



SEASONAL OCCURRENCE OF ADULT DIAPAUSE IN *GALERUCELLA PLACIDA* BALY (CHRYSOMELIDAE: COLEOPTERA)

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ABSTRACT

An experiment was conducted in the laboratory of Department of Entomology, College of Agriculture, Central Agricultural University, Imphal during 2017- 2018 to study the seasonal occurrence of adult diapause in *Galerucella placida* Baly based on feeding and oviposition behaviour. It was observed that the induction of diapause in adult based on feeding behaviour commenced from first fortnight of October, 2017 onwards which was 9.00% and increased gradually and complete diapause reached by the end of December, 2017 (100% from second fortnight of November to second fortnight of December, 2017). Based on Oviposition the initiation of diapause in adult was observed from first fortnight of October onwards, which accounted for 39% adult diapause and complete diapause reached by the end of December (100% from first fortnight of November to second fortnight of December). Maximum ($r = -0.603^{**}$) and minimum temperature ($r = -0.583^{**}$) were observed to be negatively correlated with the incidence of diapause based on feeding whereas minimum temperature ($r = 0.573^{**}$) had a significant positive relationship as seen with oviposition.

Key words: *Galerucella placida*, *Polygonum hydropiper*, adult, oviposition, feeding, diapause, seasonal incidence, temperature, correlation coefficients

The Polygonaceae is a family of flowering plants occurring worldwide, and this name is based on the genus *Polygonum*. This family includes about 50 genera and 1120 species (Anjen et al., 2003). *Polygonum* is a large genus consisting of weedy plants, common in agricultural lands and marshy areas, and *P. hydropiper* L. is a competitive weed in crops. Huda et al. (2017) reported the relative abundance of *P. hydropiper* as 52.5% in weed vegetation of *boro* rice and wheat. Khatun et al., (2014) stated that in wheat the most dominant weed is *Polygonum* sp. In Korea, Park et al. (1995) showed that cyhalofop + bentazone and cyhalofop + pendimethalin gave good control of dryland rice weed mixture which included this species. Seong et al. (1991) observed this with butachlor or benthocarb (thiobencarb) followed by bentazone + quinclorac. Use of herbicides has detrimental effects on non-target wetland plants and potentially hazardous to environment. Biological control is highly cost effective, long term, non-polluting and self-sustaining method of weed control. *Galerucella placida* Baly is a small leaf beetle belonging to the family Chrysomelidae. The insect feeds on the aquatic *P. hydropiper* (Indranisana, 2014). Lefroy (1909) reported *G. placida* as feeding on *P. hydropiper* from India. Both the beetle and grub feed voraciously on *Polygonum* leaf. Description of the

insect was given by Lefroy (1909) and Maulik (1936) and biology of the insect was studied by Indranisana (2014). However, it also on other Polygonaceae such as *P. chinense* (L.) var. *brachiata* Meissn, *P. perfoliatum* L., *P. minus* Huds, *P. labrum* Willd., *P. densiflorum* Blume etc.

The success of a biocontrol programme will depend on achieving large and self-sustaining populations of a biocontrol agent, and for this the knowledge on basic biology and life history is essential (Chippendale, 1982; Tauber et al. 1986). The induction of diapause, developmental stage of its occurrence, the physiological changes occurring during the course of diapause, and the process of its termination are genetically determined and characterize in each species (Kostal, 2006). Members of the family Chrysomelidae commonly enter diapause as adults, although many species overwinter as eggs or in some cases, these might overwinter in either life stage (Cox, 1994). Initial observations indicate that *G. placida* overwinters as adults in the leaf litter beneath the host plant. *G. placida* starts its life cycle from the month of February onward and feeds on the leaves of *P. hydropiper* till October. During this period, the insect completes >15 generations/ year with overlapping generations. However, from the later part of October,

the feeding of leaf and egg laying comes to cessation and the adult insects have been observed to enter leaf litters and cracks and crevices as host plants starts dying during this period. The diapausing adults again resume their activity from February onwards. Therefore, a better understanding of diapause in *G. placida* is needed for successful execution of *P. hydropiper* biocontrol programme, and hence the present study.

MATERIALS AND METHODS

The experiment was carried out in the laboratory of the Department of Entomology, College of Agriculture, Central Agricultural University, Imphal during March 2017-February 2018. Adult beetles of *G. placida* were collected from the field and reared on host leaves in glass petriplates in the laboratory (Figs. 1-5). The egg masses laid were kept for hatching over a moist blotting paper and on hatching, neonate grubs transferred with a fine camel hair brush to leaves of host plant, *P. hydropiper* and reared till pupation. The adult beetle emerged from pupae were released on the leaves for feeding and oviposition (Fig. 6). Fresh leaves were provided daily. The adults emerged on the same day were paired and kept in small petri plates (60x15 mm)

for fortnightly observations (Fig. 7). Observations like feeding, oviposition, and cessation of feeding and oviposition were monitored daily and % worked out. Observations on the cessation of oviposition and non-feeding in adults indicated induction of diapause. The data on weather factors viz., day length, minimum and maximum temperature, morning and evening relative humidity were recorded. Collections and observations of adult were started from March 2017 and continued till February 2018.

RESULTS AND DISCUSSION

The data on the occurrence of diapause in adult *G. placida* based on feeding given in Table 1 reveal that adults feed on the leaves of *P. hydropiper* from March 2017 onwards and until September, with 100% adults observed feeding without any diapause. Cessation of feeding was observed from the first fortnight of October onwards, diapause was 9.00%, reaching 34% by first fortnight and 100% by second fortnight of November, which continued till second fortnight of December. Feeding was initiated again from first fortnight of January 2018 and only 3 pairs of adults resumed feeding out of 10 (30%), and thereafter the feeding increased

Table 1. Diapause in adult *G. placida* (assessed based on feeding, oviposition)

Month	Fortnight	No. of pair observed	No. of pair feeding	Feeding		Oviposition	
				Adult feeding (%)	Adult in diapause (%)	% female laying eggs	Adult in diapause (%)
March	I	50	50	100	0.00	100	0.00
	II	50	50	100	0.00	100	0.00
April	I	50	50	100	0.00	100	0.00
	II	50	50	100	0.00	100	0.00
May	I	50	50	100	0.00	100	0.00
	II	50	50	100	0.00	100	0.00
June	I	50	50	100	0.00	100	0.00
	II	50	50	100	0.00	100	0.00
July	I	50	50	100	0.00	100	0.00
	II	50	50	100	0.00	100	0.00
Aug	I	50	50	100	0.00	100	0.00
	II	50	50	100	0.00	100	0.00
Sept	I	50	50	100	0.00	100	0.00
	II	50	50	100	0.00	100	0.00
Oct	I	46	42	91	9	61	39
	II	32	19	59	41	28	72
Nov	I	29	10	34	66	0.00	100
	II	22	0	0.00	100	0.00	100
Dec	I	18	0	0.00	100	0.00	100
	II	16	0	0.00	100	0.00	100
Jan	I	10	3	30	70	10	90
	II	10	6	60	40	50	50
Feb	I	15	13	87	13	73	27
	II	20	20	100	0.00	100	0.00

to 60% in second fortnight of January 2018 and 87% and 100% in first and second fortnight of February 2018, respectively. The induction of diapause in adult was observed from first fortnight of October onwards (9.00%) and in the subsequent fortnights, this increased and reached 100% by the end of December (100% adult diapause from second fortnight of November to that of December 2017).

Termination of dia pause was observed from first fortnight of January 2018 onwards as indicated by % of adult in diapause which came down to 70%, and feeding also resumed (30% adult feeding) simultaneously. The diapause decreased gradually reached 13% in first fortnight of February 2018, with feeding increased to 87.0%, and diapause completely terminated with 100% feeding by the second fortnight of February. Maximum temperature ($r = -0.603^{**}$), minimum temperature ($r = -0.583^{**}$), relative humidity- evening ($r = -0.304$) and daylength ($r = -0.799^{**}$) were negatively correlated with diapause while it was a positive one with relative humidity morning ($r = 0.359$). The multiple regression analysis revealed that all the abiotic factors contributed 70.9% ($R^2 = 0.709$) in the incidence of adult diapause based on feeding.

As regards oviposition, its cessation was used as criteria for induction of diapause as given in Table 2. The data indicates that all the females oviposit from March and continue till September reaching 100% from first fortnight of March to second fortnight of September 2017. Cessation of oviposition was observed from the first fortnight of October, reaching nil oviposition by first fortnight of November, which continued till second fortnight of December 2017. Oviposition started again from first fortnight of January 2018 with only 1 female resuming of 10 females, and increased to 50%, 73% and 100% in second fortnight of January 2018, first fortnight and second fortnight of February 2018, respectively. Thus, the initiation of diapause was observed from first fortnight of October 2017 (39%) reaching 100% by the end of December 2017. Maximum temperature ($r = -0.563^{**}$), relative Humidity evening ($r = -0.348$) and daylength ($r = -0.821^{**}$) were observed to be negatively correlated with diapause, while a positive correlation was observed with relative humidity morning ($r = 0.381$) and minimum temperature ($r = 0.573^{**}$), with all the weather factors contributing to 75 % ($R^2 = 0.750$) of diapause based on oviposition.

Previous studies of *G. placida* had focused on life history, host specificity and consumption indices

(Indranisana, 2014; Dey, 2015; Debbarma, 2016). *G. calmariensis*, *G. nymphaeae* and *Galerucatanaceti* reveal high incidence of adult diapause (Siew, 1966; Tauber et al., 1996; Velarde et al., 2002). Bean et al. (2007) studied the seasonal timing of diapause induction limits the effective range of *Diorhabdaelongata* (Coleoptera: Chrysomelidae) and reported that diapause induction curve for *D. elongta* had a steep slope going from nearly 100% reproductive with 1h change in day length. The *P. hydropiper* plants are found in the water logged low lying areas from March onwards till September. The plant starts flowering in August and September and withers thereafter in October. The peak period of diapause also coincides with the non-availability of the *P. hydropiper* in the field. The overwintering generations of *G. placida* emerge from the leaf litter and begins feeding as soon as *P. hydropiper* foliage become available from January. Reproductive development begins immediately, requiring no specific photoperiodic stimulation, the same as with many insects' species (Tauber and Tauber, 1970).

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Table 2. Seasonal occurrence of diapause in *G. placida* (based on feeding, oviposition)

(**significant at p=0.01)

Month	Fortnight	Temperature (°C)		Relative Humidity (%)		Daylength (hours)	% diapause	
		Maximum (X1)	Minimum (X2)	Morning (X3)	Evening (X4)		Feeding	Oviposition
March	I	24.0	11.06	82.8	45.7	11.0	0.00	0.00
	II	24.0	12.4	84.8	57.9	12.0	0.00	0.00
April	I	25.9	16.7	86.1	59.8	12.0	0.00	0.00
	II	25.6	18.0	83.9	77.1	12.3	0.00	0.00
May	I	29.6	18.8	80.1	54.2	13.0	0.00	0.00
	II	28.9	20.6	88.8	70.4	13.0	0.00	0.00
June	I	28.9	22.0	89.5	73.6	13.0	0.00	0.00
	II	29.1	22.7	94.2	73.1	13.0	0.00	0.00
July	I	28.3	22.3	95.2	79.5	13.0	0.00	0.00
	II	29.6	22.8	90.5	72.8	13.0	0.00	0.00
Aug	I	28.8	22.8	93.2	79.1	13.0	0.00	0.00
	II	29.4	21.7	94.5	77.6	12.0	0.00	0.00
Sept	I	29.6	21.9	91.8	73.3	12.0	0.00	0.00
	II	28.5	21.6	95.2	78.6	11.8	0.00	0.00
Oct	I	30.1	21.1	91.4	72.1	11.0	9	39
	II	26.2	17.8	93.6	67.5	11.0	41	72
Nov	I	27.1	13.3	94.2	48.0	10.7	66	100
	II	25.9	13.6	92.2	69.5	10.0	100	100
Dec	I	21.7	10.8	94.8	64.4	10.0	100	100
	II	23.0	8.5	92.7	53.6	10.0	100	100
Jan	I	21.1	6.7	92.3	49.5	10.0	70	90
	II	22.4	6.3	86.6	47.0	10.1	40	50
Feb	I	23.9	9.2	87.7	43.7	11.0	13	27
	II	25.6	8.7	88.3	45.9	11.0	0.00	0.00

Based on feeding

Meteorological parameters	Correlation coefficient(r)
Maximum temperature (°C)	-0.603**
Minimum temperature (°C)	-0.583**
Relative humidity morning (%)	0.359
Relative humidity evening (%)	-0.304
Daylength (Hours)	-0.799**

Coefficient of determination (R^2) = 0.709; Multiple Regression Equation: $96.026 - 2.062 (X1) - 0.894 (X2) + 2.150 (X3) + 0.497 (X4) - 9.777 (X5)$

Based on oviposition

Meteorological parameters	Correlation coefficient(r)
Maximum temperature (°C)	-0.563**
Minimum temperature (°C)	0.573**
Relative humidity morning (%)	0.381
Relative humidity evening (%)	-0.348
Daylength (Hours)	-0.821**

Coefficient of determination (R^2) = 0.750; Multiple Regression Equation: $228.283 - 3.665 (X1) + 3.188 (X2) + 2.661 (X3) - 694 (X4) - 30.154 (X5)$

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