera* using microphones and machine learning techniques (Kawakita & Ichikawa, 2019). It is also worth noting that a study by Potamis & Rigakis (2015), compared optical sensor and microphone recordings of insect wing flapping and found almost identical results, determining that acoustic feature clarity is not adversely affected by microphone use. In our study, hornet flight noises commonly benefited from high signal-to-noise ratio, but as mentioned above, we have also determined that even when faint or brief, their flight sounds were detectable amongst background noises. Although our measurements were acquired in an apiary where background noise is relatively strong, including even traffic noise, we have not quantitated the actual threshold of external noise level beyond which the method will start failing, and it is possible that hives exposed to strong anthropogenic noise cannot benefit from our monitoring technique.

Microphones are also free from some of the constraints potentially faced by optical sensors, when used in the field. The microphone in our study captured hornet flight sounds without interfering with the flight paths of individuals. Optical sensors require insects to fly through the laser beams, and it has been noted that for hornets under natural conditions, a lure would be needed to manipulate their movements via the sensor area to capture frequency information (Herrera et al., 2023). Our results were also not affected by the heavy traffic of honeybee individuals coming and going from the hive, or the presence of multiple hornets, whereas optical sensor data capture can be negatively affected by the presence of simultaneously occurring signals (Parmezan et al., 2021). Optical sensors are also heavily influenced by changes in illuminance, requiring them to be protected from external light sources (Parmezan et al., 2021; van Roy et al., 2014). In a field setting, where sunlight levels are changeable, it would be difficult to avoid negative impacts on data capture, whereas microphones are free from this. One

study has, however, demonstrated optical sensor wingbeat detection success under various light conditions (Potamis & Rigakis, 2015).

4.3. Future utilisation of the training algorithm and experimental set-up over the long-term at apiaries

This study has identified the advantages of a training algorithm that includes both the frequency and spectral repetition characteristics of insect flight sounds, as well as the benefits of using inexpensive microphones.

It is important to note that this is a preliminary study and therefore the algorithm is presently not expected to be fully functional on any future data set. The training database for this study using the video data that has been captured so far, is exhaustive in capturing all categories of sound. Within ‘deep learning approaches’, the size of the database is paramount to the quality of the machine learning obtained. In our work, we simply require the full extent of the deviations within each category to be captured in the training data, and that each category of the database be comprised of elements that have generic features, to promote correct feature identification in further assessment of the data.

It is now timely to test this experimental design at an apiary over the long-term. Future investigations that use such novel data to test the robustness of the algorithm will undoubtedly lead to more improvements being made.

For this purpose, we have already recorded continuous, remote microphone measurements at the apiary over a 3-month period, additionally capturing web-cam footage of the landing board area at a rate of 1-frame-per-second, for the purpose of corroborating audio data detections with visual evidence of hornet presence/absence. Whilst it was not within the scope of this study to investigate the success of the training algorithm on this long-term dataset, the measurements are now available for a follow-up investigation.

Verifying the audio data with the web-cam footage will also allow us to further establish the success of the set-up, as well as identify and account for the effects (if any) of variables such as weather conditions and colony activities on the captured acoustic data, e.g., in this study we observed audible differences between winter and summer colony recordings, related to wing fanning activity.

This future exploration will additionally determine how often external noise affects the classification outcome. In the current study, only 0.9 % of data that partially contributed to the creation of the training database and 1.3 % of novel tested data corresponded to mis-categorised sounds, amounting to just over one minute of the overall tested recorded time. Prior to the fourth and final training database, vehicle noise was included as a separate category to explore if the machine learning algorithm could discriminate it from hornet sounds. Attempts were unsuccessful, but as mis-categorised vehicle noise was not often encountered, it was deemed to be unproblematic for the current training algorithm. Carrying out analysis on the long-term data will confirm whether this remains the case, or if further training is required to reduce the impact of external noises on detection success.

4.4. Conclusion

We have here explored a remote *V. velutina* monitoring system under natural field conditions, to promote the early detection of this invasive predator at apiaries. Implementing a simple microphone set-up to capture acoustics has shown to be highly successful in recording the flight sounds of both hornets and honeybees for automated species detection and discrimination purposes. We have shown that our algorithm can detect hornet presence even when there is only one individual in the vicinity of a hive. Previous studies have only characterised hornet frequency and amplitude flight features, whereas we have emphasised the benefits of pairing these acoustic elements with spectral repetition frequency, to produce an enhanced discrimination outcome. The 2DFT has previously only been employed by our research group for specific

honeybee and Varroa mite (*Varroa destructor*) vibrational pulses (Ramsey et al., 2018; Ramsey et al., 2020; Hall, 2022; Hall et al., 2023). Our current study has revealed an additional application for this analysis technique in the field of bioacoustics, where we show that it can be used to detect a hovering flying insect. Our successful training algorithm now requires testing over the long-term, but shows remarkably good promise for remote, continuous detection of *V. velutina*.

Data availability statement

All relevant data are found in the paper and its mentary material. Additional raw data in the form of microphone recordings that were used in the analysis for this study can be found on Figshare and all relevant Matlab code on Github.

Recordings used to build and test the training base: <https://doi.org/10.6084/m9.figshare.24975117>; <https://doi.org/10.6084/m9.figshare.24975174supple>; <https://doi.org/10.6084/m9.figshare.24975180>; <https://doi.org/10.6084/m9.figshare.24975195>; <https://doi.org/10.6084/m9.figshare.24975213>.

Novel recordings used to test the training database: <https://doi.org/10.6084/m9.figshare.24975219>; <https://doi.org/10.6084/m9.figshare.24975231>.

All relevant Matlab code: https://github.com/HThomasntu/hornet_TDB_MLA_matlab.git.

CRediT authorship contribution statement

Harriet Hall: Writing – original draft, Software, Methodology, Investigation, Formal analysis, Data curation. **Martin Bencsik:** Writing – review & editing, Supervision, Software, Conceptualization. **Nuno Capela:** Methodology, Data curation. **José Paulo Sousa:** Supervision. **Dirk C. de Graaf:** Writing – review & editing, Project administration, Funding acquisition, Conceptualization.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.compae.2025.110307>.

Data availability

Our data and code has been made available as DOI links that are included in the data availability statement of the manuscript

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