

1           **Phototactic response of the oriental armyworm, *Mythimna separata***  
2           **(Lepidoptera: Noctuidae), to light-emitting diode lights of different**  
3           **wavelengths**

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## 13      **Abstract**

14      Recently, light traps using light-emitting diode (LED) lights have been applied to monitor or  
15      control insect pests. The oriental armyworm, *Mythimna separata* Walker, is an important insect  
16      pest that has caused damage to several cereal crops, including corn, wheat and rice. The present  
17      study aims to seek out a sensitive wavelength causing high phototactic response in *M. separata*.  
18      The study evaluated the phototactic responses of *M. separata* moths to several LED lights of  
19      different wavelengths and luminance intensities under laboratory condition. Results showed  
20      that green (520 nm) LED light resulted in significant phototactic response of *M. separata*  
21      moths compared to LED lights of other wavelengths. Additionally, the highest attraction rate of  
22      the moths to green LED light appeared in luminance intensity group of 200 lux compared to the  
23      other intensities groups. Experiments under optimum conditions based on the above  
24      experiments revealed that the green LED light exhibited the strongest attraction rate (64.44%)  
25      among all experimental groups. An experiment performed in a net cage also showed that green  
26      LED light resulted in the highest phototactic response of *M. separata* moths, 1.7 times more  
27      than a commercial black light used as control. These findings clearly demonstrate that *M.*  
28      *separata* moths have a high sensitivity to the green LED light. Therefore, a light trap equipped  
29      with green LED light could be useful for monitoring and controlling *M. separata* moths.

30      **Key words:** phototactic response; *Mythimna separata*; LED; wavelength; attraction rate;  
31      luminance intensity; sensitivity

## 32 1. Introduction

33 The oriental armyworm, *Mythimna* (or *Leucania*) *separata* Walker (Lepidoptera:  
34 Noctuidae), is an important long-migratory pest in eastern Asia, including the Korean  
35 peninsula, China and Japan, and causes damage to several cereal crops, such as corn, wheat  
36 and rice; in addition to these major crops, the pest has damaged more than 300 kinds of  
37 agricultural and industrial plants in nearly 100 families, thus causing substantial crop  
38 production losses annually [1-7]. Widespread outbreaks of *M. separata* are related to  
39 climate change, especially increases in temperature [8]. The conventional control of this  
40 insect pest depends on synthetic chemical insecticides. However, the excessive use of the  
41 chemical insecticides has resulted in problems, such as resistance of the pest to the  
42 chemical insecticides, harm to users and eco-system pollution [9,10]. Recently, there has  
43 been increasing pressure to search for safe and more efficient alternative pest control  
44 methods for the management of *M. separata* [7].

45 Many insects have a phototactic behavior to an artificial light at night. Therefore, light  
46 traps have been widely applied to management of agriculture and forest insect pests having  
47 phototaxis. Pest control by use of light traps, which does not negatively impact eco-system  
48 conservation and human health, could be an alternative technique used for controlling  
49 insect pests in modern organic agriculture [11,12]. In recent years, light traps equipped  
50 with light-emitting diode (LED) lights, as the light source, have been applied to monitor or  
51 control insect pests [13-15]. LED lights have some benefits, including low electricity  
52 consumption, the ability to selectively emit wavelengths, the adjustability of light intensity,  
53 high emitting effect, long use time, high mechanical stability, small size and low weight

54 [13,16]. During the last decade, some studies determined the wavelengths of LED lights  
55 that strongly attract some insect pests for using as light sources in light traps for these  
56 insect pests [17].

57 Light sensitivities of insects vary in responses to different wavelength lights. In  
58 addition, the insect's phototactic behavior is influenced by several factors, such as  
59 luminance intensity, light exposure time, insect physiological status, temperature, relative  
60 humidity, wind direction and wind strength [11,18,19]. Some studies have shown that some  
61 insect pests are more sensitive to certain wavelengths of LED light sources. The spiraling  
62 whitefly, *Aleurodicus dispersus* Russell (Hemiptera: Aleyrodidae), was most sensitive to  
63 violet (405 nm) light emitted by LED light [15]. The cigarette beetle, *Lasioderma*  
64 *serricorne* Fabricius (Coleoptera: Anobiidae), was most sensitive to ultraviolet (UV 373  
65 nm) and blue (470 nm) LED lights [20]. Green (526 nm) LED light yielded the most  
66 phototactic responses from the whitefly species, *Bemisia tabaci* Gennadius (Hemiptera:  
67 Aleyrodidae) and *Trialeurodes vaporariorum* Westwood (Hemiptera: Aleyrodidae) [21].  
68 The Indian meal moth, *Plodia interpunctella* Hübner (Lepidoptera: Pyralidae), was most  
69 attracted to ultraviolet (UV 395 nm) LED light [22]. Thus, the insect phototactic responses  
70 represent a priority research topic in the promotion of physical pest control.

71 There have been few studies on the phototactic response of *M. separata* moths to light  
72 sources, especially *M. separata* responses to specific wavelengths and light intensities. The  
73 present study aimed to find a sensitive wavelength causing a strong phototactic behavior in  
74 *M. separata* moths. We evaluated the phototactic response levels of *M. separata* moths to  
75 several wavelengths, luminance intensities, and light exposure times by using LED lights

76 and a Y-maze bioassay chamber under laboratory condition. Then, the study reevaluated the  
77 phototactic responses of *M. separata* moths under the optimum conditions (luminance  
78 intensities and exposure times) determined by our earlier experiments. Finally, we evaluated  
79 some of the above the experimental results within a net cage condition. Our findings may  
80 provide a theoretical and scientific basis for improving the light trap technique in physical  
81 pest control of *M. separata* moths.

## 82 **2. Materials and methods**

### 83 *2.1 Experimental insects*

84 Adults of the experimental insect, *M. separata*, were reared at the temperature  $25\pm 3^{\circ}\text{C}$ ,  
85 with  $65\pm 10\%$  relative humidity (RH) and a light: dark photoperiod of 14 h: 10 h in an  
86 insect rearing room. We used an artificial diet described by Bi [23] for larvae and 10 %  
87 honeybee water was used as adult's diet. During the rearing period, all insects were  
88 maintained with no exposure to any chemicals. Only three-day-old *M. separata* moths were  
89 used for the phototactic response experiments.

### 90 *2.2 Y-maze bioassay chamber*

91 To assessing the phototactic behavioral responses of *M. separata* moths, Y-maze bioassay  
92 chambers designed by Kim et al. [9] were employed in the experiments under laboratory.  
93 The Y-maze phototactic bioassay chamber consisted of an opaque acrylic body  
94 ( $20\times 20\times 100$  cm), and two transparent acrylic walls were situated at both sides of the light  
95 side area and the dark side area above the bioassay chamber to enable the observation of  
96 the insect behavior. An insect entrance hole was located in the upper center of the insect

97 resource box, and nylon netting covered the outer light side area to prevent experimental  
98 insects from escaping. The light source was installed on opposite side of light side area in  
99 the Y-maze chamber.

### 100 *2.3 Light sources and luminance intensity measurement tool*

101 The LED lights (5 W) used for the experiment were measured by using a spectrometer  
102 (USB4H11915; Ocean Optics Co. Ltd., USA), the wavelengths and the light irradiances of  
103 these LED lights are displayed in Table 1. The phototactic responses of *M. separata* under  
104 optimum experimental conditions and the net cage condition were compared with those  
105 under commercial black light ( $365\pm 7$  nm; 18 W), which served as the control. All of the  
106 LED lights and black lights were purchased from Shenzhen New Photoelectric Technology  
107 Co. Ltd., China. The luminance intensity (lux) of LED lights was measured by using a  
108 digital lux meter (GM1020-CH-00; Shenzhen Jumaoyuan Science and Technology Co. Ltd.,  
109 China) at the established point in the center of light side area within the Y-maze chamber.

### 110 *2.4 Experiment in the laboratory*

111 The phototactic responses of *M. separata* to LED lights were evaluated in the Y-maze  
112 chamber under different wavelengths, luminance intensities, and light exposure times,  
113 based on the experimental method described by Kim et al. [9]. Prior to the experiment, the  
114 insect resource area was closed by boards separating the light side and the dark side. Each  
115 experiment group contained 30 robust three-day-old moths (15 males and 15 females).  
116 These moths were moved through the insect entrance hole into insect source box of the  
117 Y-maze chamber and were maintained in the dark for two hours. Every experiment was

118 started at 8:00 P.M.; while darkness was maintained at the dark side of the Y-maze chamber,  
119 LED light were irradiated at the light side of the chamber. After LED light was turned on,  
120 both of the separating boards were removed from the Y-maze chamber.

121 The phototactic response of the *M. separata* moths was determined based on the number  
122 of insects attracted to the light side. The attraction rate was calculated with the following  
123 equation: attraction rate (%) = (number of insects attracted in the “light side”) / (30 insects)  
124 × 100%. Each experiment was executed for one hour and was observed every 10 minutes.  
125 So, the exposure time indicates from the time turning on the light source to the observing  
126 time. To estimate a significant effect between several light sources causing high phototactic  
127 behavioral responses, we produced the optimum conditions (e.g., intensity and exposure  
128 time) according to each LED light source, which result in a strong phototactic behavior of  
129 the moths, based on the above conducted experiments, and reevaluated the phototactic  
130 behavioral responses to different light sources under the optimum conditions. After each  
131 experiment, the light and dark positions were switched in the chamber. Every experiment  
132 was carried out at a temperature  $25\pm 3^{\circ}\text{C}$ , with  $65\pm 10\%$  relative humidity (RH) in the  
133 darkness. There were three replicates for each experiment.

#### 134 2. 5 Experiment in the net cage

135 An oblong net cage 10 m long, 10 m wide and 4 m high was covered with nylon net cloth.  
136 The net cage was installed outdoors in the field; thus the darkness of the cage depended on  
137 the darkness of the outdoor condition after sunset. Two (450 nm and 520 nm wavelength)  
138 LED lights (5 W) and a commercial black light ( $365\pm 7$  nm; 18 W) were installed inside the  
139 cage. The distance between the lights was 3 m, and the height of each light was 1 m. Three

140 infrared cameras were set 70 centimeters in front of each light at the same height as the light  
141 sources. Each experimental group included 200 robust *M. separata* moths, which were  
142 released 5 hours before the experiment started in the cage. Each experiment started at 8:30  
143 P.M. and lasted two hours, and the experiments were conducted from 15 to 25 September,  
144 2018. The phototactic response of the *M. separata* moths was evaluated by counting the  
145 number of times they appeared in videos of each experiment. The videos of each experiment  
146 were observed for 10-minute intervals, and the total number of appearances of the moths  
147 over a two-hour period was determined. The experiment in the net cage was replicated five  
148 times.

## 149 *2.6 Statistical analysis*

150 We performed one-way analysis of variance (ANOVA) with the software SPSS (SPSS Inc.,  
151 Chicago, IL, USA) to compare the attraction rates of *M. separata* moths to several light  
152 sources. Values are expressed as the means and standard errors (mean $\pm$ SEM). Significant  
153 differences were analyzed using Tukey's HSD test (P <0.05).

## 154 **3. Results**

### 155 *3.1 Phototactic response of M. separata moths to LED lights of different wavelengths* 156 *under several luminance intensities*

157 The phototactic responses of *M. separata* moths to LED lights of different wavelengths  
158 (two ultraviolet LED lights, six LED lights with visual wavelengths and one white LED  
159 light) were evaluated under luminance intensities of 50 lux, 100 lux, 150 lux and 200 lux,  
160 respectively (Figure 1). The violet (400 nm), ultraviolet (UV 385 nm), green (520 nm) and

161 white (420-650nm) LED lights under a luminance intensity of 50 lux revealed high  
162 attraction rates (average attraction rates of 52.02%, 49.99%, 50.37%, and 52.35%,  
163 respectively) (Figure 1a). Additionally, the high attraction rates of the *M. separata* moths to  
164 the LED lights almost all appeared by an exposure time of 30 min or 40 min. So, attraction  
165 rates of the moths to different LED lights were assessed when the moths were exposed to  
166 different LED lights for 40 min (Figure 2). Under a luminance intensity of 50 lux,  
167 significant increases in the attraction rates over exposure time, up to 40 min, were observed  
168 for violet (400 nm), ultraviolet (385 nm), green (520 nm) and white (420-650 nm) LED  
169 lights, with attraction rate of 57.31%, 54.52%, 53.91% and 53.49%, respectively (Figure  
170 2a). No significant difference was observed among these four kinds of LED lights, but a  
171 significant difference was found between these lights and the red (625 nm) LED light  
172 ( $F=4.676$ ;  $df=8, 18$ ;  $P=0.003$ ). High attraction rates of *M. separata* moths under a  
173 luminance intensity of 100 lux were observed for the green (520 nm), blue (450 nm) and  
174 yellow (590 nm) LED lights, with average attraction rate of 54.44%, 51.11% and 48.89%,  
175 respectively (Figure 1b). The green (520 nm), yellow (590 nm), and blue (450 nm) LED  
176 lights under a luminance intensity of 100 lux and an exposure time of 40 min resulted in  
177 higher phototactic responses (attraction rates of 55.56%, 52.22%, and 51.11%, respectively)  
178 than the other wavelength LED lights, and the differences were significant compared to  
179 ultraviolet (365 nm) wavelength ( $F=2.695$ ;  $df=8, 18$ ;  $P=0.038$ ) (Figure 2b).  
180 Meanwhile, the green (520 nm) and blue (450 nm) LED lights with a luminance intensity  
181 of 150 lux yielded higher phototactic responses from *M. separata* moths (average attraction  
182 rates of 59.09% and 56.81% respectively) than the other LED light groups (Figure 1c).

183 When *M. separata* moths were exposed to LED lights of different wavelengths under a  
184 luminance intensity of 150 lux for an exposure time of 40 min, significant increases in the  
185 attraction rates were recorded for the green (520 nm) and blue (450 nm) LED lights  
186 (attraction rates of 59.14% and 58.53% respectively) compared to two ultraviolet (365 nm  
187 and 385 nm), violet (400 nm) and dark red (680 nm) LED lights, but there were no  
188 significant differences compared to the other wavelength LED lights ( $F=7.529$ ;  $df=8, 18$ ;  
189  $P=0.001$ ) (Figure 2c). The highest significant increase in the average attraction rates of *M.*  
190 *separata* moths under a luminance intensity of 200 lux was observed for the green (520 nm)  
191 wavelength (average attraction rate of 59.32%) (Figure 1d). Furthermore, the high  
192 attraction rates of the moths recorded for the green (520 nm) LED light after an exposure  
193 time of 30 min. A significant increase in the attraction rates of *M. separata* moths to nine  
194 wavelength LED lights under a luminance intensity of 200 lux and an exposure time of 40  
195 min was only found for the green (520nm) LED light ( $F=12.907$ ;  $df=8, 18$ ;  $P=0.000$ )  
196 (Figure 2d). Moreover, the attraction rate to the green LED light was the highest, with a  
197 value of 63.65%. Additionally, the attraction rates to ultraviolet (365 nm), violet (400 nm),  
198 yellow (590 nm) and white LED lights were significantly higher than that to the ultraviolet  
199 (385 nm) LED light, but those to blue (450 nm), red (625 nm) and dark red (680 nm) LED  
200 lights was no a significant difference compared to that of ultraviolet (385 nm) LED light.

### 201 *3.2 Effect of luminance intensity on the phototactic response of M. separata moths to green* 202 *(520 nm) LED light*

203 Effect of the luminance intensity on the phototactic response of *M. separata* to green LED  
204 light was evaluated under several luminance intensities with an exposing time of 40 min

205 (Figure 3). The attraction rate of the moths exposed to the green (520nm) LED light under  
206 a luminance intensity of 200 lux was significantly higher than that under a luminance  
207 intensity of 250 lux, but no significant difference was observed among the other intensities  
208 ( $F=3.417$ ;  $df=4, 10$ ;  $P=0.038$ ).

### 209 *3.3 Phototactic response of M. separata moths under the optimum conditions of each LED* 210 *light*

211 Phototactic responses of the *M. separata* moths to LED lights of ten wavelengths under  
212 these optimum conditions which produced by the above experimental results were  
213 reevaluated and compared to the response to a commercial black light used as a control  
214 (Table 2). In this experiment, the green (520 nm) LED light under optimum condition  
215 (luminance intensity of 200 lux and exposure time of 40 min) resulted in the highest  
216 attraction rate, with 64.44%. Moreover, it was 1.38 times more attractive than control  
217 (black light; 46.67%). The number of moths attracted to the light side of the green LED  
218 light was significantly higher than those of the other LED lights and control ( $F=5.408$ ;  
219  $df=10, 22$ ;  $P=0.001$ ). A significant difference in the dark side was recorded between two  
220 LED lights (590 nm and 625 nm) and the other LED lights ( $F=5.035$ ;  $df=10, 22$ ;  $P=0.001$ );  
221 however, there were no significant differences relative to the control. The ratio of moth's  
222 numbers in the light side and the dark side was the highest for the green LED light among  
223 all the experimental groups and control.

### 224 *3.4 Phototactic response of M. separata moths under the net cage condition*

225 The attraction effect of *M. separata* moths to blue (450 nm) and green (520 nm) LED lights

226 compared with a commercial black light as a control in the net cage was evaluated (Figure 4).  
227 A significant increase in the number of *M. separata* moths that appeared in front of the light  
228 source for two hours was recorded for green (520 nm) LED light compared with the other  
229 lights, and was 1.7 times higher than the number of moths attracted to the black light as the  
230 control ( $F=74.524$ ;  $df=2, 12$ ;  $P=0.001$ ). However, a significant decrease was recorded for the  
231 blue (450 nm) LED light, which had an even lower number than the control. We analyzed  
232 the phototactic response tendencies of *M. separata* moths to different light sources based on  
233 observations 10-minute intervals for two hours in the net cage (Figure 5). The phototactic  
234 responses of the moths to three light sources in the net cage showed many complicated  
235 peaks in the time series. However, a higher rate of phototactic responses occurred for the  
236 green LED light compared with the other lights. Additionally, relatively high attraction  
237 effects were recorded after the illumination exposure time of 30 min, and again decreased  
238 after 110 minutes.

#### 239 **4. Discussion**

240 Many insects exhibit phototactic behaviors, such as positive phototaxis (attraction to light  
241 sources) or negative phototaxis (repelled by light sources). The wavelengths of light  
242 sources causing the phototactic behavior are species-specific in insects. Furthermore, the  
243 phototactic response level is influenced by the wavelength, luminance intensity and  
244 exposure time of the light source [17,24,25].

245 Our results confirmed that the phototactic response level of *M. separata* moths depends  
246 on the specific wavelength, luminance intensity and exposure time of the light source.  
247 Some studies have evaluated the phototactic responses of several species in the

248 Lepidoptera to different LED light wavelengths. The attraction rate of the tobacco cutworm,  
249 *Spodoptera litura*, to green (520±5 nm) high-powered LEDs under 40 lux and an exposure  
250 time of 60 min was highest than those to other wavelengths [26]. The Indian meal moth, *P.*  
251 *interpunctella* has the most sensitivity to violet (405 nm) LEDs based on an  
252 electroretinogram recording test and behavior experiment [27]. However, in other study, *P.*  
253 *interpunctella* moths were most attracted to green (520±5 nm) LEDs under 60 lux and an  
254 exposure time of 30 min [28]. In addition, the oriental fruit moth, *Grapholita molesta* was  
255 significantly more attracted to violet (405 nm) and green (540 nm) LEDs [29]. The  
256 diamondback moth, *Plutella xylostella*, moths were significantly more attracted to green  
257 (520±5 nm) LED than other LEDs [30].

258 Our experimental results are similar to the results of these previous studies. When *M.*  
259 *separata* moths were exposed to LED lights under a luminance intensity of 50 lux, these  
260 moths preferred the violet (400 nm) LED light. However, under luminance intensities of  
261 100 lux to 200 lux, the moths were more attracted to the green (520 nm) LED light  
262 compared with the other LED lights. Additionally, the results of the phototactic response  
263 experiments with *M. separata* moths exposed to the optimum conditions for each LED  
264 light showed that the green (520 nm) LED light resulted in the highest attraction rate.  
265 Moreover, the experimental data in the net cage condition also was similar with the results  
266 in the laboratory.

267 Most insects have three types of photoreceptors that are sensitive to UV, blue and green  
268 wavelength ranges, which is generally the case for pest moths [27,31,32]. A study on the  
269 relation between light source of specific wavelength and the quantity of insects caught by a

270 light trap equipped with the light source have found that light sources of green wavelength  
271 and UV attract more individuals of the Lepidoptera than the light sources of other  
272 wavelengths, suggesting that this phenomena may be related to structure and function of  
273 compound eyes of insects [33]. Some studies reported that the 75 % photoreceptors were  
274 related with green sensitivity, and the other photoreceptors were related to UV and blue  
275 sensitivities in the tobacco hornworm (*Manduca sexta*) [34,35]. While, the beet armyworm  
276 (*Spodoptera exigua*) moths with *Se-lw* (opsin gene related to green photoreceptor) knocked  
277 down exhibited decreased phototactic behavior, suggesting that the green receptors play  
278 more critical role in the photosensitivity to an artificial light than other photoreceptors [36].  
279 Based on our results, *M. separata* moths demonstrate the highest sensitivity to the green  
280 wavelength, which may be due to the green photoreceptor in their compound eyes.

281 Attraction effect of insects may vary depending on oxidant stress level that a light source  
282 induces in the insects. One study explored the relationship between the quantities of  
283 underground pests caught by traps with light sources of different wavelengths and the oxidant  
284 stress levels of the pests exposed to these light sources; that study found that a trap equipped  
285 with a light source resulting in a weaker oxidative stress can attract more ground pests, such as  
286 cockchafers [19]. The some studies reported that the oxidative stress level in the *M. separata*  
287 moths under a green (520 nm) light was weak but was strong under UV irradiation [37,38].  
288 Therefore, *M. separata* moths may have a stronger phototactic behavior to the green light than  
289 lights of other wavelengths. However, more scientific evidence is needed to verify whether the  
290 phototactic behavior of the insects is passively produced by the light stress.

291 Color vision of many animals, including insects, aids them in locating their habitat, to

292 seek food or mates, and to avoid their enemies and rivals [39]. For example, phytophagous  
293 insects are sensitive to any wavelengths reflecting from host plants within the range of 500  
294 to 580 nm. Based on visual stimuli, insects can find host plants by cooperating with plant  
295 volatile chemical information [40,41]. Based on the suggestions of previous data, *M.*  
296 *separata* moths may be indiscriminately attracted to light or color of green wavelength  
297 range emitted by objects including non-host plants. The high attraction of *M. separata*  
298 moths to violet (which has a wavelength between that of ultraviolet light and that of visual  
299 light) LED light under a luminance intensity of 50 lux may be related to the ultraviolet  
300 photoreceptor in the compound eyes of the insect. Moreover *M. separata* moths under low  
301 luminance intensity may be more sensitive to short wavelengths.

302 Phtotactic response level to a light source in the same insect species is different from  
303 luminance intensity of the light source [17,42]. Luminance intensity causing a high  
304 attraction rate to a green light source in *S. litura* and *P. interpunctella* moths was 40 lux  
305 and 60 lux, respectively [26,28]. In our results, the phototactic response level of *M.*  
306 *separata* moths to green LED light gradually increased with the luminance intensity from  
307 50 lux to 200 lux, but the phototactic response level of the moths decreased under a  
308 luminance intensity of 250 lux. Similarly, previous studies have revealed that low  
309 luminance intensities result in an increased phototactic response of insects, but high light  
310 intensities result in a decreased phototactic response of insects [42,43]. Our results showed  
311 that the phototactic behavior of *M. seaprata* moths also demonstrated luminance intensity  
312 preferences, suggesting that 200 lux in green (520 nm) wavelength range is the optimum  
313 luminance intensity for these insects.

314 Our finding supports previous studies that insects can detect monochrome wavelengths  
315 and luminance intensities with different sensitivities and may provide a theoretical and  
316 scientific basis for improving the light trap technique for *M. separata* moths.

## 317 **5. Conclusion**

318 Many insects, including moths, are attracted to artificial light, because insects have  
319 phototactic behavioral characteristic, but their phototactic response level and sensitivity to  
320 light wavelengths varies among species. The present research revealed that *M. separata*  
321 moths demonstrated the highest phototactic response to green (520 nm) LED light compared  
322 to the other wavelength LED lights under laboratory and net cage conditions. Our finding  
323 shows that the moths can detect monochromatic wavelengths and luminance intensities.  
324 Consequently, LED light of green wavelength (520 nm) could be used as light source in the  
325 light trap for monitoring and controlling *M. separata* moths. In addition, the possibility of  
326 employing traps equipped with LED lights of specific wavelengths to control insect pest  
327 moths may be worthy of further investigation.

### 328 **Author Contributions:**

329 K.N.K., Q.Y.H. and C.L.L. designed this study. K.N.K., H.S.S. and Z.J.H. performed the  
330 experiments under the lab and net setup, K.N.K. and R.J.C. analyzed the experimental data.

331 The article was written by K.N.K. and Q.Y.H. in consultation with all authors.

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335       **References**

- 336   1.     Sharma, H.C.; Davies, J.C. The oriental armyworm, *Mythimna separata* (Walker).  
337       Distribution, biology and control: a literature review. Miscellaneous Reports(59). *Centre for*  
338       *Overseas Pest Research (UK)* **1983**, *59*, 1-24.
- 339   2.     Sharma, H.C.; Sullivan, D.J.; Bhatnagar, V.S. Population dynamics and natural mortality  
340       factors of the oriental armyworm, *Mythimna separata* (Lepidoptera: Noctuidae), in  
341       south-central india. *Crop Prot.* **2002**, *21*, 721-732.
- 342   3.     Chen, R.L.; Bao, X.Z.; Drake, V.A.; Farrow, R.A.; Wang, S.Y.; Sun, Y.J.; Zhai, B.P. Radar  
343       observations of the spring migration into northeastern China of the oriental armyworm moth,  
344       *Mythimna separata*, and other insects. *Ecol. Entomol.* **2010**, *14*, 149-162.
- 345   4.     Jiang, X.; Luo, L.; Zhang, L.; Sappington, T.W.; Hu, Y. Regulation of migration in  
346       *Mythimna separata* (Walker) in china: A review integrating environmental, physiological,  
347       hormonal, genetic, and molecular factors. *Environ. Entomol.* **2011**, *40*, 516-533.
- 348   5.     Jung, J.K.; Seo, B.Y.; Cho, J.R.; Kim, Y. Monitoring of *Mythimna separata* adults by using a  
349       remote-sensing sex pheromone trap. *Korean J Appl Entomol* **2013**, *52*, 341-348.

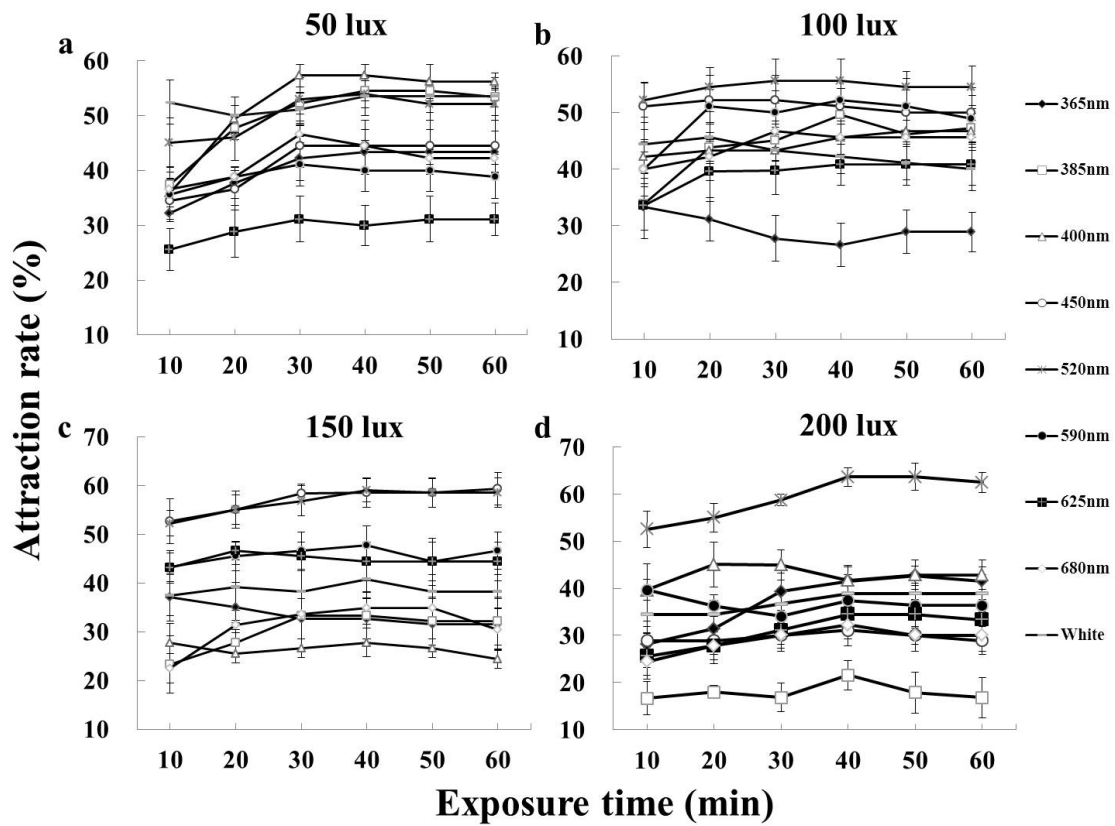
- 350 6. Liu, Y.; Qi, M.; Chi, Y.; Wuriyanghan, H. De novo assembly of the transcriptome for  
351 oriental armyworm *Mythimna separata* (Lepidoptera: Noctuidae) and analysis on insecticide  
352 resistance-related genes. *J. Insect Sci.* **2016**, *16*(92), 1-8.
- 353 7. Lihuang, K.M.; Zhang, Z.; Kim, K.N.; Huang, Q.Y.; Lei, C.L. Antennal and behavioral  
354 responses of *Mythimna separata* (Walker) to three plant volatiles. *Environ. Sci. Pollut. Res.*  
355 **2017**, *24*, 24953-24964.
- 356 8. Zhao, Z.; Sandhu, H.S.; Ouyang, F.; Ge, F. Landscape changes have greater effects than  
357 climate changes on six insect pests in China. *Sci. China Life Sci.* **2016**, *59*, 627-633.
- 358 9. Kim, K.N.; Song, H.S.; Li, C.S.; Huang, Q.Y.; Lei, C.L. Effect of several factors on the  
359 phototactic response of the oriental armyworm, *Mythimna separata* (Lepidoptera:  
360 Noctuidae). *J. Asia-Pac. Entomol.* **2018**, *21*, 952-957.
- 361 10. Song, J.; Jeong, E.Y.; Lee, H.S. Phototactic behavior 9: phototactic behavioral response of  
362 *Tribolium castaneum* (Herbst) to light-emitting diodes of seven different wavelengths. *J.*  
363 *Appl. Biol. Chem.* **2016**, *59*, 99-102.
- 364 11. Jing, X.F.; Lei, C.L. Advances in research on phototaxis of insects and the mechanism.  
365 *Entomological Knowledge* **2004**, *41*, 198-203. (In Chinese with English abstract)
- 366 12. Shimoda, M.; Honda, K.I. Insect reactions to light and its applications to pest management.  
367 *Appl. Entomol. Zool.* **2013**, *48*, 413-421.
- 368 13. Tamulaitis, G.; Duchovskis, P.; Bliznikas, Z.; Breive, K.; Ulinskaite, R.; Brazaityte, A.;  
369 Novickovas, A.; Zukauskas, A. High-power light-emitting diode based facility for plant  
370 cultivation. *J. Phys. D Appl. Phys.* **2005**, *38*, 3182-3187.

- 371 14. Cohnstaedt, L.W.; Gillen, J.I.; Munstermann, L.E. Light-emitting diode technology  
372 improves insect trapping. *J. Am. Mosq. Control Assoc.* **2008**, *24*, 331-334.
- 373 15. Zheng, L.X.; Zheng, Y.; Wu, W.J.; Fu, Y.G. Field evaluation of different wavelengths  
374 light-emitting diodes as attractants for adult *Aleurodicus dispersus* Russell (Hemiptera:  
375 Aleyrodidae). *Neotrop. Entomol.* **2014**, *43*, 409-414.
- 376 16. Yeh, N.; Chung, J.P. High-brightness LEDs—Energy efficient lighting sources and their  
377 potential in indoor plant cultivation. *Renew. Sus. Energ. Rev.* **2009**, *13*, 2175-2180.
- 378 17. Park, J.H.; Lee, H.S. Phototactic behavioral response of agricultural insects and  
379 stored-product insects to light-emitting diodes (LEDs). *Appl. Biol. Chem.* **2017**, *60*,  
380 137-144.
- 381 18. Liao, H.; Shi, L.; Liu, W.; Du, T.; Ma, Y.; Zhou, C.; Deng, J. Effects of light intensity on the  
382 flight behaviour of Adult *Tirumala limniace* (Cramer) (Lepidoptera: Nymphalidae:  
383 Danainae). *J. Insect Behav.* **2017**, *30*, 139-154
- 384 19. Gao, Y.; Li, G.; Li, K.; Lei, C.; Huang, Q. Comparison of the trapping effect and antioxidant  
385 enzymatic activities using three different light sources in cockchafers. *Environ. Sci. Pollut.*  
386 *Res.* **2017**, *24*, 27855-27861.
- 387 20. Katsuki, M.; Arikawa, K.; Wakakuwa, M.; Omae, Y.; Okada, K.; Sasaki, R.; Shinoda, K.;  
388 Miyatake, T. Which wavelength does the cigarette beetle, *Lasioderma serricornis*  
389 (Coleoptera: Anobiidae), prefer? Electrophysiological and behavioral studies using  
390 light-emitting diodes (LEDs). *Appl. Entomol. Zool.* **2013**, *48*, 547-551.
- 391 21. Jahan, S.M.H.; Lee, G.S.; Lee, S.; Lee, K.Y. Acquisition of tomato yellow leaf curl virus  
392 enhances attraction of *Bemisia tabaci* to green light emitting diodes. *J. Asia-Pac. Entomol.*

- 393           **2014**, *17*, 79-82.
- 394   22.   Kishanr, S.; Thomasw, P. Responses of adult *Plodia interpunctella* (Hubner) (Lepidoptera:  
395           Pyralidae) to light and combinations of attractants and light. *J. Insect Behav.* **2008**, *21*,  
396           422-439.
- 397   23.   Be, F.C. A new artificial diet for the army worm *Leucania separata* Walker. *Acta Entomol.*  
398           *Sin.* **1981**, *24*, 379-383. (In Chinese with English abstract)
- 399   24.   Cloyd, R.A.; Dickinson, A.; Larson, R.A.; Marley, K.A. Phototaxis of fungus gnat, *Bradysia*  
400           sp. nr *coprophila* (Lintner) (Diptera: Sciaridae), adults to different light intensities.  
401           *Hortscience* **2007**, *42*, 1217-1220.
- 402   25.   Van, L.F.; Ettema, J.A.; Donners, M.; Wallisdevries, M.F.; Groenendijk, D. Effect of spectral  
403           composition of artificial light on the attraction of moths. In *Biol. Conserv.* **2011**, *144*,  
404           2274-2281.
- 405   26.   Yang, J.Y.; Kim, M.G.; Lee, H.S. Phototactic behavior: Attractive effects of *Spodoptera*  
406           *litura* (Lepidoptera: Noctuidae), tobacco cutworm, to high-power light-emitting diodes. *J.*  
407           *Korean Soc. Appl. Biol. Chem.* **2012**, *55*, 809-811.
- 408   27.   Cowan, T.; Gries, G. Ultraviolet and violet light: attractive orientation cues for the Indian  
409           meal moth, *Plodiainterpunctella*. *Entomol. Exp. Appl.* **2009**, *131*, 148–158.
- 410   28.   Park, J.H.; Lee, H.S. Phototactic behavior 10: phototactic behavioral effects of *Plodia*  
411           *interpunctella* (Hübner) (Lepidoptera: Pyralidae) adults to different light-emitting diodes of  
412           seven wavelengths. *J. Appl. Biol. Chem.* **2016**, *59*, 95-98.
- 413   29.   Sun, Y.X.; Tian, A.; Zhang, X.B.; Zhao, Z.G.; Zhang, Z.W.; Ma, R.Y. Phototaxis of  
414           *Grapholitha molesta* (Lepidoptera: Olethreutidae) to different light sources. *J. Econ.*

- 415 *Entomol.* **2014**, *107*, 1792-1799.
- 416 30. Cho, K.S.; Lee, H.S. Visual preference of diamondback moth, *Plutella xylostella*, to  
417 light-emitting diodes. *J. Korean Soc. Appl. Biol. Chem.* **2012**, *55*, 681-684.
- 418 31. Briscoe, A.D.; Chittka, L. The evolution of color vision in insects. *Annu. Rev. Entomol.* **2001**,  
419 *46*, 471-510.
- 420 32. Diaz-Montano, J.; Campbell, J.F.; Phillips, T.W.; Cohnstaedt, L.W.; Throne, J.E. Evaluation  
421 of light attraction for the stored-product psocid, *liposcelis bostrychophila*. *J. Pest Sci.* **2015**,  
422 *89*, 923-930.
- 423 33. Jing, X. Nocturnal insects' phototactic behavior to different light and the effects of black  
424 light on the enzyme in bollworm, *Helicoverpa armigera*. *Master's Thesis*, Huazhong  
425 Agricultural University, Wuhan, China, **2004**.
- 426 34. Cutler, D.; Bennett, R.; Stevenson, R.; White, R. Feeding behavior in the nocturnal moth  
427 *Manduca sexta* is mediated mainly by blue receptors, but where are they located in the retina?  
428 *J. Exp. Biol.* **1995**, *198*, 1909-1917.
- 429 35. White, R.H.; Huihong, X.; Münch, T.A.; Bennett, R.R.; Grable, E.A. The retina of *Manduca*  
430 *sexta*: rhodopsin expression, the mosaic of green-, blue- and UV-sensitive photoreceptors,  
431 and regional specialization. *J. Exp. Biol.* **2003**, *206*, 3337-3348.
- 432 36. Liu, Y.J.; Yan, S.; Shen, Z.J.; Li, Z.; Zhang, X.F.; Liu, X.M.; Zhang, Q.W.; Liu, X.X. The  
433 expression of three opsin genes and phototactic behavior of *Spodoptera exigua* (Lepidoptera:  
434 Noctuidae): Evidence for visual function of opsin in phototaxis. *Insect Biochem. Molec. Biol.*  
435 **2018**, *96*, 27-35.
- 436 37. Ali, A.; Rashid, M.A.; Huang, Q.Y.; Lei, C.L. Influence of UV-A radiation on oxidative

- 437 stress and antioxidant enzymes in *Mythimna separata* (Lepidoptera: Noctuidae). *Environ.*  
438 *Science. Pollut. Res.* **2017**, *24*, 8392-8398.
- 439 38. Kim, K.N.; Yun, C.N.; Sin, U.C.; Huang, Z.J.; Huang, Q.Y.; Lei, C.L. Green light and light  
440 stress in moth: Influence on antioxidant enzymes in the oriental armyworm, *Mythimna*  
441 *separata* (Lepidoptera: Noctuidae). *Environ. Science. Pollut. Res.* **2018**, *25*, 35176-35183.
- 442 39. Chen, Z.; Liu, X.; Zhou, J.X.; Dou, H.; Kuang, R.P. Phototactic behaviour of *Pachyneuron*  
443 *aphidis* (Hymenoptera: Pteromalidae) – hyperparasitoid of *Myzus persicae* (Hemiptera:  
444 Aphididae). *Bio. Sci. Techn.* **2014**, *24*, 1469-1480.
- 445 40. Couty, A.; Emden, H.V.; Perry, J.N.; Hardie, J.; Pickett, J.A.; Wadhams, L.J. The roles of  
446 olfaction and vision in host The roles of olfaction and vision in host - plant finding by the  
447 diamondback moth, *Plutella xylostella*. *Physiol. Entomol.* **2006**, *31*, 134–145.
- 448 41. Blackmer, J.L.; Byers, J.A.; Cesar, R.S. Evaluation of color traps for monitoring *Lygus* spp.:  
449 Design, placement, height, time of day, and non-target effects. *Crop Prot.* **2008**, *27*,  
450 171-181.
- 451 42. Wei, G.; Zhang, Q.; Zhou, M.; Wu, W. Studies on the phototaxis of *Helicoverpa armigera*  
452 (Hübner). *Acta Biophys. Sin.* **2000**, *16*, 89-95.
- 453 43. Chen, Y.; Luo, C.W.; Kuang, R.P.; Li, H.W.; Zheng, C.; Liu, Y.J. Phototactic behavior of the  
454 armand pine Phototactic behavior of the armand pine bark weevil, *Pissodes punctatus*. *J.*  
455 *Insect Sci.* **2013**, *13*, 1-10.

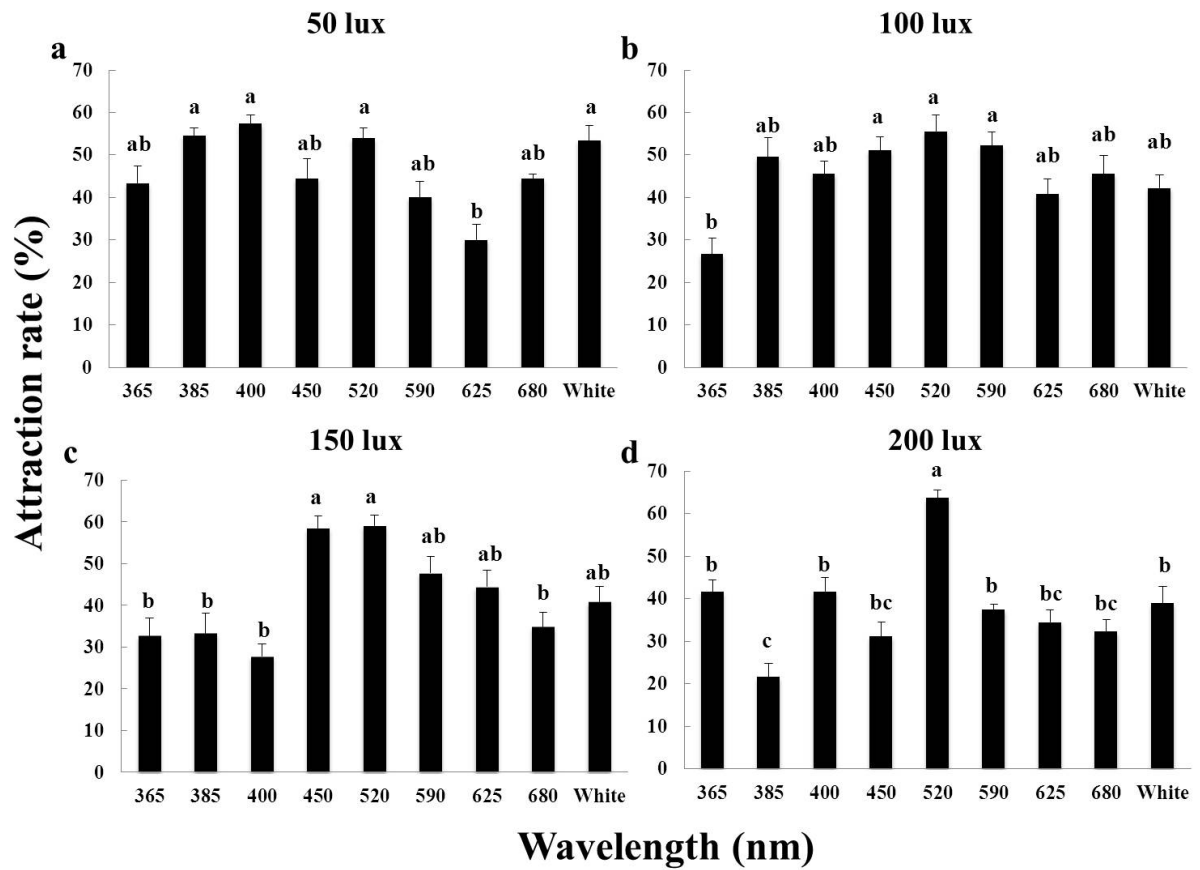
456 **Figures**

457

458 **Figure 1.** Phototactic responses of *M. separata* moths to LED lights of different wavelengths

459 under several luminance intensities. The values of the points on each line express the

460 mean, and the error bars on the points express the SEM.



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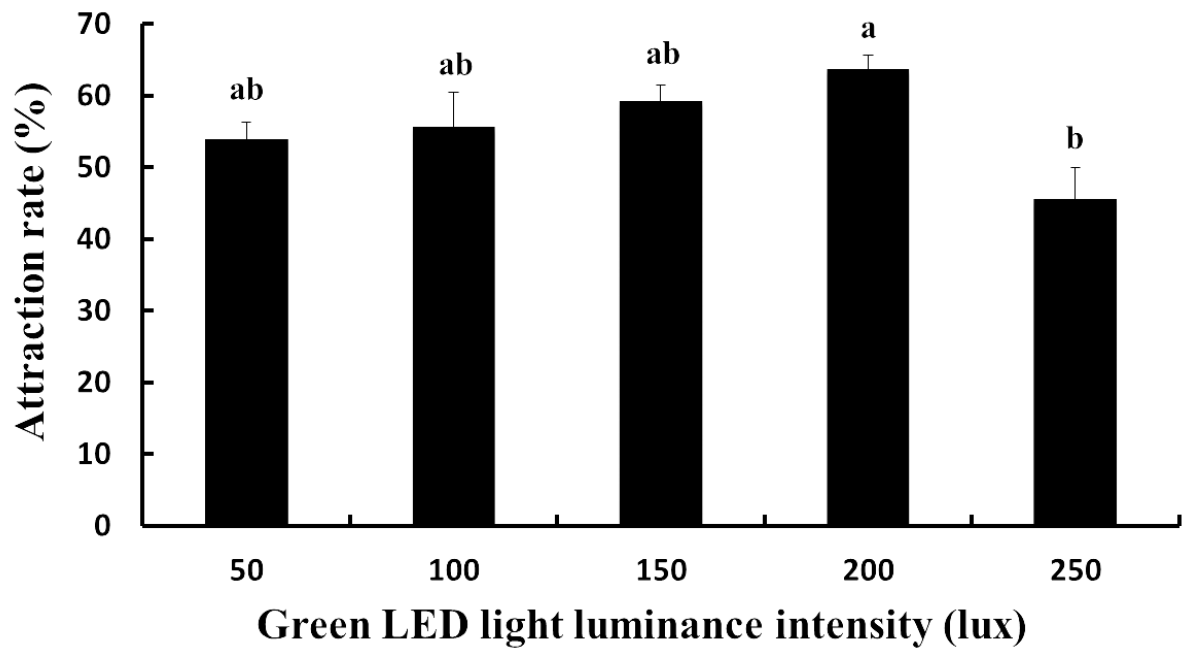
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**Figure 2.** Attraction rate of *M. separata* moths to LED lights of different wavelengths under several luminance intensities with an exposure time of 40 min. The figure was derived from Figure 1. Bars indicate the mean and error bars express the SEM. Different letters above each bar denote significant differences according to the Tukey HSD test ( $P < 0.05$ ).



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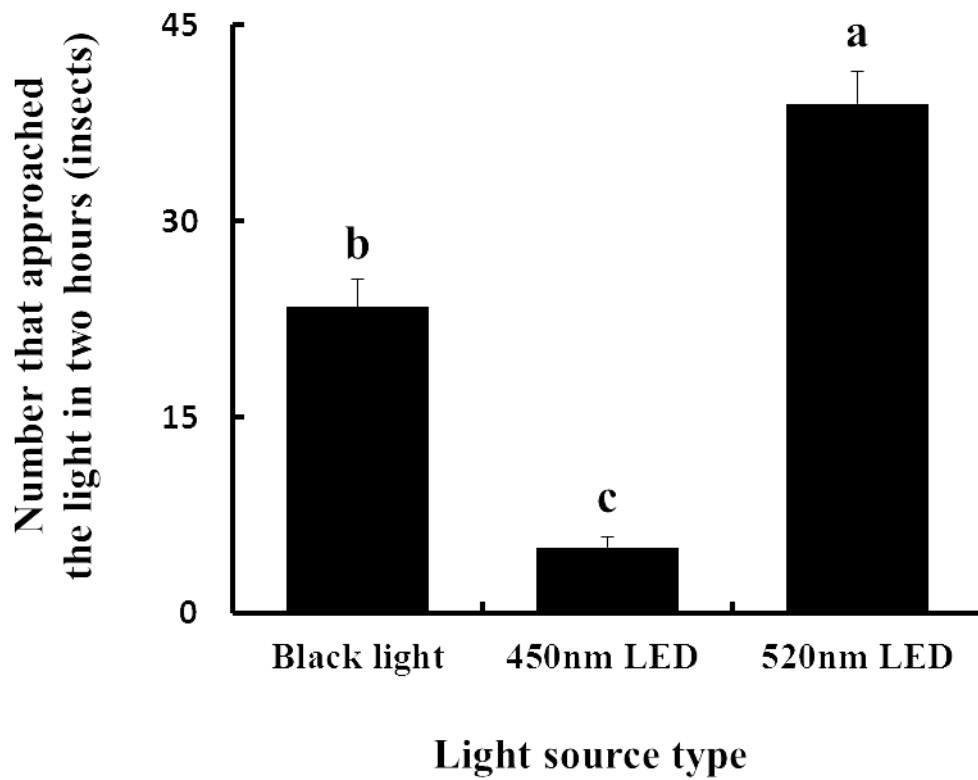
**Figure 3.** Effect of luminance intensity on the phototactic response of *M. separata* moths exposed to green (520 nm) LED light. Here, the exposure time is 40 min. Bars indicate the means and error bars express the SEM. Different letters denote the significant differences according to the Tukey HSD test ( $P < 0.05$ ).

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**Figure 4.** Attraction effects of different light sources on *M. separata* adults in the net cage.

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The value of each bar indicates the total number of adults that appeared in front of

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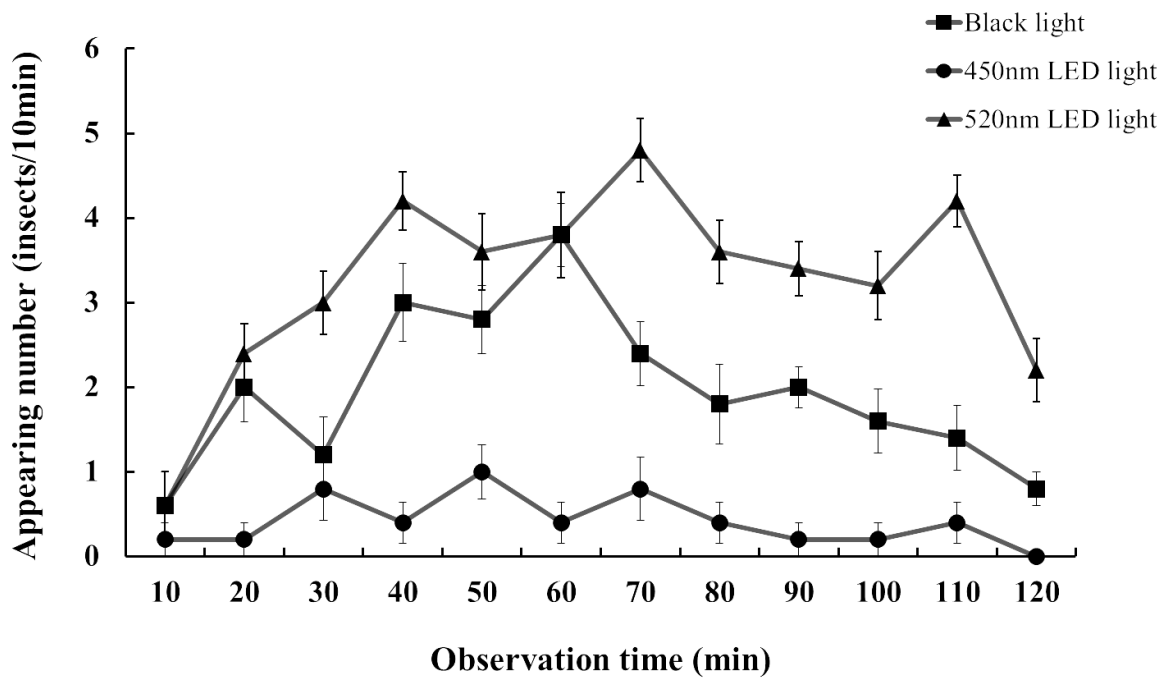
each light over a two hour period, and the error bars indicate the SEM. The different

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letters above each bar express significant differences according to the Tukey HSD test

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( $P < 0.05$ ).



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480 **Figure 5.** Phototactic response tendency of *M. separata* adults to different light sources as a  
481 time series in the net cage. The figure was derived from Figure 4. Each value indicates  
482 the number of adults that appeared for each observation time (10 minutes) in the  
483 videos of each experimental group. The error bar at each peak indicates the SEM.

484

**Tables**

485

**Table 1.** Wavelength and irradiance of LED lights

Light color	Wavelength (Mean $\pm$ S. E) (nm)	Irradiance under 200 lux (Mean $\pm$ S. E) ( $\mu$ W/cm <sup>2</sup> )
Ultraviolet	365 $\pm$ 3	89.3 $\pm$ 3
Ultraviolet	385 $\pm$ 1	107.6 $\pm$ 4
Violet	400 $\pm$ 2	41.7.8 $\pm$ 3
Blue	450 $\pm$ 3	39.4 $\pm$ 2
Green	520 $\pm$ 3	29.8 $\pm$ 2
Yellow	590 $\pm$ 3	57.8 $\pm$ 3
Red	625 $\pm$ 3	118.6 $\pm$ 5
Dark Red	680 $\pm$ 1	676.4 $\pm$ 7
Infrared	740 $\pm$ 4	2047.3 $\pm$ 12
White	420~650	64.2 $\pm$ 3

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**Table 2.** The attraction rate of *M. separata* to LED lights of 10 wavelengths and black light under optimum conditions<sup>1</sup>

Wavelength (nm)	Luminance intensity (lux)	Exposure time (min)	Insect number (mean±SEM)		Ratio <sup>2</sup> (L:D)	Attraction rate <sup>3</sup> (%)	Relative attraction rate <sup>4</sup>
			Light side (Attraction)	Dark side (Repellent)			
365	50	40	13.67±0.89b	4.33±1.20b	3.15	45.56	0.97619
385	50	40	16.33±0.33b	4.67±0.33b	3.50	54.44	1.166667
400	50	40	16.67±0.33b	5.33±0.88b	3.13	55.56	1.190476
450	150	30	17.00±0.58b	4.00±0.58b	4.25	56.67	1.214286
520	200	40	19.33±0.33a	3.67±0.78b	5.27	64.44	1.380952
590	100	40	15.33±0.88b	9.67±0.33a	1.59	51.11	1.095238
625	150	10	15.00±0.58b	9.33±0.33a	1.61	50.00	1.071429
680	50	30	13.67±0.33b	4.00±0.57b	3.42	45.56	0.97619
White	50	40	15.33±1.45b	5.33±1.20b	2.88	51.11	1.095238
IR-740 <sup>5</sup>	-	60	14.67±0.88b	5.67±1.20b	2.59	48.89	1.047619
BL (365) <sup>6</sup>	-	40	14.00±0.58b	7.00±0.58ab	2.00	46.67	1

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489 <sup>1</sup> Each value is the average of three replicates under optimum conditions, using 30 insects per time.490 <sup>2</sup> Each value indicates the ratio of insect numbers in the light side and the dark side.491 <sup>3</sup> Attraction rate (%) is the average percentage of the 30 *M. separata* adults attracted to the light side under optimum conditions.492 <sup>4</sup> Relative attraction rate indicates the ratio of the attraction rate of each group to the attraction rate of the black light (control).493 <sup>5</sup> Each value is the average of three replicates showing the attraction rate to 5 W infrared (740 nm) LED light.494 <sup>6</sup> Each value is the average of three replicates showing the attraction rate to 18 W the black light (control).495 Different letters in column express significant differences according to the Tukey HSD test ( $P<0.05$ ).