forests

Article

Larval Instars and Adult Flight Period of *Monochamus saltuarius* (Coleoptera: Cerambycidae)

Lichun Fan †, Jue Wang †, Weitao Wang and Yanan Zheng *

College of Forestry, Shenyang Agricultural University, Shenyang 110866, China; fanlichun17@163.com (L.F.); wjgz173@163.com (J.W.); wangweitao418@163.com (W.W.)

* Correspondence: rockyya@163.com or yanan@syau.edu.cn; Tel.: +86-186-4040-3865

† These authors contributed equally to this work.

Abstract: *Monochamus saltuarius* is a vector of *Bursaphelenchus xylophilus* in Japan, South Korea, and the middle temperate zone of China. However, there are only a few reports on this species in China, and its biological characteristics are still unclear. In this study, we aimed to elucidate the larval development and adult flight period of *M. saltuarius* to provide a theoretical basis for the effective control of pine wilt disease in the middle temperate zone of China. Seven morphological variables of larvae were measured to determine the number of larval instars, and the adult specimens of *M. saltuarius* were collected from traps in forests to study the flight period of adults in Fushun, Liaoning, the epidemic center of pine wilt disease in the middle temperate zone of China. The results revealed that the full larval period of *M. saltuarius* was 279.6 d, and the larvae had five instar stages, with an average duration of 7.4, 14.3, 49.8, 83.6, and 124.5 d, respectively. Additionally, 78.4% of the overwintering larvae were fourth instar, and 21.6% were fifth instar larvae. We also found that the adults began to emerge from early May to late June, and the period was from early May to mid-August in the forest. During the investigations period from 2018 to 2020, the total number of adults captured was minimal at 744, and precipitation was highest at 291.54 mm in 2019. We characterized the larval instars and adult flight period of *M. saltuarius*, which is a prerequisite for developing appropriate management strategies. The results of this study will provide an important reference for the formulation of strategies to control *M. saltuarius* and pine wilt disease.

Keywords: *Monochamus saltuarius*; larval instar; head capsule width; adult flight period; precipitation

check for updates

Citation: Fan, L.; Wang, J.; Wang, W.; Zheng, Y. Larval Instars and Adult Flight Period of *Monochamus saltuarius* (Coleoptera: Cerambycidae). *Forests* **2022**, *13*, 910. https://doi.org/10.3390/f13060910

Academic Editors: Celia K. Boone and Katherine P. Bleiker

Received: 10 May 2022
Accepted: 8 June 2022
Published: 10 June 2022

1. Introduction

Monochamus saltuarius Gebler (Coleoptera: Cerambycidae) is a species of pinewood borer that primarily damages members of the Pinaceae family, including *Picea abies*, *Pinus sylvestris*, *P. koraiensis*, *P. nigra*, *Larix* spp., *Abies alba*, etc. [1,2]. *M. saltuarius* has not been widely studied before because the mechanical damage that causes it does not lead to economic losses.

In 1987, it was confirmed for the first time in Japan that *M. saltuarius* is a vector insect of *B. xylophilus* [3], and it was subsequently found to carry *B. xylophilus* in Korea [4]. Thereafter, the studies on host trees, biological characteristics, control measures, etc., of *M. saltuarius* started to be reported. In Japan, *M. saltuarius* primarily infests *P. densiflora* and *P. thunbergii*, and its nematode-carrying capability has been studied [3,5]. In South Korea, *B. xylophilus* vectored by *M. saltuarius* infests *P. koraiensis*, and the vector prefers to oviposit on *P. koraiensis* and feed on *Sciadopitys verticillata* [6]. Meanwhile, the control measures of *M. saltuarius*, such as chemical, biological, and physical, have been explored. Adult males produce 2-undecyloxy-1-ethanol aggregation-sex pheromone, which have been used for monitoring and managing *M. saltuarius* [7]. In addition, *M. saltuarius*'s natural enemies [8], electron beam-induced sterility [9], and forecasting model of *M. saltuarius* emergence [10] have been explored.

M. saltuarius was identified as a vector of *B. xylophilus* in the middle temperate zone of China in recent years [11,12]. We have preliminarily studied the population, biological characteristics, and biological control of the vector in China [13–16], but there remain few reports in this regard. Although the life history of *M. saltuarius* has been studied in Fengcheng [17], it is necessary to study *M. saltuarius* life history in Fushun, which is regarded as an epicenter of pine wilt disease in the middle temperate zone of China [18].

The transmission of vector insects is key to the spread of *B. xylophilus*, and predicting the population dynamics of vector insects is of importance to control the spread of pine wilt disease in the areas surrounding the epidemic zones [17]. Therefore, the purpose of this study was to determine the larval instars and adult flight period of *M. saltuarius* under natural conditions through forest investigations. Additionally, basic biological research can inform the development of science-based management measures for *M. saltuarius* in efforts to contain the spread of pine wilt disease.

2. Materials and Methods

2.1. Larvae Collection

M. saltuarius larvae were collected from the Dahuofang Forest in Fushun (41°56′16.3′′ N, 124°13′6.5′′ E, 173.1 m altitude), Liaoning, China, from 16 July 2020 to 8 January 2021. Weakened or diseased *P. koraiensis* individuals with a large number of oviposition scars were cut into half-meter long logs. The larvae were then collected by peeling off the phloem and splitting the xylem of *P. koraiensis*. Approximately 100 *M. saltuarius* larvae were collected every 7 days and then measured in the laboratory. Two hundred eggs were reared in an incubator at 20 °C and 50% humidity in November 2020. One hundred newly hatched larvae (1 day old) were measured as the first instar larvae. The overwintering larvae were collected from 15 November 2019 to 8 January 2021 from a forest to study the instars of *M. saltuarius* larvae during the overwintering period.

2.2. Measuring Larval Instar

Seven morphological variables of *M. saltuarius* larvae were measured as described by Jiang [19] (Table 1).

Table 1. Morphological variables of *M. saltuarius* larvae.

No.	Larval Instar Characteristic	Measurement Method
L1	Head capsule width	Measured the width of the head capsule of the larva
L2	Mandible length	Measured the right mandible length, viewed from the back of the abdomen
L3	Distance between the main ocelli	Measured the distance between the abdominal ocelli from the abdomen
L4	Prothoracic plate width	Measured the width of the posterior margin of the sclerotized part of the anterior thoracolumbar plate, excluding the pterygoid flap
L5	Mesothoracic spiracle length	Measured the right valve of the midrib, as viewed from the back
L6	Body width	Measured the maximum width of the somite
L7	Body length	Measured the length from the mouth to the end of the abdomen

The larval mandible length, distance between main ocelli, head capsule width, prothoracic plate width, mesothoracic spiracle length, body width, and body length were measured (Figure 1). A volume microscope (Optec sz810, (Chongqing, China)) at 50× magnification and a vernier caliper with an accuracy of 0.02 mm were used for measurements. The larvae that could not be measured on the same day were placed in the refrigerator at 4 °C to stop their development and measured the following day.

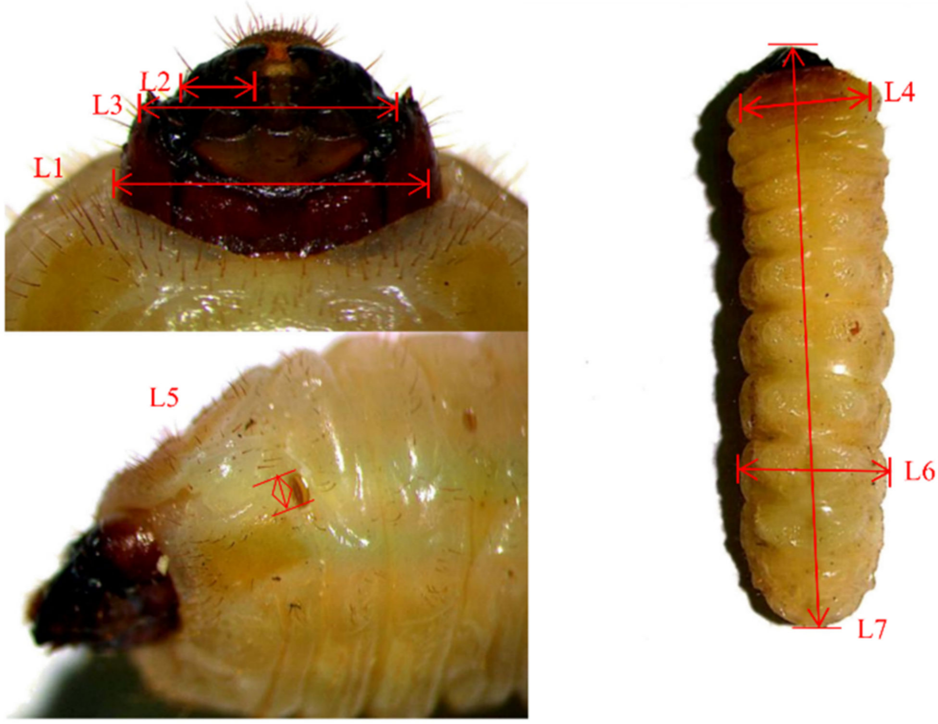

Figure 1. Seven variables measured for distinguishing instars of *M. saltuarius* larvae. (L1) Head capsule width, (L2) mandible length, (L3) distance between the main ocelli, (L4) prothoracic plate width, (L5) mesothoracic spiracle length, (L6) body width, (L7) body length.

The time taken for each larva to reach the different instar stages was recorded, and the developmental duration of each instar was obtained by calculating the average value of all larvae.

2.3. Trapping Adults

Ten cerambycid beetle traps (ZM-80B traps, APF-I attractant) were randomly hung at a height of 1.5–2.0 m at 50 m intervals in forests from 15 April to 15 August in 2018, 2019, and 2020. The trapped *M. saltuarius* adults were collected and counted every seven days, and attractants were replaced once a month. The initial and termination dates of the adult *M. saltuarius* flight period in the forests were also recorded.

2.4. Survey of Emergence Period

On 15 April 2020, *P. koraiensis* with pine wilt disease were selected and cut into one-meter long logs. The logs were sealed with wax at both ends and wrapped with a wire mesh screen (8 mesh iron net), and placed in a forest. Subsequently, *M. saltuarius* adults were collected and counted every day. The emergence period was measured as the time from the emergence of the first adult to the emergence of the last adult.

2.5. Statistical Analysis

All analyses conducting and equations fitting were performed using Statistical Package for Social Sciences software (SPSS20.0 version, SSPS Inc, Chicago, IL, USA), and significant differences were analyzed using Tukey's multiple comparisons and two-way analyses of variance (ANOVA).

The frequency distribution of each variable value was analyzed according to Wang et al. [20], and the frequency distribution map was drawn to determine the instar number of the larva. Then, the mean value, standard error, coefficient of variation (CV), growth rate, and Crosby's ratio for each ratio of each instar were calculated. CV is a normalized measure of the degree of dispersion of a probability distribution. Crosby's ratio is applied as an additional check for overlooked instars. Based on the Crosby growth rule, if the CV is less than 20% and Crosby's ratio is less than 10%, the instar classification is accurate and meets the statistical requirements [21].

$$CV = (SE/Mean) \times 100$$

$$G_n = \overline{X}_m/\overline{X}_{n-1}$$

$$C_n(\%) = (G_n - G_{n-1})/G_{n-1} \times 100$$

where G is the growth rate (Dyar index), also known as the Brooks index, X_n and X_{n-1} are the average values of the two larval instars, C_n is the Crosby's ratio, and G_n and G_{n-1} are the growth rates of two instar larvae [19].

The growth rate and Crosby's ratio were calculated using regression analysis to fit the equations between the measurement variables and instars of *M. saltuarius* larvae according to the measured values to determine the morphological variables selection and standard of instar division.

$$Y_{Ln} = ax^b$$

where Y_{Ln} is the estimated value of the seven variables, *Ln* is the seven variables in Table 1, and x is the larval instar. Parameters a and b represent the scale and shape of the function, respectively.

Climate data (temperature and rainfall) were collected from meteorological stations in China (China Meteorological Administration, 2018–2020. http://data.cma.cn/ (accessed on 24 March 2022)).

3. Results

3.1. Larval Instars

Based on the results of the measurement of 2615 *M. saltuarius* larvae, the frequency distribution of the seven variables was analyzed. The results showed that:

(1) The frequency distribution of head capsule width (L1), mandible length (L2), distance between main ocelli (L3), and prothoracic plate width (L4) exhibited five peaks (Figure 2a–d, Supplementary Data S1). The average head capsule widths of *M. saltuarius* larvae were 0.82, 1.26, 1.96, 3.17, and 4.25 mm for the first to fifth instars, respectively.

(2) The coefficient of variation in the frequency distribution of L5, L6, and L7 variables was relatively large. This could not accurately reflect the age division of *M. saltuarius* (Figure 2e–g, Supplementary Data S1).

According to Dyar's law, the larval instar of *M. saltuarius* has five instars. The overlapping degree of head capsule width was relatively small between the five instars. The variation in body length and width was large, making them unsuitable for the precise division of larval instar.

Statistical analyses of the seven variables of each instar are shown in Table 2. The CV of head capsule width, mandible length, prothoracic plate width, and body length were <20%, which met the statistical requirements. The Crosby's ratio of head capsule width, prothoracic plate width, mesothoracic spiracle length, and body width were <10%, which indicated that it was reasonable to divide the larvae into five instars (Table 2, Supplementary Data S1).

Regression analysis was conducted on the relationship between the measured average value of each index of instar larvae and the number of instars of larvae. The results showed that the relationship between the three variables (L1, L2, and L3) and the number of instars (x) conformed to the exponential relationship described by Dyar's law (Table 3). Thus, the results confirmed that dividing the larval instar of *M. saltuarius* into five instars was appropriate.

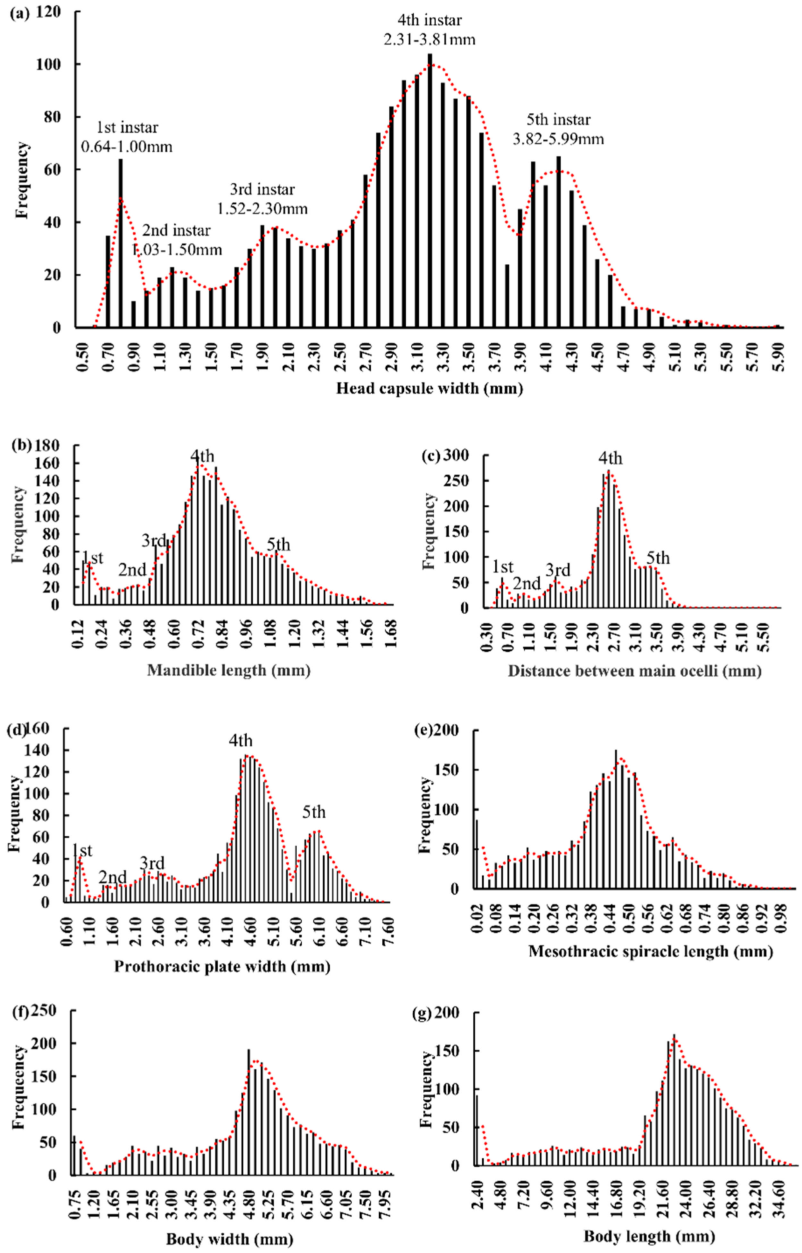

Figure 2. Frequency distribution of the variables separating larval instars of *M. saltuarius*. (**a**) head capsule width, (**b**) mandible length, (**c**) distance between the main ocelli, (**d**) prothoracic plate width, (**e**) mesothoracic spiracle length, (**f**) body width, (**g**) body length.

Table 2. Measurements and analyses of seven variables of different larval instars of *Monochamus saltuarius*.

Variable	Instar	Number of Measured Larvae	Mean ± SE	Minimum (mm)	Maximum (mm)	Coefficient of Variation (CV) (%)	Growth Rate	Crosby's Ratio (%)
Head capsule width (L1)	1	110	0.82 ± 0.06 ^{e}	0.64	1.00	18.93		
	2	89	1.26 ± 0.14 ^{d}	1.03	1.50	15.21	1.38	
	3	296	1.96 ± 0.39 ^{c}	1.52	2.30	10.96	1.62	−6.34
	4	1663	3.17 ± 0.31 ^{b}	2.31	3.81	17.21	1.54	3.21
	5	457	4.25 ± 0.31 ^{a}	3.82	5.99	13.33	1.36	3.71
Mandible length (L2)	1	110	0.18 ± 0.02 ^{e}	0.15	0.24	6.55		
	2	89	0.32 ± 0.05 ^{d}	0.24	0.39	13.92	1.42	
	3	296	0.51 ± 0.10 ^{c}	0.40	0.57	14.24	1.36	−4.92
	4	1663	0.79 ± 0.12 ^{b}	0.57	1.05	14.88	1.55	14.05
	5	457	1.21 ± 0.13 ^{a}	1.05	1.69	10.75	1.53	−1.05

Table 2. *Cont.*

Variable	Instar	Number of Measured Larvae	Mean ± SE	Minimum (mm)	Maximum (mm)	Coefficient of Variation (CV) (%)	Growth Rate	Crosby's Ratio (%)
Distance between the main ocelli (L3)	1	110	0.61 ± 0.05^{e}	0.48	0.73	23.23		
	2	89	1.00 ± 0.11^{d}	0.74	1.22	27.21	2.43	
	3	296	1.57 ± 0.31^{c}	1.23	1.86	27.42	1.73	−29.00
	4	1663	2.61 ± 0.27^{b}	1.86	3.11	21.41	1.97	13.82
	5	457	3.41 ± 0.23^{a}	3.11	5.81	16.07	1.65	−16.04
Prothoracic plate width (L4)	1	110	0.90 ± 0.13^{e}	0.61	1.34	16.07		
	2	89	1.67 ± 0.18^{d}	1.36	1.97	6.28	1.53	
	3	296	2.49 ± 0.47^{c}	1.97	2.97	13.88	1.48	7.32
	4	1663	4.66 ± 0.60^{b}	2.97	5.85	12.90	1.37	−7.20
	5	457	6.28 ± 0.33^{a}	5.85	7.65	9.78	1.50	9.45
Mesothoracic spiracle length (L5)	1	110	0.03 ± 0.01^{d}	0.02	0.07	11.95		
	2	89	0.10 ± 0.02^{c}	0.07	0.12	10.99	1.49	
	3	296	0.18 ± 0.05^{b}	0.12	0.24	25.23	1.38	−0.68
	4	1663	0.44 ± 0.08^{a}	0.24	0.58	11.95	1.52	−7.09
	5	457	0.68 ± 0.08^{e}	0.58	1.02	7.30	1.37	−9.62
Body width (L6)	1	110	0.92 ± 0.15^{a}	0.77	1.49	16.01		
	2	89	1.85 ± 0.17^{c}	1.52	2.12	13.66	1.49	
	3	296	2.85 ± 0.44^{d}	2.12	3.06	25.32	1.46	8.37
	4	1663	4.91 ± 0.70^{b}	3.06	6.07	22.99	1.34	−1.76
	5	457	6.71 ± 0.46^{e}	6.07	8.23	7.09	1.85	−8.47
Body length (L7)	1	110	3.00 ± 0.71^{e}	2.42	5.64	6.54		
	2	89	7.68 ± 1.03^{d}	5.73	9.26	10.95	1.30	
	3	296	12.67 ± 3.00^{c}	9.28	16.02	12.82	1.46	12.82
	4	1663	23.60 ± 2.62^{b}	16.04	28.02	15.63	1.45	−15.42
	5	457	30.34 ± 1.74^{a}	28.04	36.50	14.85	1.60	1.86

The same column data (mean ± SE) with different letters indicated a significant difference between instars ($p < 0.05$, Tukey's test).

Table 3. Regression relationship between the number of larval instars and variables of *Monochamus saltuarius*.

Variable	Regression Equation	Correlation Coefficient	Statistical Parameters
Head capsule width (L1)	$Y_{L1} = 0.22x^{0.37}$	0.9994	df = 1,4, F = 3561.03, $p < 0.0001$
Mandible length (L2)	$Y_{L2} = 0.04x^{0.42}$	0.9950	df = 1,4, F = 398.34, $p < 0.0001$
Distance between main ocelli (L3)	$Y_{L3} = 0.16x^{0.39}$	0.9980	df = 1,4, F = 1010.42, $p < 0.0001$
Prothoracic plate width (L4)	$Y_{L4} = 0.19x^{0.45}$	0.9990	df = 1,4, F = 2000.11, $p < 0.0001$
Mesothoracic spiracle length (L5)	$Y_{L5} = 0.0032x^{0.69}$	0.9950	df = 1,4, F = 595.68, $p < 0.0001$
Body width (L6)	$Y_{L6} = 0.19x^{0.46}$	0.9960	df = 1,4, F = 953.31, $p < 0.0001$
Body length (L7)	$Y_{L7} = 0.66x^{0.50}$	0.9950	df = 1,4, F = 843.26, $p < 0.0001$

x: larval instar number (1–5 instars).

The development duration of the full larval instar was 279.6 d, and the duration of instars 1–5 was 7.4, 14.3, 49.8, 83.6, and 124.5 d, respectively. In addition, the overwintering larvae were the fourth (78.4%) and fifth (mature) instar larvae (21.6%).

3.2. Adult Flight Period

M. saltuarius adults began to occur in Fushun in early May. The activity of adults decreased in late July, and the proportion of dead adults in traps increased. No adult individuals were found in traps after mid-August. In 2018, the maximum number of adults captured per week was 373 on 10 July. In 2019, the maximum number of adults captured per week was 110 on 3 May. In 2020, the maximum number of adults captured per week was 292 on 13 June, followed by 272 on 17 July. There were two peaks of captured adults appearing in 2018 and 2020, respectively, one in June and another in July (Figure 3).

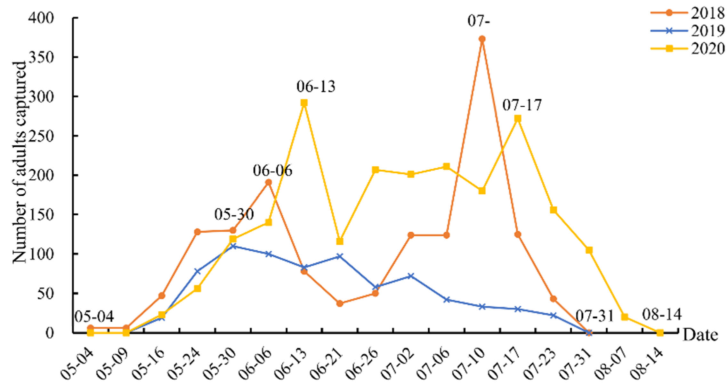

Figure 3. The number of *M. saltuarius* adults captured per week in three consecutive years. The numbers on the curves indicate the sampling date (month-day).

The total number of adults captured was 1462, 744, and 2078, from 4 May to 31 July in 2018, 2019, and 2020, respectively. Meteorological data showed that during the period of investigation, the total precipitation for this period was 269.04, 291.54, and 276.09 mm, for 2018, 2019, and 2020, respectively. The total temperature was 1959.62 °C, 1922.33 °C, and 1899.07 °C for 2018, 2019, and 2020, respectively. The number of adults captured per week was minimal, and weekly precipitation was highest in 2019 (Figure 4, Supplementary Data S1).

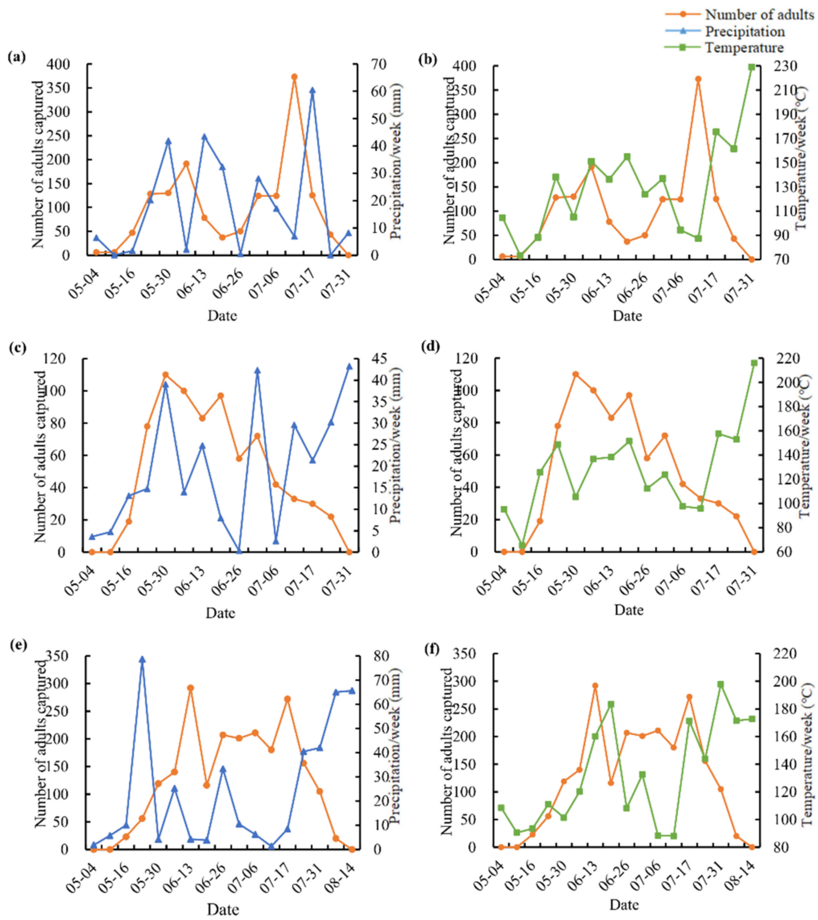

Figure 4. Relationship among the number of *M. saltuarius* adults captured per week, precipitation per week, and temperature per week. (**a**,**b**): in 2018, (**c**,**d**): in 2019, (**e**,**f**): in 2020.

In 2020, *M. saltuarius* started to emerge on 4 May, after which the number of adults emerging per week gradually increased and peaked at 30 on 24 May, then gradually decreased to zero by 21 June. The emergence period lasted for 49 days. The adults started to be captured by the traps on 16 May, and no adult individuals were trapped after 16 August. The trapping period lasted for 90 days. The mean longevity of adults in the forest was presumed to be 50–60 days (Figure 5, Supplementary Data S1).

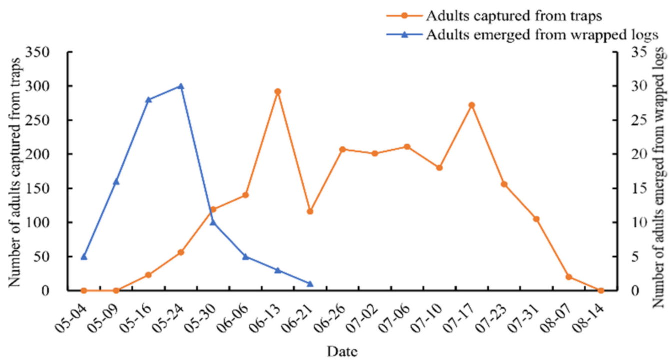

Figure 5. Relationship between the number of *M. saltuarius* adults captured per week and the number of *M. saltuarius* emerged per week in 2020. The numbers on the curves indicate the sampling date (month-day).

The ratio of males to females was 1:1.22, 1.36:1, and 1:1.02 for 2018, 2019, and 2020, respectively. There was no significant difference between the number of female adults and male adults in 2018, 2019, and 2020 (F = 0.004, df = 1,4, $p = 0.956$, ANOVA) (Figure 6, Supplementary Data S1).

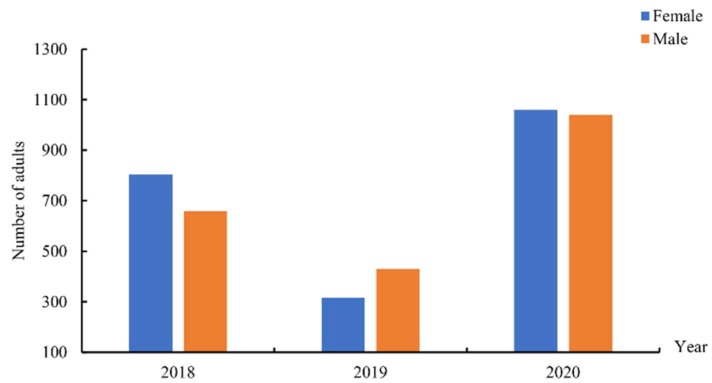

Figure 6. Number of male and female *M. saltuarius* adults collected from traps in different years.

4. Discussion

Dyar [22] found a definite geometric progression relationship between the head capsule width of adjacent larval instars of Lepidoptera insects, which is referred to as Dyar's law [23,24]. It has been widely used in the instar division of Lepidoptera and Coleoptera insects [25,26], and studies have shown that the number of instars is mainly determined by different measurement indicators and geographical location. For example, in the genus *Monochamus*, the development of *M. alternatus* larva was divided into five instars in China [27,28], but it was divided into four instars in Japan [29], and the development of *M. galloprovincialis* larva was divided into four instars [30]. Based on the results of previous studies, seven morphological variables were selected in this study, CV of head capsule width and prothoracic plate width were <20%, and the Crosby's ratio was <10%, which indicated that it was reasonable to divide the larvae into five instars. This result is consistent with most studies on *M. alternatus* [31–33]. However, Li et al. [17] divided *M. saltuarius* larvae into four instars by measuring head capsule width, which may have been caused by different measurement indicators used in their research. Our results showed that larval instar cannot be accurately determined by the variation in body length and body width. Thus, it is not recommended as a variable for the basis of instar division. However, in practice, body length and width are the simplest parameters to measure and are beneficial for field conservation workers to develop control strategies depending on different developmental instar of pests. Longhorn beetle larva has a long developmental duration, and the preference of natural enemies at different instars varies. For example, *Dastarcus helophoroides* (Coleoptera: Colydiidae) prefers to prey on mature larvae of longhorn beetle, and its release would need to coincide with the fifth instar stage larvae of *M. alternatus* to control the pest in spring (late April to early May) [34]. However, *Sclerodermus* spp. (Hymenoptera: Bethylidae) prefer to parasitize early-instar larvae of longhorn beetle, and its release would need to coincide with first to second instar stage larvae of *M. alternatus* to control the pest in summer (late June to early July) [35], which can achieve the best control effect.

The number of adults captured was the lowest in 2019 out of the three years. Analysis of the meteorological data shows that the temperature during the investigation period was similar in these three years, and the precipitation in 2019 was higher than in 2018 and 2020. Thus, we speculate that precipitation may influence the number of captured adults. We also found that *M. saltuarius* adults have a peak emergence period in May. This result is consistent with the study by Li et al., in Fengcheng, Liaoning province, China [17], and Han et al., in Korea [36]. We have confirmed that in Fushun, there are two peak flight periods of *M. saltuarius* adults in a year for the first time. Previous studies have found that there are also two peak flight periods of *M. alternatus*, which is the major vector of *B. xylophilus* in South China [37,38]. The results showed that the ratio of male to female adults captured in the forest was 1:1.22, 1.36:1, and 1:1.02 in 2018, 2019, and 2020, respectively. The ratio of female adults was lower than in South Korea, where the ratio of male to female adults was 1:1.358 [36]. This may be caused by different collection methods, i.e., *M. saltuarius* were captured with sentinel traps in forests in this study, but adults emerged from logs were collected in Korea.

The larval instars and adult flight periods of *M. saltuarius* were determined, which will aid in developing science-based management measures for *M. saltuarius*. The larval instars can provide a reference for natural enemy selection and release time in biological control applications [13,15], and the adult flight period can provide a reference for determining the best time to use traps and chemicals in control protocols [16]. In this study, we only collected *M. saltuarius* larvae in 2020 to determine its larvae instars, and the impacts of environmental conditions on *M. saltuarius* were not considered. Therefore, further investigations are necessary for a more accurate understanding of the larval instars.

5. Conclusions

This study further clarifies that the full larval period of *M. saltuarius* was 279.6 d and that the larvae had five instar stages. Additionally, 78.4% of the overwintering larvae were fourth instar, and 21.6% were fifth instar larvae. We also found that the adults began to fly at the end of April or in early May until the end of June, and the peak flight period was from late May to mid-July in the forest. During the study periods from 2018 to 2020, the total number of adults captured was lowest at 744, and precipitation was highest at 291.54 mm in 2019. Based on the conclusions arrived at from basic biological research, scientific measures for the management of *M. saltuarius* can be developed, which can help prevent pine wilt disease from spreading further northward in China.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/f13060910/s1, Supplementary Data S1.

Author Contributions: Conceptualization, Y.Z.; Formal analysis, L.F.; Funding acquisition, Y.Z.; Investigation, L.F. and W.W.; Methodology, Y.Z. and L.F.; Project administration, Y.Z.; Resources, Y.Z.; Software, L.F.; Supervision, Y.Z.; Validation, J.W.; Visualization, L.F.; Writing—original draft, L.F. and J.W.; Writing—review and editing, Y.Z. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the Natural Science Foundation of Liaoning Province, grant number 2019-MS-279.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data presented in this study are available on request from the corresponding author. The data are not publicly available due to their complexity.

Acknowledgments: We thank the Liaoning Province Station of Forest and Grassland Pest Control and Quarantine and the Dahuofang Forest for supporting this study, and undergraduates and postgraduates for their help in our investigation.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Wang, Z.C. *Monocraphia of Original Colored Longicorn Beetles of China's Northeast*; Jilin Science and Technology Publishing House: Jilin, China, 2003; pp. 348–372.
2. Cesari, M.; Marescalchi, O.; Francardi, V.; Mantovani, B. Taxonomy and phylogeny of European Monochamus species, first molecular and karyological data. *J. Zool. Syst. Evol. Res.* **2010**, *43*, 1–7. [CrossRef]
3. Heisuke, S.; Takeshi, S.; Mitsunori, K. Transmission of Bursaphelenchus xylophilus (Steiner et Buhrer) Nickle (Nematoda: Aphelenchoididae) by Monochamus saltuarius (Gebler) (Coleoptera: Cerambycidae). *J. Jpn. For. Soc.* **1987**, *69*, 492–496.
4. Kwon, T.S.; Lim, J.H.; Sim, S.J.; Kwon, Y.D.; Son, S.K.; Lee, K.Y.; Kim, Y.T.; Park, J.W.; Shin, C.H.; Ryu, S.B. Distribution patterns of Monochamus alternatus and M. saltuarius (Coleoptera: Cerambycidae) in Korea. *J. Korean For. Soc.* **2006**, *95*, 543–550.
5. Togashi, K.; Jikumaru, S. Horizontal transmission of Bursaphelenchus mucronatus (Nematoda: Aphelenchoididae) between Insect Vectors of Monochamus saltuarius (Coleoptera: Cerambycidae). *Appl. Entomol. Zool.* **1996**, *60*, 90–92. [CrossRef]
6. Han, J.H.; Kim, H.K.; King, W.J.; Kim, G. Feeding and oviposition preference of the Sakhalin pine sawyer Monochamus saltuarius (Coleoptera: Cerambycidae) for various tree species. *Entomol. Res.* **2016**, *46*, 331–336. [CrossRef]
7. Lee, H.R.; Lee, S.C.; Lee, D.H.; Choi, W.S.; Jung, C.S.; Jeon, J.H.; Kim, J.E.; Park, I.K. Identification of the aggregation-sex pheromone produced by male Monochamus saltuarius, a major insect vector of the pine wood nematode. *J. Chem. Ecol.* **2017**, *43*, 670–678. [CrossRef]
8. Lee, K.S. First record of the family Bothrideridae (Coleoptera) in Korea represented by the wood-boring beetle ectoparasite, Dastarcus helophoroides. *J. Asia-Pac. Entomol.* **2012**, *15*, 273–275.
9. Cho, W.S.; Koo, H.N.; Yun, S.H.; Seon, L.J.; Hun, J.D.; Jin, K.W.; Lee, S.J.; Kyung, K.H.; Han, I.H.; Kwon, Y.D. Electron beam-induced sterility and inhibition of ovarian development in the sakhalin pine longicorn, Monochamus saltuarius (Coleoptera: Cerambycidae). *J. Econ. Entomol.* **2018**, *111*, 725–731. [CrossRef]
10. Jung, C.S.; Koh, S.H.; Nam, Y.; Ahn, J.J.; Lee, C.Y.; Cho, W.I. A model for predicting spring emergence of Monochamus saltuarius (Coleoptera: Cerambycidae) from Korean white pine, Pinus koraiensis. *J. Econ. Entomol.* **2015**, *108*, 1830–1836. [CrossRef]
11. Yu, H.Y.; Wu, H. New host plants and new vector insects for pine wood nematodes discovered in Liaoning. *For. Pest Dis.* **2018**, *37*, 61. (In Chinese)
12. Fan, L.C.; Shi, Y.; Jiang, S.W.; Zheng, Y.N. Study on the species of long-horned beetles carrying Bursaphelenchus xylophilus in Liaoning. *For. Res.* **2021**, *34*, 174–181. (In Chinese)
13. Zheng, Y.N.; Jiang, S.W.; Huang, Y.F.; Yang, Z.Q.; Gao, G.P.; Liu, P.X.; Shi, Y.; Fan, L.C.; Zhang, Y.L. Dastarcus helophoroides, natural enemy of Monochamus saltuarius was found in pine tree infected by Bursaphelenchus xylophilus in Liaoning, China. *Chin. J. Biol. Control* **2020**, *36*, 31. (In Chinese)
14. Zheng, Y.N.; Shi, Y.; Li, Y.; Fan, L.C.; Wang, J.; Wang, W.T. Feeding preference of Monochamus saltuarius (Coleoptera: Cerambycidae) adults for four host pine trees. *Acta Entomol. Sin.* **2021**, *64*, 1478–1482. (In Chinese)
15. Zheng, Y.N.; Zhang, Y.L.; Shi, Y.; Fan, L.C.; Li, Y. Preliminary study on the control efficiency of Monochamus saltuarius by Sclerodermus spp. *Chin. J. Biol. Control* **2022**, *2*, 306–311. (In Chinese)
16. Zheng, Y.N.; Fan, L.C.; Shi, Y.; Chen, G.F.; Wang, J.; Jiang, S.W. A Comparative Study on Trapping Effect of Different Attractants on Monochamus saltuarius. *J. Northeast For. Univ.* **2021**, *49*, 102–104+115. (In Chinese)
17. Li, M.; Dai, Y.; Wang, Y.; Wang, L.C.; Sun, S.H.; Chen, F.M. New insights into the life history of Monochamus saltuarius (Cerambycidae, Coleoptera) can enhance surveillance strategies for pine wilt disease. *J. For. Res.* **2021**, *6*, 2699–2707. [CrossRef]
18. Wang, K.H.; Zhong, Q.L.; Nie, X.M. Analysis of occurrence reasons of pine wilt disease caused by Bursaphelenchus xylophilus in Fushun. *Liaoning For. Sci. Technol.* **2021**, *3*, 56–58. (In Chinese)
19. Jiang, S.N. *Chinese Longhorn Beetle Larva*; Chongqing Publishing House: Chongqing, China, 1989. (In Chinese)
20. Wang, X.Y.; Yang, Z.Q.; Tang, Y.L.; Jiang, J.; Yang, Y.L.; Gao, C. Determination of larval instar number and duration in the oak longhorn beetle Massicus raddei (Coleoptera: Cerambycidae). *Acta Entomol. Sin.* **2012**, *55*, 575–584. (In Chinese)
21. Loerch, C.R.; Cameron, E.A. Determination of larval instars of the Bronze Birch Borer, Agrilus anxius (Coleoptera: Buprestidae). *Ann. Entomol. Soc. Am.* **1983**, *76*, 948–952. [CrossRef]

22. Dyar, H.G. The number of molts of lepidopterous larvae. *Psyche* **1980**, *175*, 420–422. [CrossRef]
23. Dai, Z.Y. Method for determining the age of insect larvae species. *Chin. Bull. Entomol.* **1980**, *4*, 185–187. (In Chinese)
24. Cai, W.Z.; Pang, X.F.; Hua, B.Z.; Liang, G.W.; Song, D.L. *General Entomology*; China Agricultural University Press: Beijing, China, 2011. (In Chinese)
25. Jaime, G.; Brenda, Y.C.; Alfredo, C.; Francisco, J.V.; Fernando, E.V. The coffee berry borer (Coleoptera: Curculionidae), How many instars are there? *Ann. Entomol. Soc. Am.* **2015**, *108*, 311–315.
26. Rachael, A.S.; Whitney, S.Q. Life history of Allokermes galliformis (Hemiptera: Kermesidae) in Colorado. *Ann. Entomol. Soc. Am.* **2018**, *111*, 265–270.
27. Huan, J.S.; Tang, C.S.; Song, H.F.; Chen, Q.S.; Kang, W.T.; He, X.Y. The biological characteristic of Monochamus alternatus in Xiamen City I: Life history. *Entomol. J. East China* **2005**, *14*, 118–121. (In Chinese)
28. Liu, J.D.; Li, B.W.; Wang, J.Y.; Zhao, B.A.; Shu, J.P. Primary study on methods to determine instars of Monochamus alternatus larva. *J. Zhejiang For. Sci. Technol.* **2008**, *4*, 84–86. (In Chinese)
29. Togashi, K. Life table for Monochamus alternatus (Coleoptera: Cerambycidae) within dead trees of Pinus thunbergii. *Jpn. J. Entomol.* **1990**, *58*, 217–230.
30. Koutroumpa, F.A.; Vincent, B.; Rouxmorabito, G.; Martin, C.; Lieutier, F. Fecundity and larval development of Monochamus galloprovincialis (Coleoptera Cerambycidae) in experimental breeding. *Ann. For. Sci.* **2008**, *65*, 707. [CrossRef]
31. Kozima, K.; Katagiri, K. On the larval instar and changes of its composition of Monochamus alternalus HOPE. *J. Jpn. For. Soc.* **2008**, *46*, 307–310.
32. Myeong, S.G.; Soon, H.K.; Su, B.K.; Dong, S.K. The developmental characteristics for the head capsule width of Monochamus alternatus (Coleoptera: Cerambycidae) larvae and determination of the number of instars. *J. Insect Sci.* **2019**, *19*, 26.
33. Hu, X.; Li, M.; Kenneth, F.R.; Luo, Q.Y.; Fu, H.J.; Wu, S.Q.; Liang, G.H.; Wang, R.; Zhang, F.P. Bacterial communities associated with the pine wilt disease vector Monochamus alternatus (Coleoptera: Cerambycidae) during different larval instars. *J. Insect Sci.* **2017**, *17*, 115. [CrossRef]
34. Wen, X.S.; Liao, S.L.; Tang, Y.L.; Yang, Z.Q. Study on the efficacy of releasing Dastarcus helophoroides eggs against Monochamus alternatus in pine forests. *Sci. Silvae Sin.* **2017**, *53*, 133–138. (In Chinese)
35. Lai, Y.X.; Wang, Y.H.; Wang, X.Y.; Yang, Z.Q.; Tang, Y.L.; Qin, R.H.; Zhang, Y.N.; Zhang, Y.F. A field test of controlling the pine wilt disease with the technique of isolating Monochamus alternatus on forest spot and releasing parasitoid Sclerodermus guani. *Chin. J. Biol. Control* **2012**, *28*, 460–465. (In Chinese)
36. Han, J.H.; Yoon, C.M.; Shin, S.C.; Kim, G.H. Seasonal occurrence and morphological measurements of pine sawyer, Monochamus saltuarius adults (Coleoptera: Cerambycidae). *J. Asia-Pac. Entomol.* **2007**, *10*, 63–67. [CrossRef]
37. Huang, W.L. Monitoring population dynamics of Monochamus alternatus and the effect of climate factors using pheromone trapping method. *For. Pest Dis.* **2020**, *39*, 19–23. (In Chinese)
38. Yang, A.M. Study on the change law of population dynamics of Monochamus alternatus Hope adults in forests. *J. Fujian For. Sci. Technol.* **2004**, *31*, 62–64. (In Chinese)