Bark beetles as lidar targets and prospects of photonic surveillance

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Abstract
Forestry is raising concern about the outbreaks of European spruce bark beetle, *Ips typographus*, causing extensive damage to the spruce forest and timber values. Precise monitoring of these beetles is a necessary step towards preventing outbreaks. Current commercial monitoring methods are catch-based and lack in both temporal and spatial resolution. In this work, light scattering from beetles is characterized, and the feasibility of entomological lidar as a tool for long-term monitoring of bark beetles is explored. Laboratory optical properties, wing thickness, and wingbeat frequency of bark beetles are reported, and these parameters can infer target identity in lidar data. Lidar results from a Swedish forest with controlled bark beetle release event are presented. We concluded that our entomological lidar is a promising tool for remote monitoring bark beetles.

KEYWORDS
bark beetle, coherent scattering, entomological lidar, environmental monitoring, *Ips typographus*, target characterization, thin films

Abbreviations: DoLP, degree of linear polarization; FoV, field of view; IQR, interquartile range; NIR, near infrared; OCS, optical cross-section; SWIR, short-wave infrared; WBF, wing beat frequency; WIP, wing interference pattern.

Precise monitoring of European spruce bark beetle *Ips typographus* is necessary for fighting bark beetle infestation early. In this work optical properties wing thickness and WBF of bark beetles are reported and these parameters infer target identity in lidar data. Lidar results from a Swedish forest with a controlled bark beetle release event are presented. We concluded that our entomological lidar is a promising tool for remote monitoring bark beetles.
1 | INTRODUCTION

The spruce bark beetle (Ips typographus) is one of the most severe insect pests in European forests, capable of killing millions of spruce trees in large outbreaks [1]. During normal conditions, the adult insects mainly attack trees that are already dead. Storm damage or severe drought stress, however, lead to ample availability of brood material for the beetles. Rapid population increases have then been observed, resulting in outbreaks during which healthy trees are attacked and killed [2]. Spruce bark beetles communicate via pheromones to form aggregated attacks, which overcome the defense capacity of living trees [3]. Whereas most attacks occur within a distance of 500 m from a previous attack [4], the spruce bark beetle can disperse several kilometers [5]. Both the flight activity and the development from egg to mature bark beetle are temperature-dependent. The flight mainly occurs during days with a temperature above 16–20°C [6]. A new generation of bark beetles is initiated by overwintering adults in May and June, and maturity is reached in July to August. In central Europe and Denmark, the first generation commonly initiates a second generation peaking during late summer [7]. In Sweden, this has been rare historically, but the likelihood of two generations per year increases due to climate change [8] and it has been observed that there is a second generation emerging beetles in the south of Sweden in July [9].

A common countermeasure against bark beetles outbreaks is the timely sanitary cutting of recently attacked trees. Pheromone traps are used for monitoring flying beetles, and when there are a large number of beetles in the trap, it indicates a high risk of attacks. Traps are usually emptied weekly during the swarming season. The Swedish Forest Agency (Skogsstyrelsen), in 2020, continuously operates traps in 60 locations throughout the country [10]. Even though the traps are emptied weekly, it is still laborious to collect the trap catches, and the trap catches are therefore also limited in time resolution. They do not show the population dispersal and cannot be used to infer how the weather changes throughout the week affect bark beetle activity. The catch efficiency is also highly location-dependent, and trapped rotten insects can deter the beetles away from traps [11].

The efficiency of outbreak control depends on knowing the time, location, and magnitude of infestations. Therefore, surveillance of the population density of beetles is crucial. Several methods have been used by researchers to access the population dynamic of insects, such as e-traps [12, 13], entomological radar [14, 15], and lidar [16–22]. Existing aerial topographic lidar has primarily been used for damage assessment [23]. E-trap provides a limited assessment of insects fluxes and no dispersal estimation, and radar is unable to monitor untagged insects imbedded in the forest due to ground clutter. To overcome the limitations of those photonic surveillance methods, our group developed a kHz entomological lidar for non-intrusive remote sensing [17, 19, 22, 24, 25]. It provides optical oscillatory and microstructure information of the in-flight target, based on the retrieved spectral and polarization information.

This study aims to evaluate the feasibility of entomological lidar as a tool for long-term monitoring of bark beetles. For this reason, we characterized ex-vivo bark beetle with hyperspectral imaging and goniometry, and we recorded in-vivo dynamic scattering of beetles in a flight chamber. We investigated the relationship between the acquired optical properties and lidar measurement in-situ. In addition, we demonstrated the ability of lidar in visualizing and monitoring an otherwise unperceivable plume of pheromones together with insect activity simultaneously. In summary, this study provides improved knowledge for surveillance and identification of bark beetles. We concluded that our entomological lidar is a promising tool for remote monitoring of bark beetle population density, allowing for fighting bark beetle infestation early.

2 | MATERIALS AND METHODS

Several laboratory instruments were used to characterize bark beetles optically. Hyperspectral push-broom imaging was carried out to assess spectral features from the body, elytra, and wings. An imaging polarimetric goniometer system was used to study the scattering cross-section and de-polarization of anatomical features of beetles from various aspects of observation. A dual-band polarimetric setup was used to investigate the dynamic scattering properties and wingbeat frequency (WBF) of free-flying beetles in a flight chamber. Finally, the laboratory recordings were compared to in-field lidar measurements of released beetles.

2.1 | Field collection

Beetles were captured by using pheromone traps [26] during a lidar in-field campaign in April and May 2019 in the vicinity of Nytebodskagen, Sweden (56°20′04.0″N 14°22′59.3″E). Beetles were preserved by freezing or drying. For the mounting purpose, specimens were re-moisture, and the softened beetles were pinned vertically through the thorax with small stainless steel insect pins (size 100 μm). The elytra were opened up, and the hind wings extended and positioned horizontally to the body.
The body sizes and wingspans of 30 randomly selected beetles were measured with a caliper.

2.2 Hyperspectral imaging (ex-vivo)

We carried out hyperspectral push-broom scans with an instrument similar to previous studies [27]. The source was a halogen-tungsten lamp, which delivered light from a Ø8 mm fiber bundle from an 80 mm distance (5.7° light cone). The light source and camera were arranged in a specular condition of ±22° to the bark beetle wing surface. The specimens were mounted on a black neoprene sheet with pins to minimize the background reflectance. The hyperspectral camera is based on a visible extended InGaAs imager (Hyspex, Norsk Elektro Optikk, Norway) with a spectral coverage of 600 nm to 1600 nm filter settings in this study. The camera was operated with a microscope objective yielding a swath width of 40 mm with a resolution of 62.5 μm per pixel and 3 nm per spectral bands. The objective had an aperture of Ø 16 mm and a working distance of 80 mm, and the collected light cone was 11°. The recorded hyperspectral images were calibrated to diffuse reflectance using a Lambertian gray reference with a 50% diffuse reflectance (Spectralon®). Note that the objective only captures 3.0% of the Lambertian distribution, calibrate a reflected signal from a specular surface to a diffuse reference could produce up to ~3400% times stronger reflectance signal than from a diffuse object.

2.3 Imaging Polarimetric Goniometry (ex-vivo)

Studies have shown that the body and wings of insects display distinct degrees of linear polarization (DoLP) from various aspects and scattering angles [28–30]. We use the definition of DoLP:

$$\text{DoLP} = \frac{I_{co}}{I_{co} + I_{de}}, \quad (1)$$

where $I_{co}$ is the intensity of measured co-polarized reflected light and $I_{de}$ is the intensity of de-polarized reflected light (the dark current and background are assumed subtracted).

A spectral polarimetric optical tomographic imaging goniometer system (SPOTIG, see details in [30]) was used to study the DoLP contributed from anatomical features of the beetles. Such a system has been previously used in 3D reconstruction of insects [31, 32], and a similar technique has been used for optical tomography with a light sheet [33]. SPOTIG uses an LED as the light source (680 mW, 810 nm), and the light is horizontally polarized and collimated into a Ø 25 mm beam. A sample is mounted on another rotational stage in order to investigate different projection aspects, and the light source can be rotated around the sample to change the scattering angle. The light scattered from the sample is collected by a horizontal microscope (f100mm, Ø25mm, WD 20 cm). A linear polarization analyzer on a third rotation stage allows retrieval of both co- and de-polarization measurements (HH and HV, respectively). The camera used is a 12 bit, USB3 CMOS imager, with 1240 × 980, 4.8 × 4.8 μm pixels (Basler Ace aC2500-60 um USB3 Mono). The strobe modulated light source for automatic background subtraction.

Backscattering is recorded by placing the light source on the same side of the sample as the camera. Forward scattering/extinction can be recorded by rotating the light source to the opposite side of the sample. Flat-field calibration is done by setting the system in different illumination configurations and acquire an image of a homogeneous calibration target. The backscattering ($\theta_{sc} = 165°$) and forward scattering ($\theta_{sc} = 14°$) properties of beetles were studied in all anatomical planes, which was done by mounting each specimen in the corresponding plane and rotating the aspect stage 360° in steps of 5° (aspect angle is presented by $\varphi$). The same procedure was repeated for a beetle specimen with removed wings and specimens with removed wings and elytra in co- and de-polarization. In extinction mode ($\theta_{sc} = 0°$), the same experimental procedure as in backscattering mode was used.

The scattering phase function of beetles was also studied. Each specimen was mounted on the aspect stage, and the light source was rotated around the specimen to scan the scatter angle between $-164° < \theta_{sc} < -14°$ and $14° < \theta_{sc} < 164°$ with a step size of 5°. The same procedure was repeated for the beetle specimen without wings and specimen without wings and elytra. The phase function measurement was done in both co- and de-polarized modes.

2.4 Entomological flight chamber measurement (in-vivo)

Dual-band backscattering properties of beetles in free flight were studied in our entomological chamber, see details of the setup in [29]. A dichroic beamsplitter was used to combine an 808 nm NIR laser beam (5 W) and a 1550 nm SWIR laser beam (3 W). The superimposed NIR/SWIR beam was horizontally polarized and collimated with an Ø 50.8 mm lens. Detection optics (another
(ø 50.8 mm lens) was placed adjacent to the light source, and the field of view (FoV) of the detector was overlapped with the NIR/SWIR laser beam to form the probe volume. A long-pass filter (RG780) was used to remove ambient LED light. A polarizing beamsplitter was used to separate the received backscattered light into co- and de-polarized components. Each polarization was measured by a Si/InGaAs sandwiched photodiode (K3413-09, Hamamatsu, Japan), with a trans-impedance amplifier (TIA, OPA404) with 47 MΩ transimpedance and ~ 4 kHz bandwidth. Both NIR/SWIR laser beam and detection optics FoV were terminated in separate dark cones of neoprene. An enclosed chamber was built around the probe volume without blocking the beam or detectors [29]. White diffuse (Lambertian 100%) Teflon balls (ø 6.35 mm) was dropped through the probe volume to calibrate the backscattered light signals in all 4 channels from volts to optical cross-sections (mm²) [18, 34, 35]. Beetles were released inside the chamber, and the modulated signals were measured with a sampling rate of 20 kHz in both wavelength- and polarization bands (DAQ-USB6012, National Instruments, USA).

2.5 | Lidar at field site measurement (in-vivo and in-situ)

A lidar in-field campaign was undertaken in Nytebodaskogen, Sweden (56°20'04.0"N 14°22'59.3"E) on May 13th - 17th, 2019, see Figure 1. The lidar system was set up on a small hill next to a barn. The lidar transect was about 120 meters long and 4 meters above the ground. The transect stretched over a clearing in a valley and terminated on a termination board covered with black neoprene (1.8% diffuse reflectance at wavelength 808 nm). A pheromone trap was positioned at mid-distance of the lidar transect and right underneath the lidar beam to lure the bark beetles, and a weather station was set next to the pheromone trap. The lidar was running 24 hours every day except the times when hard drives were replaced to enable gathering more data. On May 16th, 2019, we carried out a controlled bark beetle release event and a smoke release event separately right next to the pheromone trap. The smoke is a chemical mixture of hydrochloric acid and ammonia. The release was used to visualize the movement of pheromone plume from the trap, and the controlled bark beetle released was to obtain reference measurement of bark beetles in-situ.

We used a kHz entomological Scheimpflug lidar at 808 nm, resembling previous work [36, 37]. It was a single band lidar system, and it transmitted a 3.2 W laser beam. The beam was expanded and collimated by a refractor telescope (ø 76 mm, f = 200 mm). The backscattered light was collected by a Newtonian telescope (ø 205 mm, f = 800 mm) and focused onto a CMOS sensor array with 2048 pixels and 16-bit dynamic count. An RG780 long-pass filter and a bandpass filter (808 nm, FWHM: 3 nm) were used to block background light. The baseline of expander and collector are separated by 814 mm, and the CMOS array was tilted 45° in Scheimpflug configuration to achieve infinite focal depth [17, 18, 22, 35–38]. The laser and the CMOS camera were connected to a multiplexer and a laser driver, and the laser was controlled by a strobe signal sent from the CMOS sensor to switch on and off alternately, the sensor line rate was 3.5 kHz, and the sampling rate for both backscattered signal and background were 1.75 kHz [37]. Each acquired data file is 10 seconds long and contains 35 000 lines exposures.

3 | RESULTS AND DISCUSSION

Here, we report on a successful characterization of the spectral property of bark beetles from both in-vivo and ex-vivo experiments. Using the hyperspectral camera, we studied membrane thickness and its variance within and between individuals. Soap-bubble color patterns found on beetle wings in Figure 2 are caused by the thin-film effect, a typical example of coherent and incoherent light interference. Cross-section and polarization of scattered light from beetles were studied with goniometer from various observation angles. Several beetles were released

![FIGURE 1](image-url) Aerial view of the lidar site in Nyteboda. The entomological lidar was set on a small hill, and it was monitoring over a small clearing in a valley. The lidar beam was terminated by a black neoprene board. A pheromone trap and a weather station were placed halfway of the lidar transect path.
in the dual-band polarimetric flight chamber to determine the WBF and dynamic properties of flying beetles. When laser light interacts with free-flying bark beetles, the recorded signal can be decomposed into a non-oscillatory and oscillatory scatter contribution [21, 22, 24, 25]. The oscillatory signal is contributed from the wing throughout many wingbeats, and the non-oscillatory scattered signal is contributed from the body and elytra. Both frequency components of the signals can be further decomposed into coherent ($I_{co} - I_{de}$) and incoherent scattering ($I_{de}$). The light interaction with the body and elytra is primarily diffuse [39], with the dominant chromophores being melanin acting as a gain factor on the reflectance ($-\lambda^{-3.48}$) [40] and for the body also liquid water scaling with the interaction path length.

3.1 | Hyperspectral imaging (ex-vivo)

Beetles captured in-situ from Nyteboda, Sweden, were measured according to Section 2.1 and had a median body length of 4.8 mm (IQR: 0.25 mm), median body width of 1.9 mm (IQR: 0.16 mm), and a median wing length of 6.7 mm (IQR: 0.40 mm).

Beetle wings appear transparent in the diffuse angle of light incidence (Figure 2A) but can display structural colored patterns at the specular light condition. A false-color image was formed from the hyperspectral images in Figure 2B, C. Three bands (1320, 1064, and 808 nm) coinciding with commercial laser diode wavelengths are displayed in red, green, and blue lines. The reflectance spectra of three specular pixels are shown in Figure 2D, exhibiting strong spectral fringes. Thicker wing sections display narrow spectral fringes. The extreme wave numbers (indicated with closed and open circles in Figure 2D) were used to calculate the wing.

膜厚度morphology by a linear fitting method [41]. We also present the spectrum of the elytra, which is sloped due to the melanization of the insect carapace and is approximately by,

$$R_{Elytra} = 13.6 \times \exp \left(-8.65 \times 10^{19} \lambda^{-6.83}\right) + 2.92$$

where $R_{Elytra}$ is in percentage and $\lambda$ is in unit of nm. Parameter 13.6 corresponding to 13.6% of diffused light from elytra was collected, parameter $8.65 \times 10^{19}$ is for scaling the fitting equation and parameter 2.92

![Figure 2](image-url)
corresponding to the reflectance offset due to refractive index. The melanization gain factor value from our fitting is 6.83 which is larger than the one provided by the reference, 3.48 [40].

The wing interference pattern (WIP) can roughly map out the wing membrane thickness distribution, see Figure 3A, B. Specular pixels were selected with an intensity threshold, enabling calculation of the membrane thickness distribution of the insect wings [42]. The thickness of the membranes decreases from the anterior wing margin to the posterior wing margin, as the veins confined to the anterior wing margin thicken the membrane to support the vein-free structure near the posterior wing margin.

Fringes shown in Figure 2D only provide wing thickness information of the selected pixels. It is like perform a point measurement on the membrane with a spectrometer. But for the purpose of remote sensing, it is more likely for the lidar to retrieve a strong backscattered specular signal reflected from the whole wing, rather than a small section of the membrane. Therefore, we spatially integrated all spectral fringes for all specular pixels and the result effective fringe is shown in Figure 3C. If such fringes survived a spatial integration into an effective fringe from the entire wing, it would enable us to retrieve the nanoscopic wing thickness remotely, which could be highly species-specific. Note that spatially integrated fringe magnitude is more moderate than the ones from individual pixels in Figure 2D. The same measurements were carried out on 10 randomly chosen beetles to estimate the variation among individuals. The obtained effective wing membrane thickness of the entire wings of beetles was thereby calculated to 525 ± 28 nm. This biological variance of just 5% is exceptional by itself, considering that the variance of wingbeat-measurement displays at least five times higher relative spread (>25%).

All wavelengths of effective fringe were corrected for the angle of incidence of 22°, and the fringe maxima and minima from Figure 3C have wavelength values of 1592 nm, 1063 nm, 802 nm, and 646 nm under normal incidence. Three commercial laser diode wavelengths were also displayed in Figure 3C, where laser bands 808 nm and 1064 nm are very close to the destructive and constructive wavelength of 802 nm and 1063 nm.

### 3.2 Imaging Polarimetric Goniometry, DoLP (ex-vivo)

Using a goniometer, we investigated the DoLP of beetles in different anatomical planes in backscatter- and forward scatter configurations. DoLP results are presented in Figure 4-5.

In backscattering configuration, see Figure 4, lights scattered from the beetle body and elytra are highly depolarized regardless of the aspect angles. Small features...
such as hair on the beetle maintain a high DoLP due to light undergoes less scattering events within small features. Beetle wing exhibits minimal de-polarization when the light impinges in specular condition (±8°), as shown in Figure 4B. However, the DoLP of the wing changes when the observer moved away from the specular condition, see Figure 4A,C. Hence, when the beetle wing is in a normal incidence with the lidar probe beam, we will receive the strongest co-polarized backscattered signal.

In forward-scattering mode (see Figure 5), the impinging light maintains a high degree of polarization. It could be due to the forward scattered light undergoes a single or few scattering events through the thin body structure, such as wings and elytra. Since light did not penetrate through the thicker body part of the beetle, the polarization features of those areas could not be determined in the forward scattering configuration.

### 3.3 Imaging Polarimetric Goniometry, OCSs and phase function (ex-vivo)

The goniometer was also used to study the optical cross-sections (OCSs) and phase function of ex-vivo beetles. The measured OCS (in mm²) of a beetle is presented in Figure 6 in all anatomical planes for co- and de-polarization. Anatomical terms in the figure are used to describe the beetle positioning to the camera. In both extinction and backscattering modes, the largest OCS value is observed when measuring the beetle from ventral (belly) or dorsal (back). According to Figures 4 and 5, a beetle has the largest projection if the beetle is observed from the dorsal/ventral side, due to the positioning of wings and elytra corresponding to a large cross-section area.

Figure 4 shown that the wing membrane retains a high degree of polarization in backscattering mode. The
de-polarized signals in backscatter-roll and backscatter-pitch diagrams in Figure 6 indicate that light undergoes multiple scattering in the membrane, which could be from the thicker sections of the wing, such as the veins.

In the backscattering mode, the backscattered signal intensity is related to the reflectance of the target and how much light was collected by the camera. Therefore, the OCS values in extinction mode are way larger than the backscattered OCSs. The contribution of wings or elytra to the co- and the de-polarized signal can be observed in Figure 6 in each diagram by comparing the OCS values before and after removing wings and elytra. With the OCS values provided in Figure 6, we can calculate the DoLP of beetle with Equation (1) from different aspects. For the frontal aspect, the beetle body+wing+elytra has a DoLP of 56%, beetle body+elytra has a DoLP of 55%, and beetle body has a DoLP of 55%, which is in good agreement with the DoLP visualization result in Figure 4A. The DoLP was calculated for the dorsal aspect, where beetle body+wing+elytra has a DoLP of 76%, beetle body+elytra has a DoLP of 65%, and a beetle body has a DoLP of 55%.

The backscattering OCSs of beetle is parameterized into a set of spherical harmonics with scaling and axial flipping [34]. Furthermore, spherical harmonic coefficients were then obtained through regression. The coefficients are presented in Table 1, where the unit sphere has a radius of 1 and center at the coordinate’s origin.
Spherical harmonics have two indices, where \( l \) is the degree, and \( m \) is the order. The values of the real part of spherical harmonic were stored for all \( l \) and \( m \) (degree up to \( l = 3 \)) and sum up to real cross-section values (in the unit of \( \text{mm}^2 \)), see Figure 7. Such table values in Table 1 can confirm or disconfirm a given field observation could be a bark beetle observed from an arbitrary direction. The ratio between co- and de-polarized signals from the spherical harmonics in Table 1 can be used to identify possible angular dependence of DoLP or melanization.

The phase function of a beetle at two observation aspects was investigated with the goniometer. The scattering pattern was measured by rotating the light source in a circle and illuminate the beetle from different scattering angle \( \theta_{sc} \).

When the beetle is facing the camera (Figure 8C), the angular distribution of scattered light intensity shows a

**TABLE 1** Parameterized table values of a set of harmonics bases used to generate the backscattering OCS in 3D. The corresponding images are presented in Figure 7

| Backscattering                  | Copol. (\( \text{mm}^2 \)) | Depol. (\( \text{mm}^2 \)) | Copol. (\( \text{mm}^2 \)) | Depol. (\( \text{mm}^2 \)) | Copol. (\( \text{mm}^2 \)) | Depol. (\( \text{mm}^2 \)) |
|---------------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|
| Body + elytra + wings           | 0.67                        | 0.71                        | 0.50                        | 0.42                        | 0.24                        | 0.19                        |
| Body + elytra                   | 0.27                        | 0.21                        | 0.27                        | 0.21                        | 0.30                        | 0.26                        |
| Body                            | -1.75                       | -1.95                       | -1.75                       | -1.95                       | -1.75                       | -1.95                       |
| **Y_{20}**                      |                             |                             |                             |                             |                             |                             |
| **Y_{21}**                      |                             |                             |                             |                             |                             |                             |
| **Y_{22}**                      | -0.35                       | -0.19                       | -0.17                       | -0.14                       | -0.05                       | -0.05                       |
| **Y_{30}**                      | 3.11                        | 0.72                        | 0.43                        | 0.13                        | 0.03                        | 0.03                        |
| **Y_{31}**                      | 0.16                        | 0.12                        | 0.33                        | 0.03                        | -                           | -                           |

**FIGURE 7** Backscattering OCS of a beetle from three anatomical planes are parameterized by a set of spherical harmonics. Dots represent measured values, and lines are projections of harmonic fits. The color coding is blue to purple shading by height.
strong forward scattering feature for co-polarized light at the given wavelength, where its intensity is roughly 20 times stronger than the de-polarized signal in the forward direction (Figure 8D). Even when wings and elytra were removed, the body of the beetle is still strongly forward scattered, which could be due to the body size of a beetle is small, so the light undergoes fewer scattering events. Lidar probe and detect a small amount of backscattered light from in-flight insects, other sensors such as E-trap [43] could, in principle, capture forward scattered light. For insects that are highly forward scattering, a forward detection configuration will require less probe intensity and still acquire strong forward scattered signals.
When the side of the beetle is facing the camera (Figure 8E), it also shows a strong forward scattering feature in the phase function. However, its scattered light intensity is 3 times weaker comparing to Figure 8C when wings and elytra are still attached to the beetle. This magnitude difference is due to the changing observation aspect of the beetle has changed the size of projection area from the wings, see the difference in Figure 5B,C, and this magnitude difference is diminished when the wings are removed from the beetle, see the difference in Figure 8C,E. Changing aspects of the beetle do not seem to have a strong impact on changing the de-polarized signal magnitude in Figure 8D,F for backscattering. This could be due mainly to that, the body and elytra are the main contributors to the de-polarized backscattered signal, and that there are no big projected area changes when the beetle is turned around from the front to the side. The big intensity changes for the de-polarized forward scattering signal when the beetle was turned could be due to the scattered light from the left-wing were blocked by the beetle body.

### 3.4 Entomological chamber (in-vivo)

Roughly, 200 beetles were released inside the entomological flight chamber to study the dynamic properties of wingbeats of free-flying beetles. However, not all the beetles were flying within the probe volume, and the same beetle could have entered the probe volume more than once. We also discarded observations that do not contain at least three clear wingbeats, as we cannot resolve a WBF if it is less than three wingbeats in the time series are available, and we discard signals that contain significant changes in body orientation, as we could not apply our parameterization method when the body signal is not consistent. Therefore, the amount of observations we presented does not correspond to the number of beetles recorded. Their backscattered signals were recorded for two laser bands in co- and de-polarization. Suppose we assume all the beetles have a similar flight speed and mostly fly horizontally. In that case, we expect a long observation recording from a beetle when it is flying along the beam and a short observation when it is flying perpendicularly to the beam. Examples of long and short transit observations of flying beetles are presented in Figure 9. A short observation should contain at least three clear wingbeat signals. In the bark beetle case, a short observation with at least three clear wingbeats is roughly 30 to 40 ms long in time. And a long observation for bark beetle typically contains more than six clear wingbeats, and its corresponding transit time is roughly longer than 70 ms.

The backscattered signal rises and drops throughout many wingbeat cycles, as seen in Figure 9A,C. The oscillatory signal components are contributed from the wing during the wingbeat, and the non-oscillatory component is contributed from the body elytra. The WBFs of 68 bark beetle signals were acquired in the entomological flight chamber, and bark beetles have a median of a WBF of 104 Hz with an IQR of 26 Hz.

A parameterization method [24, 34] was used to project the observation data onto a set of discrete harmonics, which can then estimate in a unit of contributions to the cross-section (mm²). The strength of the fundamental tone appears different in respect to the second harmonic tone for long and short transit observations, which could relate to the orientation and wing dynamic of the flying beetle [34, 44]. Brydegaard and collaborators [34] suggested an insect wing dynamic model, when an insect is observed from the side with lidar, will result in a strong first overtone in the power spectrum. When the insect is observed from the front, or back it yields a strong fundamental tone.

InGaAs co-polarized signal (at 1550 nm) in Figure 9, the difference between the fundamental tones to the first overtone is large in the short transit and small in the long transit observation is in disagreement with the model [34]. However, the diffuse signal from Figure 9 is in agreement with the proposed model. For example, the Si de-polarized signal (at 808 nm) has a large tone difference between the fundamental tone to the first overtone in the long transit, and such difference becomes smaller for short transit. Jansson and collaborators [44] also provided several results that argue that the model in [34] only applies to the diffused signal instead of specular.

### 3.5 Lidar (in-vivo, in-situ)

To achieve the most effective management of damage control of bark beetles, we need to monitor their distribution to determine the active measures. Bark beetles release pheromones to attract other beetles for breeding or initiating an attack on healthy trees [3, 45]. This released pheromone is invisible to human eyes and cannot be easily monitored. We employed our entomological lidar system in a field together with pheromones mixed with aerosols to illustrate that our lidar can simultaneously monitor the pheromone plume and insects attraction. The measurement result is presented in Figure 10.

As presented in Figure 10, our lidar system has successfully monitored both aerosols smoke and insects within the same time-range map. Insect observations are shown as intensity snippets in the map instead of a
**FIGURE 9** Dual-band polarimetric modulated signal acquired from the entomological chamber for beetles. A, C, The backscattered signal in time series from in-flight beetle for long and short transit time observation. B, D, The corresponding power spectrum for each transit time and time series were parameterized by a set of discrete harmonics weighted with the body contribution.

**FIGURE 10** An example of lidar observation. A, Time range map of insect and smoke release event. B, A small section of the time range map is zoomed in and presented. C, The time series of the observation in B.
continuous intensity distribution as the smoke. A presumed bark beetle observation is highlighted in Figure 10B,C based on the event distance and release time. A long and short lidar transit observation are presented in Figure 11. Both are presumed to be bark beetles based on the release location and time. Their recorded WBFs are also within the bark beetle WBFs span measured from the flight chamber (a median of 104 Hz and IQR of 26 Hz).

The parameterization method was used to retrieve the strength of the body and harmonic tones from the lidar observations. The interpretation of the signals is important when the body to fundamental tone ratios are close. The relation between the fundamental tones to the first overtone of the lidar measurement is similar to what we observed in the flight chamber in Figure 9. When the transit is long, the fundamental tone is stronger than the first overtone. When the transit is short, the difference in tone strength decreases.

Then we investigated if the ratio between body and fundamental tone from the lidar measurement in Figure 11 is comparable to the flight chamber recordings in Figure 9. Unlike the flight chamber, lidar only had one laser band at 808 nm, and it records the sum of both co- and de-polarization. The body to fundamental tone ratio is calculated for lidar by dividing the body signal strength by the fundamental tone strength in Figure 11B,D. The long observation gives a ratio of 3.2, and the short observation gives a ratio of 5.3. The body to fundamental tone ratio is calculated for the flight chamber by dividing the sum of the co- and de-polarized body signal with the sum of co- and de-polarized fundamental tone signal in Figure 9B,D for Si measurements only. Six long observations from the flight chamber within time interval 70 to 100 ms were randomly selected, and they gave a median value of 10.6 (IQR:4.4) for the body-to-fundamental-tone ratio, where 6 short observations within time interval 30 to 40 ms gave a median of 5.5 (IQR:1.2). The body to fundamental tone ratio is nearly the same for both lidar and flight chamber short transit measurement. However, their values were fairly different for the long observation, which could be due to a long transit time means the beetle could have several orientation changes within the same recording, while for a short transit beetle is most likely just flying perpendicular through the beam without big adjustments to its heading. Changes of orientation within the same recording could lead to erroneous strength estimations for body and harmonic tones.

We also looked into if the lidar measured OCS values are comparable to the flight chamber and goniometer measurements. Our lidar measured OCS value for a presumed bark beetle is a factor 10 times larger than the lab measurement. Lidar does face the challenge of calibrating the signal intensity into absolute OCS precisely. Unlike the goniometer and flight chamber that allows the placement of calibration targets in the detection path, lidar is commonly placed in a complex environment and monitoring a long distance (>100 m). Most of the time, the monitoring environment does not allow the placement of a calibration target. Lidar uses its beam size on the termination board to mathematically convert the lidar signals into OCS (mm²). Some of the beam widths are imaged.

**FIGURE 11** Representative example of insect long and short detection event observed in the field measurement. A, C, Time series of observations with different transit times. B, D, Observations in time series were parameterized by a set of discrete harmonics weighted with the body contribution.
outside of the lidar linear array at a short range, leading to a wrong OCS estimation for observations. The lidar transect in Nyteboda had a range of 120 m, but the beetles were released halfway, at 60 m, which is considered a short range for the lidar system.

4 | CONCLUSION AND OUTLOOK

This paper evaluated the feasibility and characterized optical scattering from bark beetles as lidar target, ex-vivo, in-vivo, and in-situ. We explored various optical domains such as imaging of anatomical features, spectral content, polarization properties, goniometric scatter analysis, modulation properties, and remote sensing. We applied hyperspectral imaging, polarimetric imaging and goniometry, multiband modulation spectrometry, and laser radar.

The body melanization and thickness across bark beetles wings in the range of 600–1600 nm was measured. It was showed that wing interference patterns survive spatial integration over the entire wing producing an effective spectral fringe that can be exploited for remote sensing. By determining this effective fringe from numerous specimens, we discovered an extraordinary small variance of wing thickness less than one magnitude lower than the variance of WBFs. This not only raises questions of fundamental nature in biology, but such small variance also has great implications for differentiating insect species remotely should this be a generally occurring feature across species. Further studies are needed to validate this feature for remote identification of insects.

Anatomical features responsible for the depolarization of backscatter light were pinpointed. Beetle wings maintain a high degree of co-polarization even when the wavelength is not in condition for resonant backscatter. We also demonstrated that both the body and elytra almost randomizes the polarization entirely. This is noteworthy because both body and elytra are melanized (a feature normally increasing DoLP) and elytra are thin, providing a limited optical path length for multiple scattering and depolarization.

Analyzing forward scattered light, we concluded that the body is mainly opaque for near-infrared light (in the tissue window) despite being a small organism of a few mm size. From the highly co-polarized light transmitted through both wings and elytra, we concluded that the transmitted light experiences a shorter interaction path length compared to the reflected light. This is counterintuitive compared to classical medial optics but compatible with similar findings in mosquitoes [30].

Quantitative optical backscatter cross-sections from all anatomical projections for both co- and de-polarized laser light are presented as well as extinction coefficients. These estimated values are useful for designing field monitoring instrumentation and interpreting entomological lidar data, such as determining plausible bark beetle targets in an ensemble of insects. We concluded that the DoLP is invariant with heading and observation angle, and such it could be a feature for identification. This finding is similar to earlier findings for mosquitoes [29, 30].

By repeated scans and relieving the beetles from wings and elytra, we could attribute different fractions of the cross-section to different anatomical parts. This is interesting because entomological lidar is capable of isolating oscillatory scattered light from wings from the non-oscillatory light scattered of the body and therefore provides a stronger foundation for remote detection. We also notice that the angular dependence of cross-sections and (consequently, the circular and spherical harmonics required to explain them) are three folded symmetrical for both the body and wings. However, this is not the case for the contribution from the elytra, and beetles can thus be expected to display asymmetric cross-section depending on the observation aspect.

By identifying symmetries, we provided spherical functions of cross-sections using a limited set of spherical harmonics. Thus scattering by an insect target from an arbitrary observation aspect can be given with quantitative values in a table. This paves the way for establishing a library for cross-section for insect species of key importance such as pests, disease vectors, or pollinators.

With goniometric scatter analysis, we demonstrated dominant co-polarized forward scattering. The scattering phase function can be understood as a product of the cross-section from the illumination and observed aspect in conjunction with an intrinsic Henyey-Greenstein scattering function. The retrieved phase function helps us understand the light interaction with such small biological samples. It could also have implications for field detection schemes where the strong forward scattering is exploited to ease laser power requirements.

By polarimetric dual-band modulation spectroscopy on free-flying bark beetles, we have identified the range of WBFs. We applied parameterization and demonstrate quantitative cross sections values for a high number of overtones. The proposed wing dynamic model from [34] apply to diffuse signal observation. The DoLP of long transit observation is in agreement with the goniometry measurement.

We contributed a solution to monitor an invisible pheromone released by bark beetle by employing our lidar system in the field and mixing aerosols with pheromones. We successfully observed the bark beetle signals and pheromones smoke plumes simultaneously. Moreover, they both have distinguishable characters in the lidar time and range map, so it is easy to tell them apart.
We compared the lidar observations to the flight chamber measurements to prove the feasibility of having the lidar as a tool for monitoring the bark beetles. We had some challenges with absolute cross-section calibration, but there are plenty of other features to identify the beetles. Firstly, both lidar and flight chamber measurements showed the same relative tone strength relation for long and short transit observations. Secondly, the WBF measured by lidar is within the WBF span measured by the flight chamber. Thirdly, the body to fundamental tone ratio is nearly the same for short transit observation in both lidar and flight chamber measurements.

With clues provided from this work, we can distinguish bark beetle observations from in-situ lidar or other sensors measurements. Our methodology can be applied to other key insect species and enable developing a specific monitoring system. Future studies should focus on developing an insect outbreak index system to indicate the risk by employing several lidar systems at various locations.

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CONFLICTS OF INTEREST
The authors declare that there is no financial or commercial conflict of interest.

DATA AVAILABILITY STATEMENT
Data is available by contacting the correspondence author upon a reasonable request.

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