Identification and spatial negotiation through a hole, and possible influence of individual size

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Flying insects are able to negotiate complex environments such as tropical rainforests as well as less complex environments such as flowering meadows although little is known about how they avoid collisions while doing this. In this study, insect negotiation through holes was investigated using Bumblebees *Bombus terrestris* and Orchid bees *Euglossa imperialis* as test animals. This was done by allowing the individual to fly through a tunnel with an interchangeable endwall, with a varied assortment of attachable exitholes. The flight position were analysed, and ability to exit through the hole, light intensity, hole shape and/or size and individual size where recorded. We found that orchid bees in general are more willing to fly through holes than bumblebees. However, both species tend to fly through the hole at the safest point, i.e. point furthest from the surrounding edge. In addition, orchid bees tend to fly closer to the safest point than bumblebees do when negotiating larger holes . Lastly, we find no correlation between individual size and capability to negotiate through a hole.

Keywords: Collision avoidance * Navigation * Bumblebee * *Bombus terrestris* * Orchid bee * *Euglossa imperialis*

Introduction

As an insect flies through its surrounding it is able to navigate through its proximate area without crashing into obstacles. How are insect able to avoid crashing into obstacles? Especially when the insects do not have stereovision? As an individual flies through a tunnel it maintains an equal distance to the surrounding walls (Linander et al., 2015). It does so by balancing the optic flow, balancing the speed of motion passing over each eye (Linander et al., 2015). This results in that the individual remains centralized in a tunnel (Kirshner and Srinivasan, 1989, Srinivasan et al., 1991, Srinivasan et al., 1996, Dyhr and Higgins, 2010). Kirshner and Srinivasan (1989) tested this by allowing bees to fly through a tunnel, that had moving or stationary walls lined with vertical stripes. In the tunnel with the stationary walls the individual maintained centralized in the tunnel during flight. But in the tunnel with the moving walls the individual would remain centralized as long as the walls moved at the same rate in the same direction. However, when one wall was moving in the flight direction and the other one was moving against the flight direction the individual tended to fly closer to the wall moving in the same direction as the flight. This, as the rate of optic flow is lower from the wall moving in the same direction compared to information from the wall moving against the flight direction.

By using the centering response individuals could quickly avoid flying into obstacles by utilizing the suddenly increased optic flow information over one eye, thereby quickly steer in the opposite direction of the obstacle (Srinivasan and Lehrer, 1984, Srinivasan and Zhang, 1996). Interestingly, navigation and visual flight control in the Hymenopteran subfamily Apinae tends to be based on the usage of visual cues (Collett, 1996, Cameron, 2004, Baird et al., 2005). Here, studies have mainly concerned honeybees and bumblebees (Avarguès-Weber, 2011). Gibson (1950) stated that animals could extract information about the relative distance to obstacles using information extracted from the image of the environment as it moves over the retina, referred to as optic flow. This is the case for bumblebees and other hymenopterans (Baird et al., 2010).

For an animal to be able to discern its environment using vision, there is a need for light input (Chittka et al.,1999, Reber et al,2015). In nocturnal animals, a morphological adaptation is often present to allow for a greater inlet of light, such as enlarged eyes or anatomical difference in eye structure (Warrant, 2004). There is also a correspondence present between individual body sizes and eye size (Jander and Jander, 2002, Hagen and Dupont, 2013). A larger eye size could reduce the relative amount of photon noise (Warrant, 1999), otherwise hinders visual stimuli to leave out information (Lillywhite, 1977). Generally individual size variations are present within a species (Hagen and Dupont, 2013); in this regard bumblebees are highly fluctuant in size (Jander and Jander, 2002), whereas orchid bees have small individual size variations within species (Francoy et al. 2012).

The Buff-Tailed bumblebee *Bombus terrestris* can mainly be found in open habitats such as meadows (Goulson et al., 2001), a habitat with few obstacles that primarily is comprised of bright areas but also include some dim areas (Kreyer et al., 2003). This in great contrast to the orchid bee habitat, which is comprised of a complex habitat (Pokorny et al., 2015), a dense neotropical rainforest (Dodson, 1966, Roubik, 1989) reducing the amount of sunlight that can reach the forest floor (Dodson, 1966).

In this study we allowed bumblebees and orchid bees to fly through differently sized or shaped holes. The accuracy, measured as the distance from the safe point, was compared between the species and the different types of holes. The experiment was conducted in various light intensities to investigate how this affected hole negotiation. To investigate the possible importance of individual size each individual was measured and analysed in regards to individual success rate.

Here we hypothesise that orchid bees are more successful when negotiating holes compared to bumblebees. This is because orchid bees has evolved in a highly complex environment where a developed collision avoidance strategy is crucial for survival, whereas bumblebees have evolved in a less complex environment, potentially not requiring as developed collision avoidance skills as orchid bees. We also hypothesise that orchid bees would be able to fly in lower light intensities and not continuously fly as close to the safe point as the bumblebees would. This would allow the rain forest living bees to fly the shortest route possible without too many detours. In regards to individual size it could be assumed that a larger individual would be able to negotiate in lower light intensity than a smaller would. This as larger eyes would have a larger inlet of photon thus producing a clearer image on the individuals' retina.

Materials & Methods

Study animals

Commercially bred (Koppertt, UK) Buff Tailed Bumblebees *Bombus terrestris* (Linnaeus, 1758) were subjected to tests indoors at Lund University, Sweden. While Orchid bees *Euglossa imperialis* (Cockrell, 1922) were subjected to tests outdoors at the Smithsonian Tropical Research Institute (STRI) on Barro Colorado Island (BCI), Panama.

General methods

The experimental setup consisted of a tunnel (30cm x 30cm x 60cm) lined with a randomised black and white chequered pattern (2cm x 2cm squares), while the black wall with the entrance hole for the bees was lined with a red chequered pattern to provide strong contrast of the bee against the background in the camera view.

The tunnel was lined with patterns to provide sufficient visual texture for the bees to control their flight in the tunnel. The end of the tunnel had an interchangeable side where black foam boards with different exit holes could be attached (fig.1). Seven different holes were used: circular holes with a diameter of 5cm, 7.5cm, 13cm or 15 cm, a triangular hole, no hole or a hole made from overlapping circles (fig. 2).

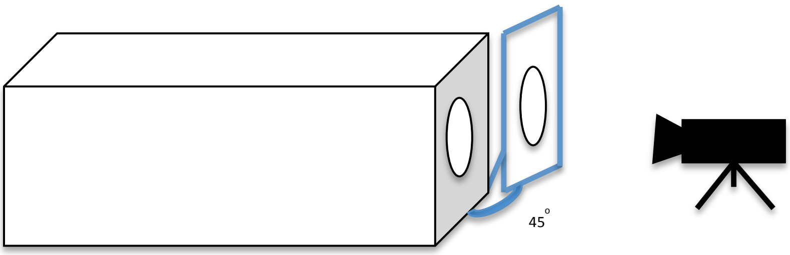

Figure 2: Experimental setup for flight tunnel experiment. Note: The blue square indicating a mirror used in the experiment, thus making it possible to see the exact moment when the bee exited through the hole.

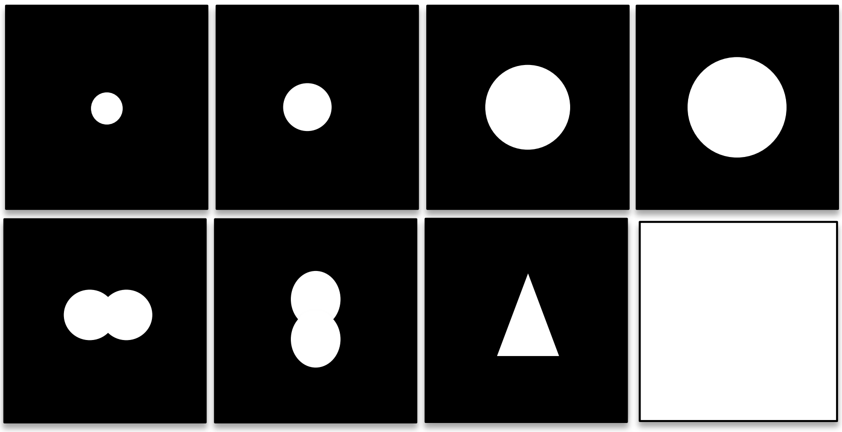

Figure 1: Holes and shapes used for flight experiments. Top row (left to right): 5cm, 7.5cm, 13cm, 15cm. Bottom row: Doublecircle, Doublecircle-Vertical, Triangle and a Control, consisting of no hole i.e. an open end of the tunnel.

Each bee was captured in a small plastic tube and released into the tunnel by inserting the open end of the tube into a small hole in the centre of the back wall, opposite to the exit hole. The bees were given five minutes to exit the experimental tunnel. Cases in which the bees did not fly out within five minutes were defined as failed exits. Individuals were timed from the moment they left the tube until they exited through the hole. Each flight was timed when the experiment was conducted and this time was rechecked when the videos of the flights were analysed.

Individual bumblebees were caught directly in the lab in which they were kept. Individual orchid bees were caught at scent baits that were made by applying a couple of drops of 1,4-Cineol (Aldrich Chemistry, USA) or Eucalyptol 99% (Aldrich Chemistry, USA) on a piece of toilet paper enclosed in a tea strainer ball suspended in a transparent thread 1 meter from the ground. Scent would be applied every morning, around 10 am and after lunch and/or as needed.

The experiments were conducted during the day, as *B.terrestris* and *E.imperialis* are diurnal. Bumblebee experiments were all conducted indoor at a constant room temperature of 22°C and Orchid bee experiments were conducted outdoor at a temperature ranging from 24.93 to 32.87^{o}C.

Bumblebee experiments were conducted in three light intensities, 6, 60 or 120 lux and each bee was allowed to acclimatise to the light intensity for 30 minutes prior to flight. Each individual was only allowed to fly once to avoid any learning effects. After the test, each individual was marked dorsally on its thorax with a numbered plastic plate.

The orchid bee experiments were conducted over a range of light intensities that were grouped into three ranges, low (0-599 lux), medium (600-1199 lux) or high (> 1200 lux). Each individual was marked dorsally on its thorax prior to flight using a marker pen and individuals were allowed to fly two times at most, however not in the same condition. Ten flights were recorded for each light condition for each hole in each species.

Data analysis

All flights were recorded using a Sony handycam (HDR-CX73OE, Tokyo, Japan) filming at 25 frames per second (fps) using night vision mode to allow filming in dim light. In order to determine the exact position where an individual flew through the exit hole, a mirror was placed beside or under the exit hole angled at 45 degrees, thus allowing the camera to record the moment the individual flies through the hole from two angles. The recordings were analysed using Quick time Player Version 7.5.5(1709) in order to locate the frame in which the bees exited the hole. These frames were then extracted from the film and the position of the bee with reference to the point in the hole that had the greatest clearance from all edges – that is, the safest point (SP) to fly through the hole – was determined using a custom script in Matlab (R2012b 8.0.783). Light intensities were measured using a light meter (Amprobe LM-100, USA), first inside and then outside the tunnel.

Body size measurements

The bumblebee’s inter-tegular (IT) width and eye length was measured using microscope (stemi SV6, magnification 1.6) connected to a camera (Nikon DS-Fi1c) and a tv-adapter (TV2/3”C 0.63x) and measured in the imaging software (Nikon, NIS Elements ver. 4.20). IT and eye length were measured four times for each bee and an average was extracted, which was used in later analysis.

The orchid bees IT was measured from photographs (Canon eos 450d with Canon Macrolens EF-S 60mm 1:2:8) of the individual through a fine mesh (0.2x0.2mm).

Statistical analysis

All data was subjected to Shapiro-Wilks normality test, where the data was determined as non-normally distributed. Thereafter a Spearman rank correlation was used. Statistical analysis was conducted using IBM SPSS (Version 20.0 for macintosh).

Ethics

No permits were required for this study as the study uses invertebrate species, which are not threatened (Jordbruksverket, 2014). However, handling of individuals was done swiftly and kept to a minimum in order to reduce stress. Bumblebees died naturally with free access to their hive, food source and 12:12h light condition. Orchid bees were released directly after flight tunnel experiment.

Results

General results

The bumblebees exited the hole in 54.33% of the flights, while the orchid bees made it through the exit hole in 93.6% of the flights. In contrast to the orchid bees, the bumblebees were not able to negotiate a 5 cm hole in any light condition, whereas the orchid bees were able to do this in all light conditions.

The effect of light intensity on the ability to negotiate holes

Light intensity had a significant effect on the ability of bumblebees to exit the tunnel via a hole (5cm, 7.5cm, 13cm, 15cm, triangle, doublecircle and a vertical doublecircle) ($P<0.001$, $n=720$ Light intensity), (fig 3). A significant relationship was also seen between time for the individual to exit the tunnel and light intensity ($P<0.001$, $n=720$, fig 4).

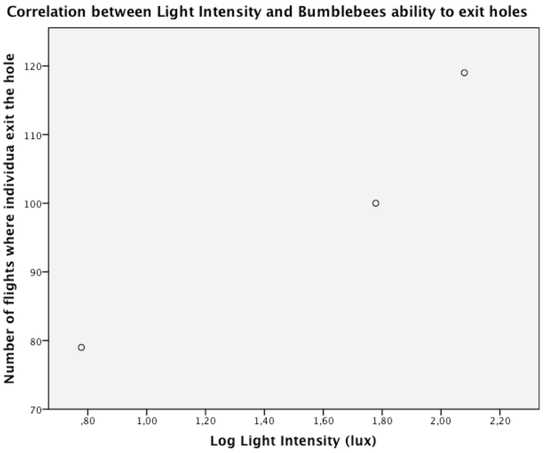

Figure 3: A correlation between individuals exiting holes and light intensity was found in Bumblebees. Indicating that, as light intensity increased, more individuals were able to negotiate the exit hole. In low light intensities n =251 (5cm=25, 7.5 cm=29, 13 cm=45, 15cm=40, Doublecircle=39, Doublecircle Vertical=25 and Triangle=48). Medium light intensities n=182 (5cm=24, 7.5 cm=31, 13 cm=29, 15cm=26, Doublecircle=24, Doublecircle Vertical=24 and Triangle=24) and in high light intensities n=187 (5cm=23,7.5 cm=24, 13 cm=26, 15cm=24, Doublecircle=32, Doublecircle Vertical=30 and Triangle=38).

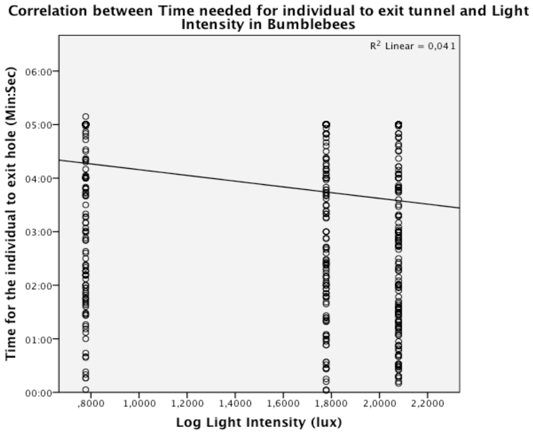

Figure 4: A correlation between flight time and light intensity in Bumblebees. Suggesting a longer flight time in lower light intensities (P<0.001, n=720). In a 15 cm hole the average flight time for a bumblebees in 6, 60 and 120 lux was 3:15, 2:08 and 1:42 minutes. With trendline (R^2:0.041).

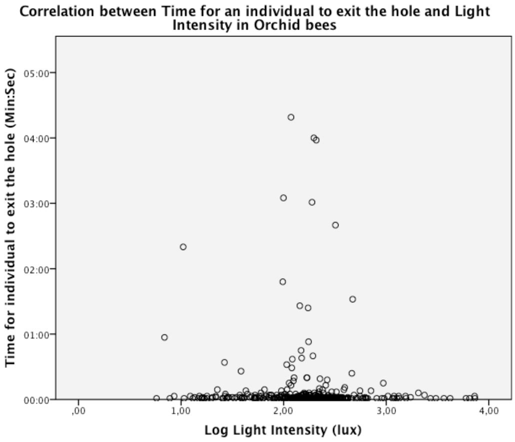

Figure 5: A correlation between flight time and light intensity was detected in Orchid bees. Suggesting a longer flight time in lower light intensities (P=0.001, n=390).

Light intensity also had a significant effect on the ability of orchid bees to exit holes (P=0.020, n=470). The time that it took orchid bees to exit the tunnel through a hole was also affected by light intensity (P=0.001, n=390), fig 5. This indicates that, as light intensity decreases, both bumblebees and orchid bees take a longer time to exit the tunnel through the holes.

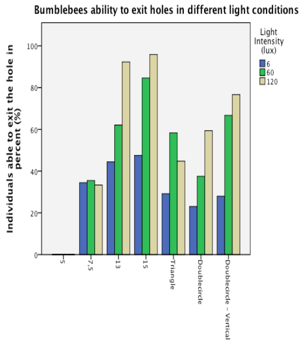

Figure 6: Bumblebees exiting holes (in percent) in different light intensities. In low light intensities (5cm (n:25), 7.5 cm (n:29), 13cm (n:45), 15cm (n:40), Triangle (n:48), Doublecircle (n:39), Doublecircle-Vertical (n:25)), Medium light intensity (5cm (n:24), 7.5 cm (n:31), 13cm (n:29), 15cm (n:26), Triangle (n:24), Doublecircle (n:24), Doublecircle-Vertical (n:24)) and high light intensity (5cm (n:23), 7.5 cm (n:24), 13cm (n:26), 15cm (n:24), Triangle (n:38), Doublecircle (n:32), Doublecircle-Vertical (n:30)).

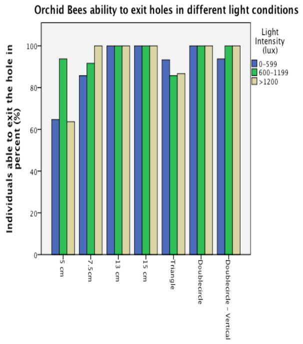

Figure 7: Orchid Bees exiting holes (in percent) in different light intensity. In low light intensities (5cm (n:17), 7.5 cm (n:14), 13cm (n:18), 15cm (n:18), Triangle (n:15), Doublecircle (n:18), Doublecircle-Vertical (n:16)), Medium light intensity (5cm (n:16), 7.5 cm (n:23), 13cm (n:25), 15cm (n:22), Triangle (n:21), Doublecircle (n:20), Doublecircle-Vertical (n:21)) and high light intensity (5cm (n:21), 7.5 cm (n:15), 13cm (n:12), 15cm (n:11), Triangle (n:15), Doublecircle (n:16), Doublecircle-Vertical (n:15)).

The effect of light intensity on position when negotiating holes

After establishing that light intensity is an important factor for the ability of bees to exit a hole of a given size (fig 6 and fig 7), we next investigated the effect of light intensity on the position of individuals as they flew out of circular holes (diameter 7.5, 13 and 15cm), a triangular shaped hole and a through a horizontal and a vertical double circle, (fig 8). This was done by measuring the lateral and vertical distance from the point that provides them with the greatest clearance from the edges, referred to as the safest point, as they flew out of a hole, (fig 9).

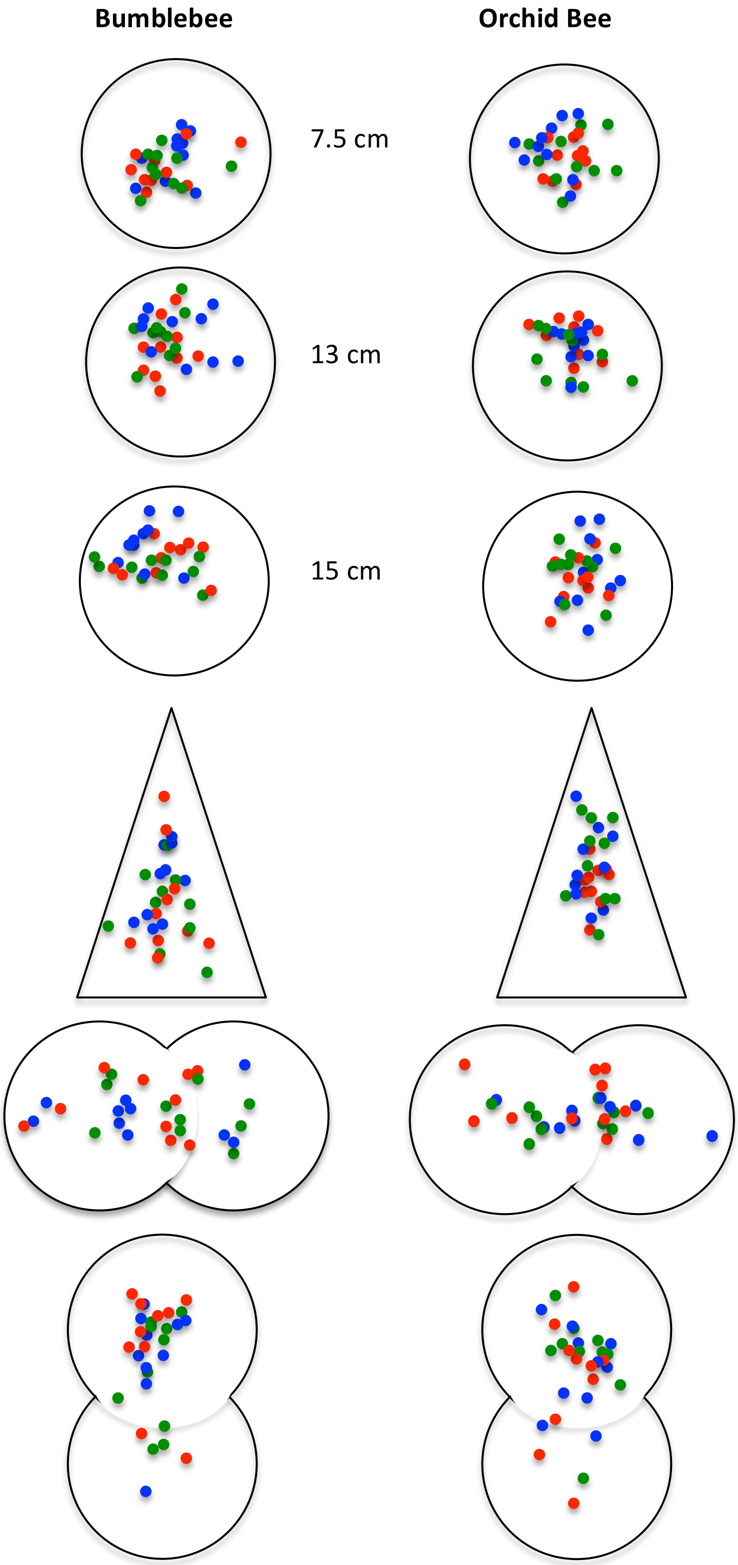

Figure 8: Bumblebee and orchid bee individuals exit point as individuals fly through various types of holes. Each point represents an individual (n=10/light intensity)

Red – low light intensities, Blue – medium light intensities, Green – high light intensities.

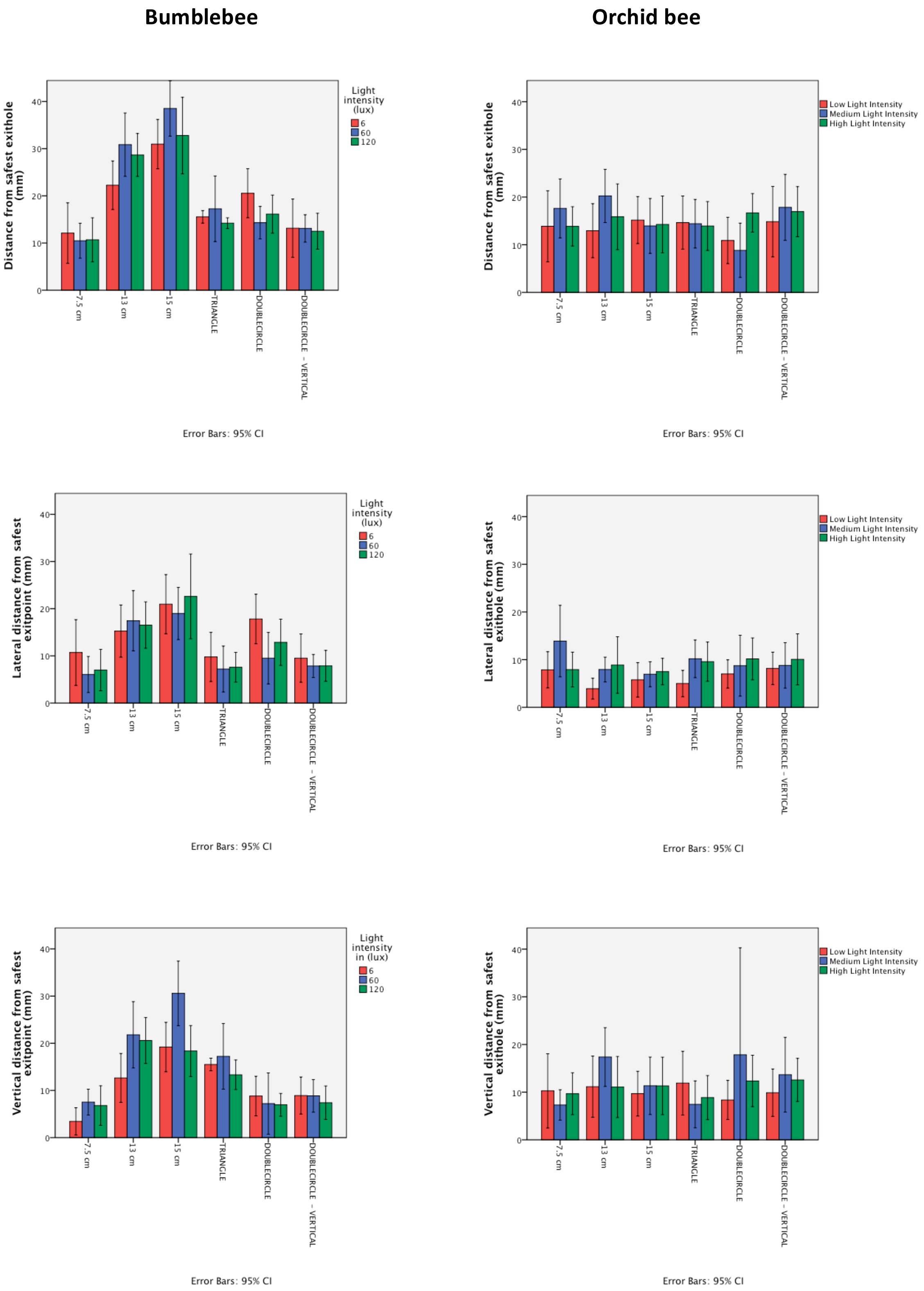

Figure 9: Distance from safe point for Bumblebees and orchid bees as they fly through various types of holes (n=10/light intensity).

Red – low light intensities,
Blue – medium light intensities
Green – high light intensities.

General results for all hole types

Bumblebee position when exiting the hole is not significantly affected by light intensity. 72.8 % (total n=182, std dev:0.03) of the bumblebees flew out above the safest point and 70% of the individuals flew out to the right of the safest point (n= 173, std dev:0.03). As with the bumblebees, the position of orchid bees when flying out of the holes was not affected significantly by light intensity. Again, the distribution of positions in the lateral direction was not equal as 45 % (n=91, std dev:0.49) flew out left of the safest point while 55 % (n=111, std dev:0.49) flew out right of the safest point. This difference was even more pronounced in the vertical direction where 60.2% (n=127, std dev:0.49) exited above the safest point while 39.8 % (n=84, std dev:0.49) exited below the safest point.

Specific results for each hole type

In regards to the different holes and shapes certain correlations were seen. For bumblebees flying through the 7.5 cm hole a negative correlation was seen between flight time and vertical distance from safe point (R:0.572, p:0.002, n:27), where a longer vertical distance from safe point correlates with a shorter flight time, (fig 10). In contrast, in orchid bees flying through a 7.5 cm hole no significant correlations were found.

In regards to bumblebee individuals flying through the 13 cm hole, a correlation between the vertical distance from safe point and light intensity, (fig 11)(R:0.274, p:0.043, n:55) could be observed. For orchid bees no statistically significant correlations were found for this hole size.

As for bumblebees flying through the 15 cm hole a correlation between flight time and light intensity was found (R:0.442, p<:0.001, n:64), which indicates that flight time in the tunnel is reduced with increased light intensity, (fig 12). Again, no significant correlations for orchid bees flying through a 15 cm hole were found.

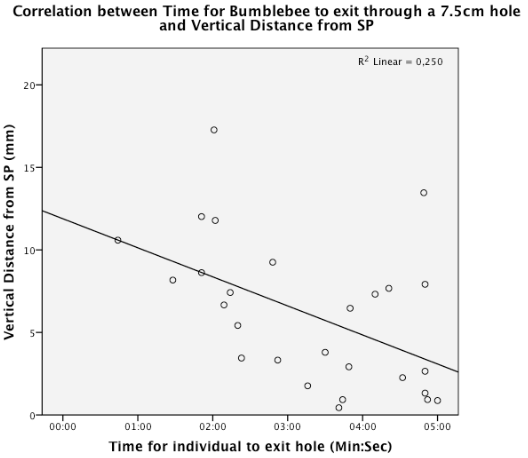

Figure 10: A correlation between vertical distance from safe point and flight time (R:0.572, p:0.002, n:27) for individuals flying through a 7.5 cm hole. With a trendline (R^2:0.250).

No significant correlations were found for bumblebees or orchid bees flying through the triangular shaped hole. Neither was it for bumblebees flying through the doublecircle or the vertical doublecircle. However for orchid bees flying through the doublecircle a weak correlation was found between flight time and vertical positioning from safe point (R:0.347, p:0.048, n:33). Indicating that vertical positioning of safe point was more frequently below the safe point than above for individuals flying through the doublecircle (below: 70.6%, n: 34). Individuals with a flight time over 30 seconds consistently flew below the safe point, while individuals

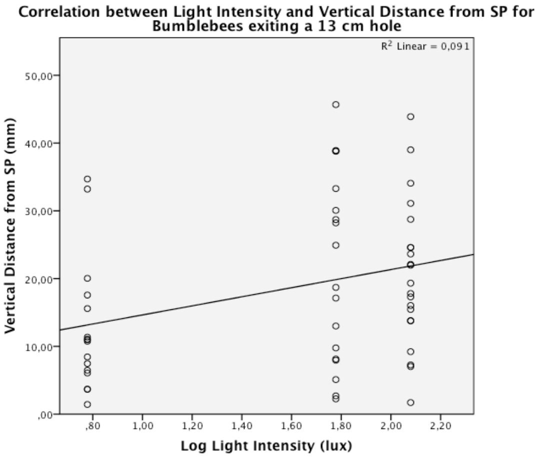

Figure 11: A correlation between light intensity and vertical distance safe point (R:0.274, p:0.043, n:55), for bumblebee individuals flying through a 13 cm hole. With a trendline (R^2 linear:0.091).

with a flight time below 30 seconds flew above as well as under the safe point. A correlation was also found for orchid bees flying through the vertical doublecircle, where vertical distance from safe point correlates with light intensity (R:0.384, p:0.023, n:25). This indicates that a higher light intensity correlates with a shorter vertical distance from safe point.

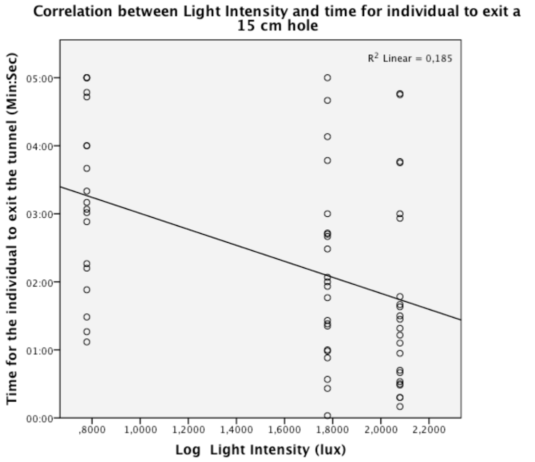

Figure 12: A correlation between flight time and light intensity (R:0.442, p<:0.001, n:64), for bumblebees flying through a 15cm hole. With a trendline (R^2 linear : 0.185)

The effect of body size on hole negotiation at different light intensities

Bumblebees vary in size, individuals with a larger inter-tegular width (IT) have larger eyes (Spaethe & Chittka, 2003), which could suggest that larger individuals might be able to more successfully negotiate through various holes, especially in dim light. This is because larger eyes would allow a greater sensitivity to light in the eye and the individual could be more accurate when negotiating in dim light conditions. We tested this by investigating if an individuals' body size affected its position as it flew through the hole.

General results for all hole types

This experiment tests the possible effect of individual body size on the ability to negotiate holes in various light intensities. The IT measurement for bumblebees was not of significant factor when negotiating holes, One-Way ANOVA (P=0.890, F:0.850, df:169). Nor was any significance seen in regards to eye length and ability to negotiate holes (Left eye P=0.306, n=596 and Right eye P=0.883, n= 596). In parallel, the orchid bees' IT measurement displays no correlation to the individuals ability to negotiate holes (P=0.952, n=470).

Discussion

In this study, we investigated the effect of dim light on the ability of bumblebees *B. terrestris* and orchid bees *E. imperalis* to negotiate holes. Using these species allowed for a comparison between a bee species subsisting in an open habitat (Goulson et al, 2002) and a species subsisting in a dense neotropical rainforest (Dressler, 1982) The study was divided into three sections; (1) the effect of light intensity on the ability to negotiate holes, (2) the positioning of individual when negotiating holes and (3) the individual body size influencing the ability to negotiate holes.

Effect of light intensity on the ability to negotiate holes

Light intensity had a significant effect on the ability of bumblebees and orchid bees to negotiate holes: In higher light intensities they could negotiate smaller holes (fig 9). However, orchid bees were able to negotiate a 5 cm hole, while bumblebees were not. There is a possibility that bumblebees would be able to negotiate 5 cm holes if there was a greater visual contrast between the inside of the exit hole and outside of it. If the hole was brighter the individual might have been able to negotiate it, as the individual would perhaps be able to see the hole and its edges. Future studies could determine if this is the case. Although it is possible, it might not necessarily be the case as bumblebees' habitat would not require it to the same extent as orchid bees.

Light intensity also influenced the individual flight time in the tunnel, as time to exit increases as light intensity decreases. This since the individual would require longer time to counteract the relatively high amount of photon noise reflected from the background in the dimmer light condition. Which, has also been seen in previous studies with bumblebees, where individuals flying through a tunnel lowered their flight speed in dimmer light conditions (Reber et al., 2015). Another possibility is that the individual would need to fly back and forward between the walls of the tunnel to scan its environment in order to find the exit hole. Or that it needs to fly back and forth from the entrance of the tunnel to scan its environment, as a learning flight, which has been seen in bumblebees when leaving their nest (Phillipides et al., 2013).

The orchid bees displayed a higher general success rate in exiting holes (93.6%) than bumblebees (54.33%) (fig 6 and fig 7). Orchid bees had a lower than 100% success rate negotiating the 5cm, triangular, 7.5cm and vertical doublecircle, in the highest light intensity, the bumblebee had a lower success rate in all types of holes and in all light conditions, when compared to orchid bees. A decrease in light intensity displays a lowered success rate, where the bumblebees success rate diminishes with lowered light intensity. This was also seen in the orchid bees, however not as cohesively as for bumblebee negotiation. As orchid bees have a more complex habitat the results were as expected. The lower light intensities used indoors compared to outdoors could also have influenced the results. In the future, the experiments with the bumblebees should be performed outdoors with higher light intensities.

From these results it can be concluded that light intensity is a determinant factor in the individuals ability to negotiate holes. A more complex habitat could possibly be a cofactor, allowing individuals to negotiate smaller holes.

Positioning of individual when negotiating holes

Generally the positioning of the bumblebees and orchid bees were not significantly correlated with light intensity. Individuals mostly exited vertically above the safest point (bumblebees 72.8%, orchid bees 60.2%). Likely due to that the major light source was from above the tunnel. Either as a fixed lighting, bumblebees, or as the sun, orchid bees. Orchid bees were however not exposed to direct sunlight from above the tunnel. In lateral direction individuals tend to fly to the right of the safest point (bumblebees 70%, orchid bees 55%). Why this is so is not known.

Specifically for the holes, some correlations were found. For bumblebees a correlation was found for the 7.5cm hole, where a longer vertical distance from the safe point correlates with a shorter flight time (fig 10). A similar correlation was also found for the 15 cm hole where flight time was reduced with an increased light intensity (fig 12). These correlation would suggest that if the individual can see the exit hole and deem it safe to exit it will likely fly directly out disregarding exiting as close to the safe point as possible. But in conditions where it is more difficult for the individual to clearly see the hole it might need to fly slower to orient itself.

Another correlation was also found for 13cm hole where a higher light intensity correlates with a greater vertical distance from the safe point (fig 11). This is likely due to that the lighting when conducting the bumblebee experiment came from above the tunnel, indicating that a stronger light source would attract the individual in a upwards direction from the safe point. However none of the correlations found in bumblebees were found for the same type of holes in orchid bees. For orchid bees a correlation was found for the doublecircle, where flight time and vertical positioning of individual was correlated: Individuals with a shorter flight time exited the hole both above and below the safe point, whereas individuals with a flight time extending 30 seconds consistently negotiated the hole below the safe point. A correlation was also found for vertical doublecircle where a higher light intensity correlates with a shorter vertical distance from the safe point. As expected and tested light intensity is an important factor for how efficient individuals negotiate holes.

From these results the conclusions that can be drawn are that, in dim light conditions, individuals are likely to exit close to the safe point of the hole, whereas in brighter light conditions individual negotiate faster and not necessarily close to the safe point. Thus reducing the risk of collision or wasting resources. As bees rely on horizontal optic flow to negotiate the environment, a lowered light intensity would increase amount of noise, disturbing the pattern of visual motion. Thereby resulting in a lowered flight speed and greater difficulties negotiating holes. To date no previous studies has been published on an insect's ability to negotiate through a hole, therefor material to compare this study to have not been found.

Individual body size influencing the ability to negotiate holes

Generally no correlations were found between the individual body size and the ability to negotiate holes. Even thou previous studies suggest that individual size in bumblebees affects the individual's ability to fly in dim light condition (Kapustjanskij et al, 2007). This was not the case for our study as individuals likely flew towards a contrast in light rather than only using the surrounding light intensity to enable flight.

From these results it can be concluded that the orchid bees body size is not correlated to the ability to negotiate holes. For bumblebees there were no general correlations found. Nevertheless the eye length could be of some interest as it possibly could result in an off-centred flight but further studies would be required to investigate this.

Habitat induced traits

The ability to negotiate holes is different between bumblebees and orchid bees. Orchid bees are able to more effectively negotiate holes than bumblebees, they are also able to negotiate through smaller holes and in dimmer light conditions. However, as orchid bees live in a complex and relatively dim habitat with many obstacles it would be important to be able to efficiently negotiate its environment. The bumblebees are in this regard very different, as their habitat consists of more open areas where an enhanced ability to negotiate obstacles in dim light may not be so advantageous. It is therefore interesting to consider if the orchid bees are more evolved than the bumblebees or if the bumblebees negotiation abilities has diminished to reinforce other abilities.

Overall, the results of this study show that orchid bees are more efficient flyers than bumblebees and individuals tend to negotiate closer to the safe point in low light intensities. We also find that in bumblebees body size does not seem to be correlated to the ability to negotiate holes. But, interestingly, variations between the left and right eye are present between individuals. This could indicate a dominant eye or an off-centered flight pattern for the individuals where one eye is larger than the other. However, to establish or reject this possibility would require further studies.

Conclusion

For an insect to be able to navigate through its environment light is of great importance (Warrant, 1999). High input of light allows individuals´ to successfully and speedily negotiate through various types of holes. Hymenopterans navigate through its environment using optic flow and by centralising when flying through tunnels (Srinivasan et al., 1996, Baird, Dacke, 2012). The ability to fly through holes in dim light conditions is to an extent contrived from habitat selection, where ecological factors has determined to what extent evolutionary traits are expressed. Our findings show that orchid bees are more successful at negotiating holes than bumblebees. Furthermore bumblebees and orchid bees generally keep closer to the safe point when negotiating holes in dimmer light conditions. Individual negotiation does not seem to be affected by individual body size. However, to determine the possible importance of individual body size and if variation in eye length could have an influence on collision avoidance, further investigations are needed.

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