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Functional response and control potential of adult *Arma chinensis* on Colorado potato beetle in Xinjiang, China

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Abstract

Potato is an important food crop. The Colorado potato beetle (CPB) is its key pest. CPBs are now resistant to several chemical pesticides, making their control more difficult. The predatory insect, *Arma chinensis*, is a natural enemy of other plant pests. We studied the predation of adult *A. chinensis* on CPB eggs and young larvae under indoor controlled conditions and its control of CPB in cages under outdoor conditions. Adult *A. chinensis* effectively reduces CPB egg and larva populations, and its predatory functional response aligns with Holling's Type II model. *A. chinensis* adults released within outdoor cages reduced CPB populations. Based on the predation behavior of the adults of *A. chinensis* to CPB eggs and young larvae, *A. chinensis* is an efficient and potential predator.

Keywords Colorado potato beetle, *Arma chinensis*, Natural enemy, Functional response, Predation

Background

Potato is the most important non-cereal food crop worldwide (Tang et al. 2022). Potatoes are starchy and have high nutritional value (Bradshaw and Bonierbale 2010). Compared with wheat, rice, and corn, potatoes are resistant to environmental extremes and show wide adaptability and a broader planting range (Jansky et al. 2019). Colorado potato beetle is the most harmful pest of potato in potato cultivation regions globally (Balaško et al. 2020). The larvae and adults of CPB (Coleoptera, Chrysomelidae) damage Solanaceae crops. The larvae

and adults of CPB feed on potato leaves, reducing the yield significantly (Hare 1980).

Management of CPB populations includes chemical control, agricultural and physical control, and biological control strategies. Most CPB control methods have relied on pesticide use (Grafius and Douches 2008). Pesticides were initially effective for CPB management; however, their excessive use has led to significant levels of pesticide resistance. By 2020, CPB developed resistance to most registered pesticides (Grafius 1997; Stanković et al. 2004; Sladan et al. 2012; Szendrei et al. 2012; Scott et al. 2015; Huseeth and Groves 2013; Balaško et al. 2020). Agricultural and physical control methods can be employed to control CPB, but these are expensive, inefficient, and lack precision. Therefore, low-cost, efficient, and accurate biological control methods need to be developed for CPB control.

Biological control emphasizes safety, efficiency, and environmental protection (Barratt et al. 2018). *Arma chinensis* (Fallou) (Hemiptera: Pentatomidae) (Liu et al. 2021), *Zicrona caerulea* (L.) (Shu et al. 2012), *Phalangium opilio* (L.) (Drummond et al. 1990), and *Adelphocoris*

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lineolatus (Goeze) (Feng et al. 2016) are some of the predatory natural enemies utilized for CPB biocontrol. *A. chinensis* (Hemiptera: Pentatomidae) is particularly beneficial in agricultural ecosystems, as it preys on numerous field pests and has been successfully used commercially. Therefore, we hypothesized that the use of *A. chinensis* can reduce the CPB population.

A. chinensis is a predaceous stink bug primarily distributed across China, Mongolia, Japan, and other East Asian regions (Zou et al. 2012). Within China, it is found in Heilongjiang, Jilin, Xinjiang, and other provinces (Zou et al. 2019). As a native species of China, nymphs and adults of *A. chinensis* prey on the eggs, larvae, pupae, and adults of herbivorous insects belonging to various orders, including Lepidoptera, Coleoptera, Hymenoptera, and Hemiptera (Zhao et al. 2011). While previous reports on the predation of CPB by *A. chinensis* have largely been based on indoor studies, natural environmental conditions are inherently variable and less controlled than those in a laboratory setting. Consequently, the effectiveness of *A. chinensis* in controlling CPB populations in the field is unclear. Recognizing the importance of understanding both laboratory and field dynamics, it is crucial to investigate the predation of *A. chinensis* on CPB under laboratory conditions and conduct controlled release experiments with *A. chinensis* adults in field cages. The outcomes of these experiments are expected to be important in elucidating how *A. chinensis* preys on CPB in the natural environment and assessing the efficacy of this biological control agent. In this study, we evaluated the potential of *A. chinensis* for biological control of CPB by examining its predation function response under laboratory conditions and conducting controlled outdoor release of *A. chinensis* adults. The results of these investigations can provide insights into the feasibility and effectiveness of utilizing *A. chinensis* as a biological control agent against CPB.

Methods

Test insects

The *A. chinensis* population was purchased from Henan Keyun Biopesticide Co., Ltd. *A. chinensis* were fed *Antheraea pernyi* (Geurin-Meneville) pupae in a controlled environment chamber and raised in incubators set at 27 ± 1 °C, with $65 \pm 5\%$ relative humidity (RH) and a photoperiod of 16:8 h (L:D). Experiments were conducted after rearing for two generations.

CPBs were collected in Qapqal County, Ili Prefecture, Xinjiang (E $80^{\circ}31' - 81^{\circ}43'$, N $43^{\circ}17' - 43^{\circ}57'$) and subsequently reared in controlled climate incubators at 27 ± 1 °C, $65 \pm 5\%$ RH, and 16:8 h (L:D). For two generations, they were reared on Shepody potato leaves before commencing the experiments. The primary objective

of the indoor experiments was to determine the developmental period of CPB at each life stage to calculate the survival rate of outdoor CPB at corresponding ages. In the laboratory experiments, the same conditions as rearing [27 ± 1 °C, $65 \pm 5\%$ RH, and 16:8 h (L:D)] were employed; CPB eggs were divided into three replicates, with 40–50 eggs per replicate placed in individual Petri dishes. Strict protocols were followed to handle and monitor the eggs to ensure the accuracy and reproducibility of the experiments. Upon hatching, the newly emerged larvae were provided with fresh potato leaves. Daily observations were made to record the developmental duration of each larval instar and the pupal stage within the Petri dishes.

Predation functional response of the adult of *A. chinensis* indoors on CPB eggs and young larvae

Adult individuals of *A. chinensis* were starved for 24 h. Each adult of *A. chinensis* was placed in Petri dishes with eggs or young larvae (1st instar larvae, 2nd instar larvae) of CPB at different densities. Prey densities were 15, 20, 25, 30, 35, and 40 CPBs per Petri dish, respectively. After 24 h, the predation of *A. chinensis* on CPBs was recorded in each Petri dish. The number of preyed-upon CPBs was determined by counting the dead bodies that had been sucked dry. For each prey density treatment, there were six replicates.

Outdoor control effect of *A. chinensis* on CPB

Nine cages (1 m × 1 m × 1 m), each containing nine pots of potatoes were used. When the potato plants reached approximately 20 cm in height, each cage was inoculated with 8 adult CPBs (4♀ + 4♂). The number of surviving adult CPB and the number of eggs produced were monitored every 2 days. If CPB eggs were observed for 14 consecutive days (7 occasions), adult *A. chinensis* was released. Adult *A. chinensis* were starved for 24 h before being released. The benefit/harm ratios were set as follows: Control (Not releasing *A. chinensis*), 1:40 (Release 1 *A. chinensis* per 40 CPB), 1:20 (Release 1 *A. chinensis* 20 per CPB) (the number of CPBs includes the total number of CPB eggs and larvae). Each treatment was repeated thrice. The numbers of surviving *A. chinensis* and CPBs were observed and recorded every 48 h. The experiment lasted 60 days. A thorough examination was conducted to inspect all leaves of the nine plants in each cage and record the number of CPB individuals in each cage. The experiment ran from July 22, 2021, to October 4, 2021. Potatoes were planted in the shade, where the ambient temperature was maintained at 15–29 °C.

Data analyses

Predatory function

The functional response was analyzed in two phases in the SAS statistical environment (version 8). The first phase involved determining the type and estimating the parameters of the functional response curve. Finding the type of functional response for calculating the functional response parameters using a proper model was compulsory. The type was determined by applying logistic regression of the proportion of prey eaten as a function of initial prey density offered. A polynomial logistic regression equation assuming a binomial distribution of data to define the type of functional response was fitted as follows:

$$N_a = \frac{\exp(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3)}{1 + \exp(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3)} \quad (1)$$

where N_a and N_0 indicate the number of prey consumed and the initial prey density offered, respectively, and N_a/N_0 is the proportion of prey consumed. P_0 , P_1 , P_2 , and P_3 are the regression parameters representing intercept or constant, linear, quadratic, and cubic coefficients, respectively. If $P_1 > 0$ and $P_2 < 0$, the proportion of prey consumed was positively density-dependent, representing a type III functional response. If $P_1 < 0$, the proportion of prey consumed declined monotonically with the initial prey density, a type II functional response was considered (Juliano 2001).

The Holling II type predation function response model reflects the change of predation of a single natural enemy within a fixed time with changes in prey density (Huang et al. 2021, 2019; Park et al. 2021; Roubinet et al. 2017). Following this analysis, we used Holling's equation to calculate the functional response if our data fit a type II functional response. Since the experiment was conducted without prey replacement during the course of the experiment, the appropriate model to estimate the handling times (T_h) and the attack rates (a) for a type II functional response was Holling's random predator equation (Holling 1959):

$$N_a = \frac{aTN}{1 + aT_h N} \quad (2)$$

where T represents the time that predator and prey are exposed to each other ($T = 1$ day), and a is the predation coefficient or the predator attack rate. The value a/T_h indicates the effectiveness of the predator, calculated by dividing a by T_h , and the maximum theoretical predation rate, $K = T/T_h$, was also calculated.

Comparison of the survival rate of CPB for different instars

We used Zhao Zhimo's average duration method to calculate the egg-hatching rate and the larval stage survival rate (Zhao and Zhou 1984):

$$N_{im} = \frac{N_{is}D}{T_i} \quad (3)$$

$$N_{ib} = \frac{T_{i-1}N_{im} + T_iN_{(i-1)m}}{T_i + T_{i-1}} \quad (4)$$

$$S_i = \frac{N_{(i-1)b}}{N_{ib}} \quad (5)$$

where N_{is} represents the cumulative number of individuals in the larval stage; D represents the survey time interval; T represents the developmental duration; N_{im} represents the number of individuals surviving in the middle of each stage; N_{ib} represents the starting number, and S_i represents the survival rate.

We used Excel to record and count the survey data and calculated the predation of CPB eggs and young larvae, number of tubers per plant, yield per plant, and the survival rate of CPB for each instar stage. Statistical analysis was performed using a one-way analysis of variance (ANOVA), conducted using the IBM SPSS Statistics 21.0 software. The LSD method was used to differentiate experimental results when the homogeneity of variance was satisfied; else, the non-parametric Kruskal–Wallis test method was used to evaluate differences among groups.

Results

Functional response

Logistic regression yielded a negative linear parameter ($P_1 < 0$) for adult *A. chinensis*, suggesting that the predator displayed a type II functional response for CPB eggs and young larvae (Table 1).

Table 1 Results of logistic regression analysis for the proportion of nymphs of adult *A. chinensis* predate on CPB eggs and young larvae relative to the initial number of nymphs provided

CPB stage	Parameter	Estimate	Standard error	Pr
Egg	P_0	12.9546	3.2117	<0.0001
	P_1	−1.4689	0.3779	0.0001
	P_2	0.0512	0.0140	0.0003
	P_3	−0.0006	0.0002	0.0006
Young larva	P_0	9.3415	3.1505	0.0030
	P_1	−1.0336	0.3691	0.0051
	P_2	0.0364	0.0137	0.0079
	P_3	−0.00041	0.000162	0.0107

The mean number of CPB eggs consumed by the adult of *A. chinensis* at prey densities of 15, 20, 25, 30, 35, and 40 was 9.00, 8.83, 7.67, 11.33, 16.00, and 16.67, respectively (Fig. 1a).

The mean number of CPB young larvae consumed by the adult of *A. chinensis* at the prey densities of 15, 20, 25, 30, 35, and 40 was 9.50, 10.67, 9.33, 15.67, 17.17, and 17.67, respectively (Fig. 1b).

The mean number of prey a predator consumes increases with the initial density offered. The highest number of CPB eggs and young larvae was consumed at a prey density of 40.

Data in Table 2 reveal that the coefficient of attack rate (a) for adult *A. chinensis* to the eggs and young larvae of CPB are 1.07 and 0.87, respectively. The handling time for adult *A. chinensis* to the eggs and young larvae of CPB was 0.06 and 0.04, respectively. The maximum theoretical predation rate for adult *A. chinensis* on eggs and young larvae of CPB was 17.60 and 28.96, respectively.

Outdoor control effect of *A. chinensis* on CPB

Different benefit-to-harm ratios did not significantly influence the number of tubers per plant ($p=0.293$, $p>0.05$). Different benefit-to-harm ratios significantly influenced the potato yield per plant ($p=0.000$, $p>0.05$). When the benefit-to-harm ratio was 1:20, the yield per potato plant was the highest, while at the Control ratio, the potato yield per plant was the lowest (Fig. 2).

The survival rate of young and old larvae of CPB was not significantly influenced by different benefit-to-harm ratios ($p>0.05$). Different benefit-to-harm ratios significantly influenced the total immature survival rate of CPB ($p=0.019$, $p<0.05$). When the benefit-to-harm ratio was 1:20, the total immature survival rate was the lowest (Table 3).

The effects of *A. chinensis* on the population density of CPB are illustrated in Fig. 3. After 46 days, CPB adults

Table 2 Parameters of functional response of adult *A. chinensis* to CPB eggs and young larvae

CPB stage	Parameters			
	a±SE	T _h ±SE	a/T _h ±SE	K±SE
Egg	1.07±0.20	0.06±0.01	18.89±3.62	17.60±1.40
Young larva	0.87±0.10	0.04±0.01	25.33±3.00	28.96±2.83
p-value	0.345	0.002	0.200	0.005

a = Coefficient of attack rate, T_h = Handling time, K = maximum theoretical predation rate

began to emerge. The number of adult CPB emerging from the soil at benefit-to-harm ratios of 1:40 and 1:20 was significantly lower than that of the control treatment ($p=0.023$, $p<0.05$) (Fig. 3).

Discussion

The adults of *A. chinensis* exhibited the highest predation on eggs of CPB. Consequently, the release of *A. chinensis* adults during the CPB's reproductive season is recommended for effective control. Analysis of the benefit/harm ratios indicated a decrease in the CPB population with increased numbers of released *A. chinensis* adults, signifying a substantial predation impact. These findings can facilitate the determination of optimal release quantities of *A. chinensis* for optimal CPB management.

The functional response of a predator is pivotal in the population dynamics of prey (Schenk and Bacher 2010). The results of this study show that the predatory functional responses of the adult of *A. chinensis* to CPB eggs and young larvae are consistent with the Holling II disk equation. Previous studies indicate that the predatory functional responses of *Coleomegilla maculate* to CPB eggs conform to the Holling II disk equation (Munyaneza and Obrycki 1997). The results of this study are in line with previous findings.

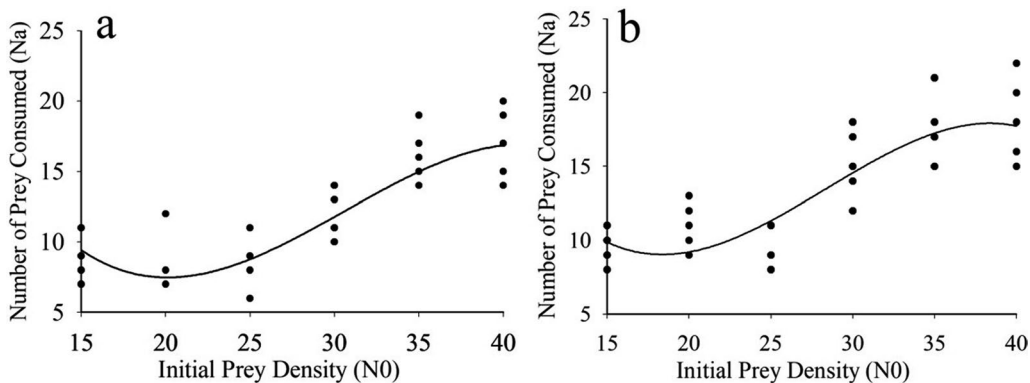


Fig. 1 Functional response of adult of *A. chinensis* at different prey densities of CPB eggs (a) and young larvae (b) during the 24 h period

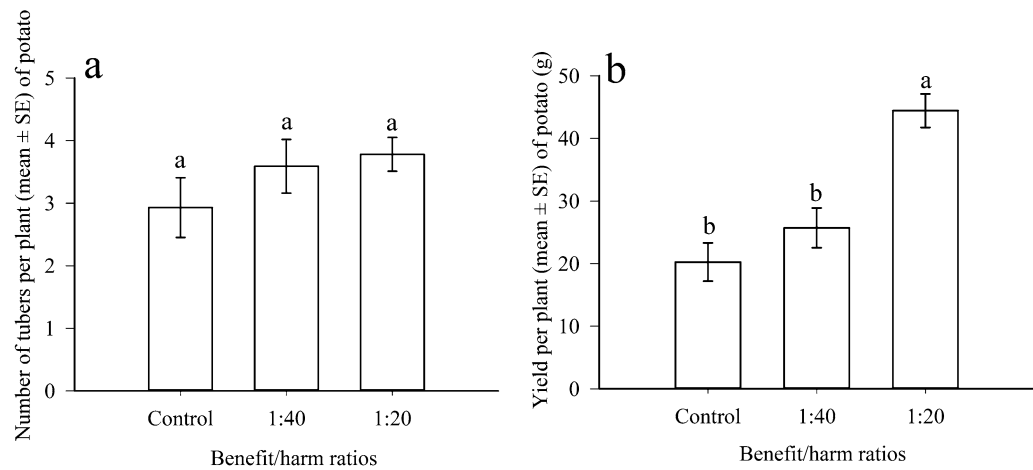


Fig. 2 Number of potato tubers per plant (a) and yield per plant (b) under different benefit/harm ratios (number of *A. chinensis* adults/number of CPBs)

Table 3 Survival rate of CPB under different benefit/harm ratios

Benefit/harm ratios	Survival rate of young larvae	Survival rate of old larvae	Immature total survival rate
Control	83.22 ± 4.68a	79.91 ± 8.72a	31.43 ± 6.96a
1:40	78.03 ± 3.83a	64.65 ± 8.28a	13.79 ± 4.92a
1:20	67.53 ± 5.47a	52.09 ± 0.22a	3.64 ± 0.31b

Data in the table are mean ± SE, and different lowercase letters following the data in the same column indicate a significant difference ($p < 0.05$)

The release of *A. chinensis* in the field has shown effective pest control. Field releases of *A. chinensis* inhibit the population growth of *Laphygma exigua* (Hubner) (Gao et al. 2012), *Spodoptera litura* (Fabricius) (Gao et al. 2019; Tang et al. 2020), and *Ambrostoma quadrimopressum* (Zhang et al. 2016). Previous studies have shown that the release of *Chrysoperla rufilabris* (Nordlund et al. 1991),

Perillus bioculatus (F.), or *Podisus maculiventris* (Say) (Hough-Goldstein and McPherson 1996) in the field can suppress the population growth of CPBs. For example, in field cage experiments, CPB populations were reduced by 84%, with release rates of 80,940 *Chrysoperla rufilabris* larvae per hectare (Nordlund et al. 1991). We found that after the release of *A. chinensis* adults in outdoor CPB-infested areas, according to different benefit/harm ratios, potato yields increased, the CPB population decreased, and the survival rate of old larvae declined. The results of this study are consistent with previous findings.

Zhang and Chen concluded that the utilization of natural enemies should ideally be practical, safe, effective, and economical (Zhang and Chen 2014). *A. chinensis* can now be propagated using artificial food, with a short propagation cycle (Forestry Industry Standard of the People’s Republic of China). Consequently, *A. chinensis* is increasingly being applied in fields for pest control and feeds on a diverse range of pests (Gao et al. 2012; Lei

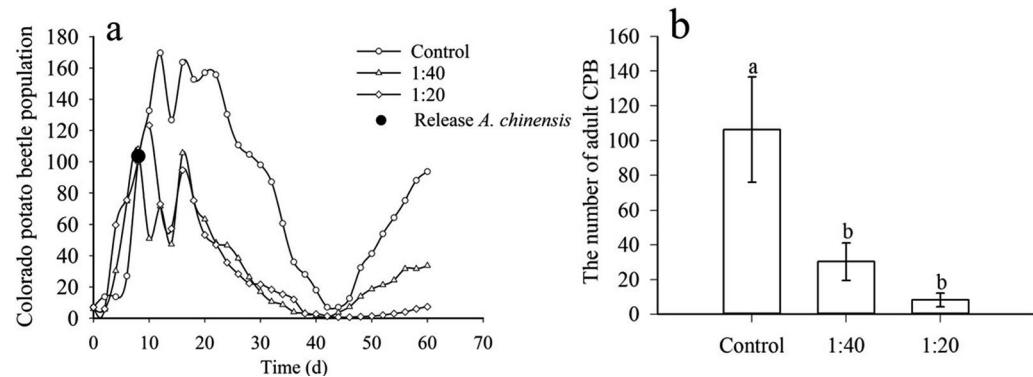


Fig. 3 Changes in population (a) and adult emergence number (b) of CPB under different benefit/harm ratios

et al. 2020; Shu et al. 2020). Although the results of this study showed that the *A. chinensis* preys on CPB under natural conditions, importantly, these experiments were conducted in a controlled cage environment. Therefore, when considering the release of *A. chinensis* into the field, numerous factors must be considered. First, potatoes are subject to infestation by various pests besides CPBs, such as *Phthorimaea operculella* (Zeller) (Rondon 2020) and *Henosepilachna vigintioctopunctata* (Fabricius) (Wang et al. 2017). Given that *A. chinensis* is not an obligate predator of CPBs, further research is necessary to determine if it preferentially targets CPBs in field conditions. Second, this study has only demonstrated that adult *A. chinensis* prey on CPBs under natural conditions. When planning to release *A. chinensis* into the field, factors such as the optimal release time, the instar to be released, and the proportion of *A. chinensis* to release will require further evaluation and experimentation.

Conclusions

CPB is a highly detrimental potato pest and frequently inflicts significant losses on the potato industry. Our findings demonstrated that *A. chinensis* can effectively manage CPB populations. Specifically, adult *A. chinensis* exhibit significant predation effects on the eggs and larvae of CPBs, with their predation functional response conforming to Holling's Type II model. Furthermore, in field cage experiments, the release of adult *A. chinensis* successfully reduced the number of CPBs. Investigating the predation capabilities of *A. chinensis* on CPB in field conditions could pave the way for expanding potato cultivation, enhancing potato yields, and accelerating the adoption of potatoes as a staple food. In the future, we plan to further investigate the predation behavior of *A. chinensis* in field conditions and its influencing factors, in order to optimize their application as biological control agents.

Abbreviations

CPB	Colorado potato beetle
N_a	Number of prey consumed
N_0	Initial prey number available
T	Total time available for search (24 h)
T_h	Prey-handling time
a	Attack rate
a/T_h	Predator's attack rate per handling time
T/T_h	Maximum predation rate
N_{is}	Cumulative number of individuals in the larval stage
D	Survey time interval
T_i	Developmental duration
N_{im}	Number of individuals surviving in the middle of each stage
N_{ib}	Starting number
S_i	Survival rate

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Author contributions

Conceptualization, L.C., L.J.; Formal Analysis, L.J., L.J.H.; Funding Acquisition, L.C.; Investigation, L.J., L.J.H.; Methodology, L.C., L.J., L.J.H.; Project Administration, L.C.; Resources, L.C.; Writing: Original Draft Preparation, L.J.; Writing: Review and Editing, L.C. All authors have read and agreed to the published version of the manuscript.

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Data Availability

Not applicable.

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