Chapter

Effect of Camera Illumination on Flashing Behavior of *Pteroptyx malaccae* (Coleoptera: Lampyridae)

Anchana Thancharoen and Sirima Masoh

Abstract

Pteroptyx malaccae is a synchronous firefly that is important in firefly tourism in Thailand. Without well-managed tourism, the fireflies have faced to the problems of shooting camera flashes from tourists. Although the effect of artificial light was well understood, which causes negative impact to firefly courtship, there is no obvious information on the effect of the camera illumination. The experiment of testing four types of camera illumination was set up in laboratory using wild populations of *P. malaccae*. The flash patterns were recorded by videotaping and analyzed by using TiLIA software. The results showed that all kinds of camera illuminations affect flashing behavior of the fireflies. They prolonged flash interval by increasing pulse duration. The flashes from smartphone camera displayed the strongest effect; however, all flash types did not influence on the firefly life span, mating behavior and oviposition behavior of the fireflies.

Keywords: Lampyridae, synchronous firefly, firefly tourism, camera flashes, light pollution, TiLIA software, camera photography

1. Introduction

Recently, artificial night lighting occurs commonly in many urban areas and can be light pollution that influences negatively in many ways, i.e., waste the energy for the pollution production, causing sky glow, creating light trespass, and building glare [1] including causes of ecological effects on many organisms [2–6]. The night lighting changed unnaturally the innate behaviors of the organisms by reducing foraging behavior, predatory behavior, metabolism, growth rate, and reducing population numbers finally. Additionally, many species of insects were unavoidably trapped by phototaxis ability [4, 7, 8] including fireflies. Unfortunately, the fireflies' bioluminescent courtship signals were likely interfered [4].

The effect of artificial light on fireflies was studied in many firefly species. Under the light pollution, the abundance of fireflies displayed lower numbers than without lighting [4, 9]. Several firefly researchers have considerably examined how the light pollution impacts on the bioluminescent insects especially their courtship and reproduction. The flooding of light altered mate location behavior of *Lampyris noctiluca* males by avoiding seeking females in the lighted background areas where the female glows were less attractive [10]. It decreased the flashing activity of *Photuris versicolor* and mating success in *P. pyralis* [4]. The prolonged courtship time during dorsal mounting posture was significantly observed in *Sclerotia aquatilis* that probably results in failure of mating [11]. However, the sexual communication of some firefly species could happen in light-polluted area. The males of *A. ficta* shifted the flash signals to attract females by increasing light intensity and flash frequency in *A. ficta* [12]. Nevertheless, the mating success was not discussed. The variation of the impacts might depend on lighting types and firefly species.

Camera illumination or flash photography is another type of man-made light that is commonly used. Although the cameras do not produce long steady light, the blight flashlight probably influences animal behaviors. The effects of flash photography on organisms have been little studied. The flash testing on seahorse behaviors underwater was done and showed no significant long-term consequences [13]. The nocturnal insects such as fireflies have never been investigated.

Congregation of male *Pteroptyx* fireflies exhibited impressively synchronous courtship flashes to attract the females perching on mangrove trees. Many *Pteroptyx* habitats have been developed to be tourist attraction sites and improved local people's income in Thailand. Many consequences of firefly tourism have been considerably occurring in Thailand because of lacking knowledge of firefly ecotourism management [14]. Using flash photography of tourists might be adverse to firefly behavior, while the actual impacts have remained unknown.

In the present study, we examined the effects of camera illumination on flashing behavior of male and female *P. malaccae* under laboratory condition. Four treatments of commonly used flash photography were tested. We analyzed the fireflies' flashes by using computer software, time-lapse image analysis (TiLIA), to investigate actually the abnormal flashing behavior to expect the effect of the light on the real behavior of the fireflies in nature.

2. Materials and methods

2.1 Study species

2.1.1 Taxonomy

P. malaccae is one of more than 18 described *Pteroptyx* species distributed mostly in the region of Southeast Asia (from Boneo, Cambodia, Malaysia, Philippines, Sulawesi, and Thailand) through Australia. Their major characters for species identification are yellowish brown elytra with black deflexed elytral apices, pale brown abdominal ventrites, and dark brown head and antennae. Significantly, their males have metafemoral combs (MFC) on metathoracic legs, bipartite light organ in ventrite 7, and no lobes along the posterior margin of tergite 8 [15, 16]. From reference [17], they found morphological variation among *P. malaccae* specimens in different regions and classified them into four groups. In the group occurring in Thailand, the males have been sculpturing at the apices of the posterolateral projections (PLP) on ventrite 7. The females have similar color patterns as the males but have normal straight wing apices.

2.1.2 Ecology

P. malaccae inhabited in mangrove forest and backish water area. The larvae live on moist soil or mud and predate mangrove snails for food. At nighttime, the adults congregate on mangrove trees, or other vegetations grow nearby and flash synchronously to display their advertising flashes to their mates. *P. malaccae* is the most common synchronous species found in Thailand when compared with other three *Pteroptyx* species (*P. valida*, *P. tener*, and *P. asymmetria*). In many observations *P. malaccae* species in Thailand are usually found in the same colony with small numbers of *P. valida*. The identification between these two sympatric species can be done by both morphology and flash pattern (**Figure 1**). The flashes of *P. malaccae* are regularly shorter and faster (0.08–0.12 s of pulse duration with about 0.45 s interval), while *P. valida* flashes are brighter and longer (0.2–0.8 s of pulse duration with about 0.7–6 s interval) [18].

2.1.3 Firefly collection and maintenance

Two hundred adults of *P. malaccae* were collected from mangrove trees along Bangpakong riverbank, Chachoengsao province, after sunset time by using sweep nets. The females are difficult to observe because of having small light and low active behaviors; the proportion of the captured females was commonly lower than males. The sweeping at the low level of vegetation area of the mangrove trees where the females distribute preferably could capture the considerable numbers of them. The sexes of the fireflies were separated to maintain virgin status of adult females as possible. They were feeding on 10% honey solution to be healthy and have long life span.

Because of the congregation behavior of *Pteroptyx* fireflies on mangrove trees, four males and one female were randomly put in the same experimental box to observe mating communication between sexes and synchronous flashing behavior among males. The small group of males could display synchronous flashing behavior. Before experimental testing, each set of fireflies were randomly grouped in a 7.10 × 11.04 × 6.50 cm of transparent plastic box with a small moist cotton and allowed them to have an adaptation period for 15–30 min before starting the experiment.

2.2 Experimental design

Four treatments of different light sources of camera illumination were set up in laboratory: (1) a white flash from smartphone camera (SC), Samsung Galaxy Note 3; (2) a white flash from digital camera (DC), SLR Olympus TG 4; (3) red light for autofocus assist before a white flash from digital camera (RDC), Sony Exmor R; and (4) no flash (control). These treatments were selected from the representative flash characters found in firefly photography.

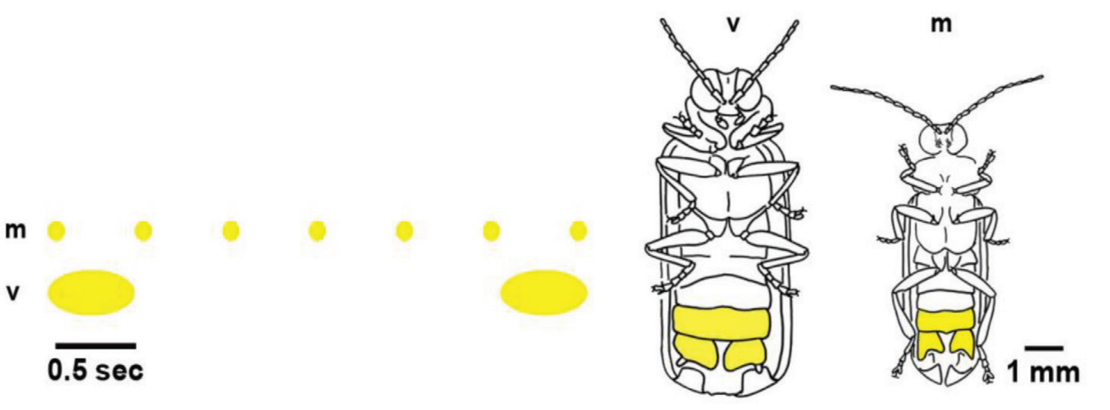

Figure 1.
Comparison of the sympatric firefly species, P. valida (v) and P. malaccae (m).

The apparatus of the experiment were designed as in **Figure 2**. The distance between the illumination point and the tested fireflies was 2 meters that is the possible distance in the real photography situation of tourists. The firefly flashing behavior was videographed using Sony Handycam™ digital camera recorder (HDR-SR11E) for 20 min in each replication (10 replications per treatment).

In the real cases of firefly photography by tourists, the fireflies would face with sequences of camera illumination; the tested fireflies were experimentally exposed to 3-time flashing to examine a consequence of sequential flashing. The flashes were shot after 5 min in the beginning of the experiment and were shot 3 times with 5-min intervals. The fireflies were continually observed and allowed to lay eggs after finishing the experiment. The adult lifespan and number of the eggs were recorded. The fireflies were tested within four nights after collection to avoid the errors from weakness and aged adults.

2.3 Flash analysis

The one of four males in each experimental set were randomly selected to analyze flash patterns. The video files were converted to Audio Video Interleave or .AVI format file to analyze by using time-lapse image analysis (TiLIA), a free software package for signal and flight pattern analyses of fireflies (available at Google Drive: https://drive.google.com/open?id=0B2o7FRVs2VohMmx2QzBVX3ZDeDA) [19]. Each video frame was converted to Tagged Image File Format (.TIFF) at each 0.03 sec. The light organs of all fireflies were defined as area of interest (ROIs) to measure the light intensities. The outputs (time and light intensities) were exported to Microsoft Excel® for flash parameter calculation applied from reference [20] (**Table 1**).

The flashes during courtship time or during synchronous flashing of the tested males were selected for flash parameter analysis by counting numbers of frames occurred in each flash parameter—pulse duration, interpulse duration, and flash interval—and then converted to second time unit.

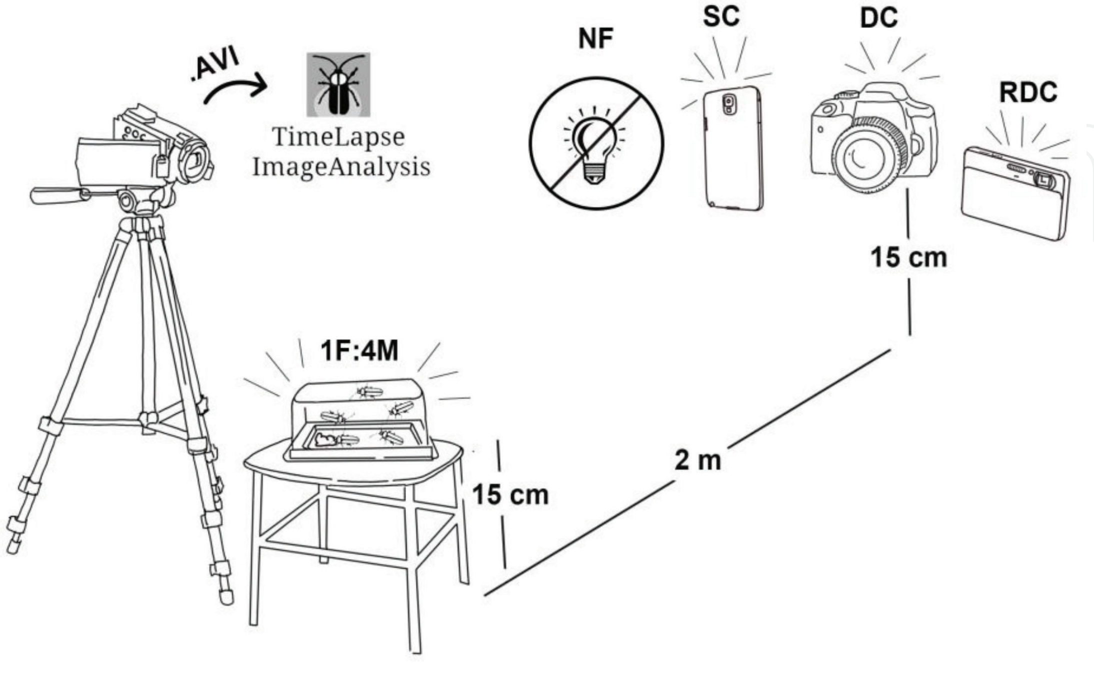

Figure 2.
Experimental apparatus.

Flash parameters	Explanation
Pulse duration	Duration between the beginning flash to the end of the flash (light period)
Interpulse duration	Duration between two pulse durations (dark period)
Flash interval	Summarizing of pulse and interpulse durations
Flash rate	Number of flash per time

Table 1.
Terminology of flash signals.

2.4 Statistical analysis

Thirty flashes of males in each treatment were statistically analyzed. The comparison of three types of flash parameters among camera illumination treatments and among different time sequences was statistically analyzed using one-way ANOVA and Tukey multiple comparison test. The flash rate of smartphone treatments was compared with control group by using independent t-test. A value of $p < 0.05$ was considered statistically significant. All statistical analyses were performed using SPSS program version 24.

3. Results

3.1 Flash parameters of *P. malaccae* males

A thousand and 33 flashes from 30 males in the control group were analyzed for flash parameters of *P. malaccae.* The *P. malaccae* males displayed variation of flash patterns. Their pulse duration could be in the range of 0.06–0.24 with 0.15–0.85 s of interpulse duration (**Table 2**). There was no correlation between pulse and interpulse duration (Pearson correlation coefficient $r = 0.033$, $P = 0.288$, n = 1023).

3.2 Effect on flash parameters

The flashing behavior of *P. malaccae* fireflies is behavioral plasticity; they could change their flash signals in response to the artificial light stimuli. From the results of flash parameter analysis, the artificial illumination from all camera flash types caused flash parameter plasticity by extending pulse duration significantly ($F = 68.461$, $df = 3$, $P = 0.000$), SC > DC > RDC > NF, and resulted in prolonging a long duration of interpulse found only in the case of smartphone camera flash ($F = 44.494$, $df = 3$, $P = 0.000$) (**Figure 3**). These resulted in longer flash interval of the smartphone treatment than the normal flash patterns. The tested fireflies in smartphone flash treatment displayed extremely slow flash rate when compared with the control group ($t = -6.346$, $df = 1682$, $P = 0.000$).

The results of the study showed that camera flashes affect temporarily the flashing behavior of *Pteroptyx* fireflies. After shooting all types of the camera flashes, most male fireflies displayed paused flashing for a period (approximately 3 sec) and then become flashing normally. The levels of sensitivity depended on flash types; smartphone camera has the highest effect (60%) followed by the digital camera with red light for focusing (30%) and digital camera (10%), respectively. Although the fireflies were shocked by the flashes suddenly, approximately 80–90% of male fireflies in all flash treatments could display courtship flashes and posted dorsal mounting during mating behavior later. Some of the males could reveal synchronous flashing behavior, DC (30%) > RDC (20%) > SC (10%).

3.3 Different time series of the flashes

After applying each types of flash photography treatment 3 times, all flash parameters in different time series were analyzed and compared in each flash type. The DC and RDC treatments displayed fluctuation of pulse duration but did not quite differently before shooting flash (**Figure 4a**). On the other hand, SC displayed an increasing trend of pulse duration (F = 62.899, df = 3, P = 0.000). The pulse duration after the last exposure of flash was the highest at 0.15 ± 0.00 s.

The flash series did not affect interpulse duration and flash interval of DC, although they could cause a small fluctuation in RDC. Alternatively, SC increased interpulse duration and flash interval after receiving the first flash (F = 34.345, df = 3, P = 0.000; **Figure 4b**) before flashing normally when received later flashes.

3.4 Female responses

The female flashes are not likely the answer response to the male flashes. They glowed irregularly. Although we could not examine the effect of the camera flashes on the female receptivity definitely, the female flashes were also recorded. The

Flash parameters	Mean ± SE
Pulse duration	0.10 ± 0.02
Interpulse duration	0.41 ± 0.01
Flash interval	0.51 ± 0.01
Flash rate (per sec)	1.97 ± 0.03

Table 2.
Flash parameters of P. malaccae.

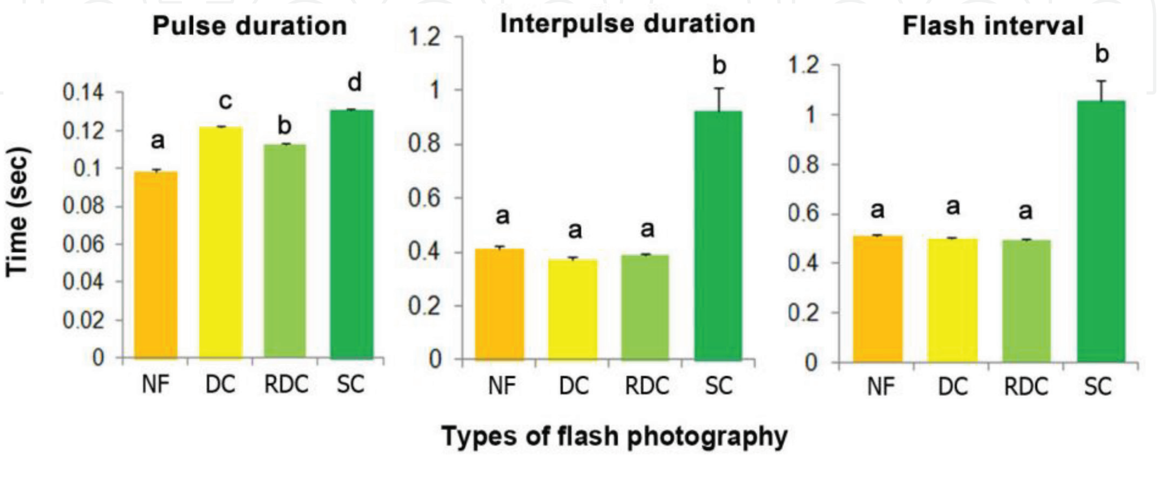

Figure 3.
Comparison of flash parameters (±SE) of P. malaccae after receiving different types of flash photography, no flash or control (NF), digital camera (DC), digital camera with red light autofocusing (RDC), and smartphone camera (SC). Letters indicate significant differences among different types of camera flash treatments.

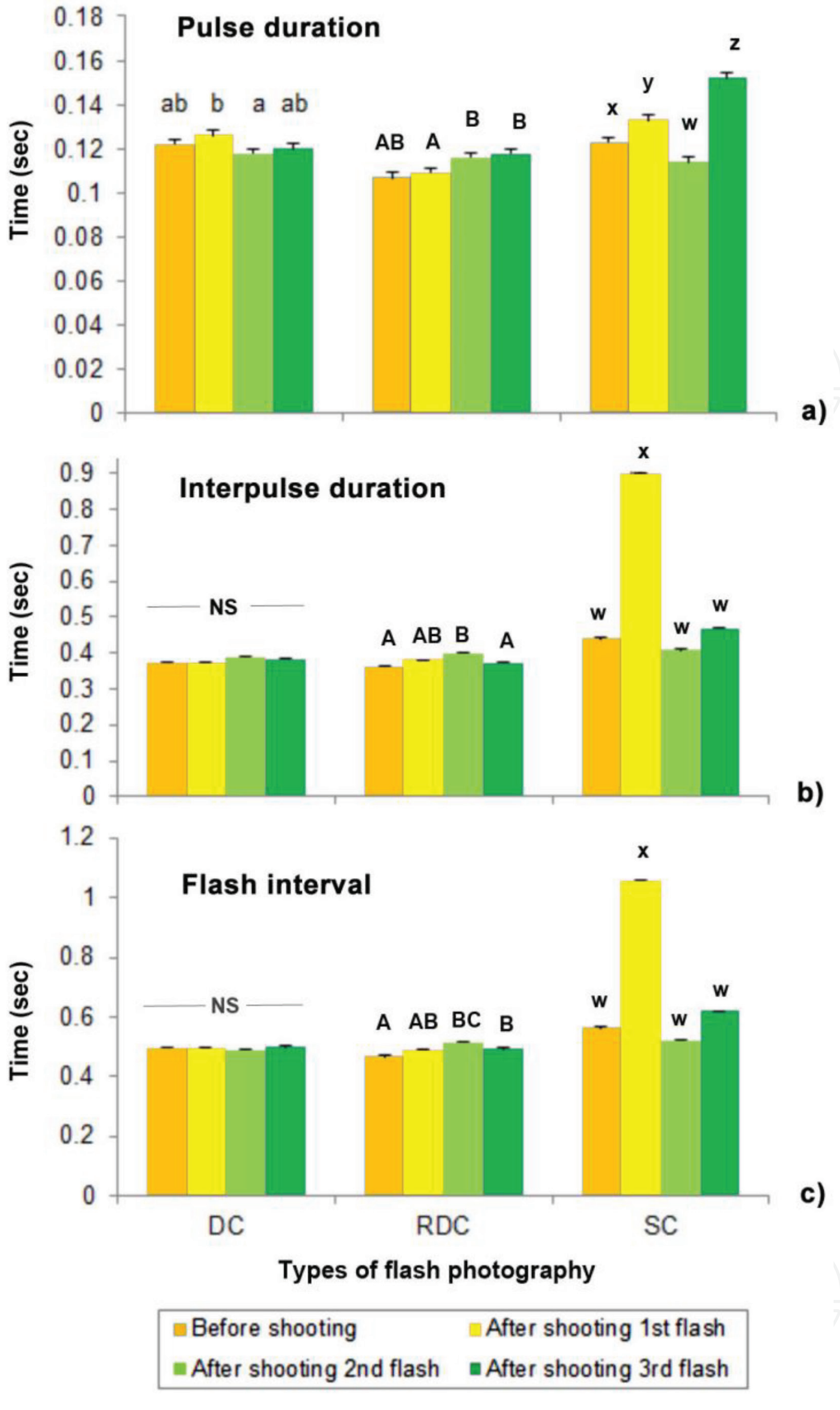

Figure 4.
Comparison of flash parameters (±SE), pulse duration (a), interpulse duration (b) and flash interval (c) of P. malaccae when a set of all camera flash types, digital camera (DC), digital camera with red light autofocusing (RDC), and smartphone camera (SC) is received. Letters indicate significant differences among different sequences of flashing time of each flash photography treatment. NS represents nonsignificant.

illumination from the smartphone caused stopped female flashes with the highest percentage (40%) followed by RDC (20%), while the females in control and DC treatment displayed normal flashing behavior. The observation of oviposition behavior after the shooting flash experiment showed that females from all treatments could lay eggs. Although the females might mate with other males before testing, they can express egg oviposition behavior after receiving flash effect. The fireflies in all flash type experiments can lay eggs for 40–50% with the maximum numbers of eggs, 40–67.

4. Discussion

The experiment of testing camera illumination was carried out using adult *P. malaccae* captured from nature that has no information of age and mating status. Some females probably already mated, and they did not respond to the male flashes and did not show mating posture during the experiment. Although female fireflies could remate after 24 h of first mating examined in *Photinus greeni* [21], there are no reports in *Pteroptyx* species. Thus, it was difficult to interpret the effect of camera illumination on female receptivity. I recommended that preparation of virgin females from culturing for the testing might be the reliable methods to provide the female response signal and mating success data. The reference [22] indicated that the flash patterns of the *Pteroptyx* fireflies in flight and on perch were different. The synchronous flashing on perch acts as attraction signals to make the aggregation groups of the species and might be important as courtship signals. Thus, this study analyzed mainly the signals of synchronous flashes. The adding of females in our experiment helped to induce males to produce the courtship flashes. Most tested males displayed courtship flashes normally. From the results, the flashes likely affect the flash synchronization that is a specific behavior of *Pteroptyx* species. There are many hypotheses of displaying the synchronous behaviors that are (1) attractive communication in dense vegetation environment [22] and (2) male competition to attract a female [23]. It would affect the species communication ability and destroy aggregation ability.

These results indicated that the firefly could adapt their flashing behavior when any strong interferences are encountered. The behavioral plasticity in flashing communication of fireflies was discussed for a long time. Although the flash codes are species specificity, fireflies can change their flash codes depending on the situation and environmental conditions [12, 24]. The plastic behavior might benefit males by increasing their advertisement to females over artificial lighting interference. *Aquatica ficta* male could change their flash signals under different ambient light intensities and different wavelengths of the light [12]. The short wavelength light induced *A. ficta* male flash brighter and slower. Additionally, the firefly could adapt their flashing behavior when they are exposed to the gradient of light (from dim to bright) by increasing their light intensities, while they displayed no flashes when they confronted with the bright light and slightly inhibit the flashing behavior which even appeared in dim environment later.

In the experiment, the tested females did not respond specific flash signals to the males that might cause from mated status of the females or captive condition. Reference [2] reported that the captive *Photuris* female did not respond to any males even the artificial males. The captive status might affect female response in this study.

The camera illuminations produced white light that are a broad spectrum composed of both short and long wavelengths of light. In this study, the response of male fireflies was quite similar to the effect of short wavelength light studied by the researchers [12]. The main biological effect of the light might come from the short wavelength light. All experimental light types were mainly LED flash technology varied with light intensities and function system. The detail of light spectrum was not described. The smartphone illumination was the brightest type that showed the strongest effect when compared with DC and RDC. The fireflies received SC flashes and showed twice the time duration of flash interval than control.

Currently, smartphone photography has become commonly used. The smartphone flashes are considerably developed to improve night shot photographs such as dual flash and Xenon flash that are stronger than LED and might cause adverse impact on the firefly behavior. The peak wavelength of LED and xenon flashes is in the blue region of visible light (400–480 nm) that could adverse biological effects including human eyes and skin when receiving long exposure [25]. The toxic effects of short wavelength light on many insects are known, i.e., mortality in immature stages of *D. melanogaster* [26] and strawberry leaf beetle [27]. In case of fireflies, the short wavelength of light caused flash signal alteration mentioned by the researchers [12]. However, the light from high-pressure sodium street lamps (with peak at low short wavelength) also likely deterred males to locate females; the males preferred to focus on females in darker areas [10]. Similarly, *Pteroptyx* fireflies showed adaptive behavior to avoid receiving the light by staying above the lamp level or staying in darker areas of the trees, even though it might have long-term effects or bring populations down and finally disappear from the habitats. From the field observation of local people in Chao Phraya riverbank area, the *Pteroptyx* fireflies stopped flashing after receiving camera flashes from tourists, moved away from the light side and could not be observed the firefly colonies back for long period up to 3 months. People wondered that the camera illumination might result in firefly mortality. As in the results of the current study, the tested fireflies in the experiment could prove that the temporary receiving of camera illumination did not cause firefly deaths. The life span of the adults is not different from control group. Additionally, the females could display egg oviposition behavior normally.

Timing of receiving flashes is probably an important factor. The tested fireflies were exposed to the camera illumination in the beginning of mating sequences. The 80% of experimental treatment could observe paired fireflies after shooting flashes. It is also possible that the copulation could not continually happen if they are interrupted during the sensitive steps of their mating sequences, i.e., dorsal mouthing. The females might remain for a long time in dorsal mouthing posture as in mating under light pollution of *Sclerotia aquatilis* [11]. Adversely, the females will not allow the male to mate with those that are often found naturally (**Figure 5**).

To conclude, any types of the camera illumination influenced *Pteroptyx* firefly behavior, i.e., mating behavior interruption, courtship signal alteration, synchronous flashing that probably influences female reception. The impact levels depend on illumination types related to light intensity, wavelength, etc. However, the fireflies could have adaptive behavior to avoid the effect after receiving the first trial; however, the repeated exposures are possibly resulting in decreasing numbers and becoming extinct in the natural habitats. Although firefly photography is not good

Figure 5.
The courtship behavior of P. malaccae in nature. The male displayed dorsal mounting on female back and flashed to her eyes. The female might reject the male during the process. Photo: Mr. Banthoon Phankaew.

when using flash illumination, many tourists preferred to use it anyway. Our finding suggests that the impact of understanding would be a useful knowledge for raising public awareness in firefly tourism activity. Besides the effect from camera illumination, the other sources of light should be concerned for firefly conservation.

5. Conclusions

Studies on the effect of artificial night lighting on fireflies have been demonstrated for concerning the effects on firefly populations and their conservation for a decade. However, they have still little understood in particular species and some different types of lights.

The book chapter has presented the impact of artificial light focusing on flash photography from both cameras and smartphones that probably have an impact on *P. malaccae*; the tourism highlighted species in Thailand and also other countries. The results indicated that the flash types could affect their courtship behavior, especially the flash from smartphones or from the brighter sources. The flash photography might bring vulnerability to wild *Pteroptyx* populations especially to the females. The consideration on firefly tourism rules about using camera flashes are needed to protect *P. malaccae* populations in tourism habitats.

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Conflict of interest

The author declares no conflict of interest.

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