

## **Influence of food on the growth, development and hibernation of Large White Butterfly (*Pieris brassicae*)**

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**Abstract.** The abundance of Large White Butterfly (LWB), *Pieris brassicae* fluctuates from year to year, and a peak in the population is reached in every five to seven years, after which there occurs an abrupt decrease in the abundance. The natural checks of the population are primarily weather, parasitoids and pathogens, but the quality of food is also an important factor. The criteria for estimating the influence of food were the duration of caterpillar stage, the mortality rate of caterpillars and prepupae, the weight of pupae and the winter mortality of diapausing pupae. Foodplants: *Brassica oleracea* var. *capitata* f. *alba*, *B. oleracea* var. *capitata* f. *rubra*, *B. oleracea* var. *gemmifera*, *B. oleracea* var. *botrytis*, *B. oleracea* var. *acephala*, *B. napus* var. *napobrassica*, *Tropaeolum majus*, *Armoracia rusticana*.

In our experiments, the most unsuitable foodplants for larvae were *Tropaeolum majus* and *Armoracia rusticana*. There appeared a high mortality rate among caterpillars feeding on both of them as well as among their hibernating pupae. It can be concluded that one of the reasons for the remarkable decrease in the pest population following the massive reproduction of LWB is the high mortality rate of caterpillars growing on less valuable foodplants. The pupae are underweight and, in most cases, they perish during winter.

**Key words:** Large White Butterfly, *Pieris brassicae*, foodplants, larval mortality, pupal body mass, hibernation

### **INTRODUCTION**

One of the greatest pests of cruciferous cultures in Estonia is Large White Butterfly (LWB), *Pieris brassicae*, who, in the years of its biggest abundance, can destroy a significant part of a crop. The larvae feed on the foliage of plants and can completely defoliate and kill a plant (Hill, 1987). In the conditions of our summers, the pest has two full generations, and, in more favourable years, partly a third generation may occur. As an oligophagous pest, the circle of its foodplants is not a large one – the plants mainly belong to five plant families, whereas the most important foodplants belong to the family Cruciferae. The caterpillars of the first generation mainly feed on natural cruciferous plants – *Sinapis arvensis*, *Raphanus sativus*, *Barbarea* spp, *Capsella bursa-pastoris*, *Cardamine* spp, etc. Butterflies of the second generation commonly appear in our fields at the end of July and at the beginning of August, and lay their eggs first of all on the varieties and cultivars of the cabbage *Brassica oleracea*. The massive reproduction and cosmopolitan nature of the pest are closely

connected with the cultivation of cabbage (Nordman, 1954; Fernando, 1971; Feltwell, 1982).

Females of LWB select host plants for their oviposition. This is based on the presence of glucosinolates (Schoonhoven et al, 1998), also known as mustard oil glycosides. The newly hatched larvae cannot exercise host-plant preference, as they lack sufficient powers of movement to leave the plant on which the eggs were laid (Singer et al., 1994). Larvae are stimulated to feed on plants based on the presence of glucosinolates, too (David & Gardner, 1962). In the years when cabbage butterflies are abundant and there is a lack of places for oviposition and feeding, caterpillars may be found feeding on secondary plants as horse-radish (*Armoracia rusticana*, fam. Cruciferae), as well as garden nasturtium (*Tropaeolum majus*, fam. Tropaeolaceae). The nasturtium belongs to different plant family but it contains glucosinolates that typically occur in the Cruciferae. Glucosinolate concentrations and compositions vary within and between species and subspecies of foodplants, different developmental stages of the plant and also among different organs and tissues.

The abundance of LWB fluctuates from year to year, and a peak in the population is reached in every five to seven years (Feltwell, 1982; Luik, 1997). The natural checks of the population are primarily weather, parasitoids and pathogens but the amount of food and its quality are also important factors. The hypothesis of the present study was: may the fact that a part of caterpillars are enforced to develop on less accepted food plants be one of the reasons for the decrease in the number of a population of a following year. To elucidate the influence of different foodplants, a number of experiments were carried out in the laboratory of applied entomology of the Plant Protection Institute of the Estonian Agricultural University. The criteria for estimating the influence of food were the duration of caterpillar stage, the mortality rate of caterpillars and prepupae, the body mass of pupae and the winter mortality rate of diapausing pupae. The results of the study enabled us to make some conclusions on the reasons for low level times following the massive reproduction periods of LWB.

## MATERIALS AND METHODS

Egg clutches of LWB were gathered in the vicinity of Tartu, and placed in Petri dishes with a wet filter paper under the lid of each dish to avoid drying. The caterpillars were raised in 1-litre glass bottles, ten individuals in each, L : D of 12 : 12 h,  $23 \pm 1^\circ\text{C}$ . The larvae of younger instars are fragile, and it is risky to remove them from leaves. Therefore we waited until the caterpillars themselves moved onto the fresh food, after which the bottle was cleaned of excrements and food residue. The experiments were carried out in three replications, each including 30 individuals.

Foodplants: white cabbage (*Brassica oleracea* var. *capitata* f. *alba*, cultivar ('Podarok'), red cabbage (*Brassica oleracea* var. *capitata* f. *rubra*), Brussel sprouts (*Brassica oleracea* var. *gemmifera*), cauliflower (*Brassica oleracea* var. *botrytis*), kale (*Brassica oleracea* var. *acephala*), rutabaga (*Brassica napus* var. *napobrassica*), nasturtium (*Tropaeolum majus*), horse-radish (*Armoracia rusticana*). Cultivar 'Podarok' was chosen as a standard because during the experiments it was a major cultivar for commercial production in Estonia.

In the course of the experiment, the length of the caterpillar stage was determined in days, the mortality rate of caterpillars and prepupae in per cents, and the mass of the

pupae in mg. The pupae were weighed on analytical scales ‘Meopta’, where a pupa was placed in a closed weighing chamber during the weighing. The precision of the weighing was 0.1 mg. The hibernating pupae were kept in a thermostat at +19°C with relative air humidity 60–65%. To retain the air humidity, there was a dish with water. Dead pupae were removed from the experiment during weekly examinations of the material. The significance of differences between the means was calculated by Student *t*-test ( $P = 0.05$ ).

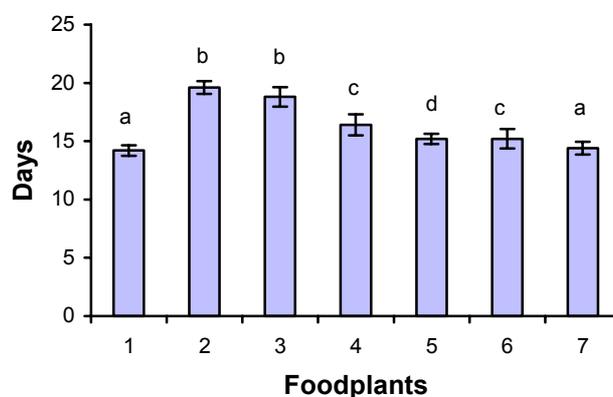
## RESULTS

### *The duration of the larval stage*

The caterpillar stage of LWB in the control (cultivar ‘Podarok’) was the shortest, averagely 14 days. With other foodplants it lasted from 14.4 (red cabbage, var. *capitata* f. *rubra*) to 18.8 days (horse-radish, *A. rusticana*) and 19.6 days for nasturtium, *T. majus*. Butterflies usually do not lay eggs on red cabbage. However, in our experiments caterpillars feeding on red cabbage developed equally with those feeding on cultivar ‘Podarok’, and they completed their larval stage as quickly as the control variant. With the equal speed (15.2 days), caterpillars developed both on cauliflower (var. *botrytis*) and Brussels sprouts (var. *gemmifera*), however, the development lasted one day longer than that of the caterpillars of the control and red cabbage variants. The development of caterpillars fed on rutabaga (var. *napobrassica*) lasted two days longer than the control (Fig. 1).

### *Death Rate of Caterpillars in the Test Period and the Number of Emerged Pupae*

Fig. 2 shows the percentage of emerged pupae. It appeared that the control variant (‘Podarok’) gave the biggest number of pupae. The smallest number of pupae was



1 – *Brassica oleracea* var. *capitata* f. *alba*, cultivar ‘Podarok’; 2 – *Tropaeolum majus*; 3 – *Armoracia rusticana*; 4 – *Brassica napus* var. *napobrassica*; 5 – *Brassica oleracea* var. *botrytis*; 6 – *Brassica oleracea* var. *gemmifera*; 7 – *Brassica oleracea* var. *capitata* f. *rubra*

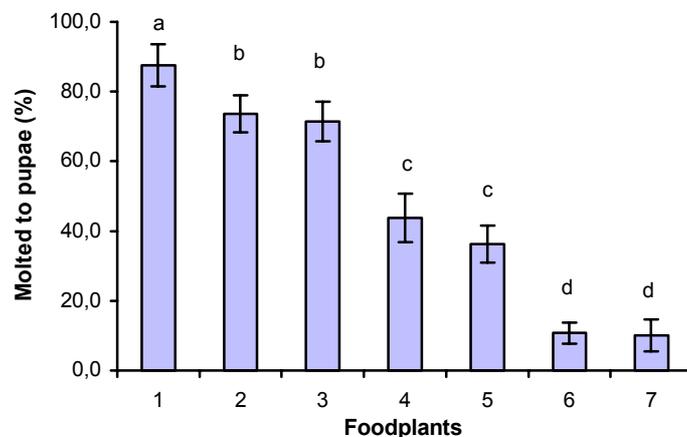
**Fig. 1.** Duration of caterpillar stage of Large White Butterfly, *Pieris brassicae* measured in days of their feeding on different foodplants. Different letters above vertical bars (standard deviation) represent statistically different values ( $P < 0.05$ ).

among caterpillars fed on horse-radish (*A. rusticana*), where only 10% of them were alive. Nearly the same result was gained with nasturtium (*T. majus*) – only 11% pupated.

The material obtained can be divided into four parts. First, the control variant (cultivar ‘Podarok’) differs from all others parts, having the lowest mortality rate and giving about 90% of pupae. The second part, where the comparison showed no significant differences, consisted of the variants of red cabbage (var. *capitata* f. *rubra*) and rutabaga (var. *napobrassica*) – the percentage of survivors 70%. The third part included the variants of cauliflower (var. *botrytis*) and Brussels sprouts (var. *gemmifera*), where the percentage of survivors was  $\approx$  40%, and there was no reliable statistical difference in the comparison. The fourth part was the variants with nasturtium (*T. majus*) and horse-radish (*A. rusticana*), where the rate of survivors was only 10–11%, while between these variants no significant difference existed either.

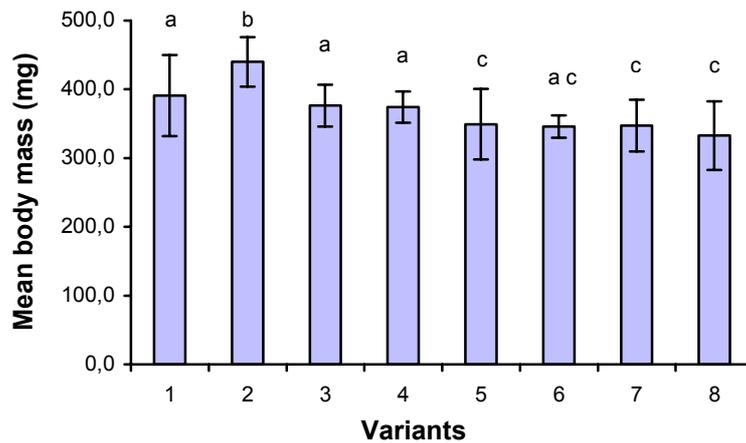
#### Body Mass of Pupae in Different Variants

In comparing body mass in each variant, the mean body mass of 1-week-old pupae was taken as a basis. The lightest pupae were those in the variant with horse-radish (*A. rusticana*): 332 mg on the average, the heaviest in the variant with kale (var. *acephala*) the mean body mass was 440 mg (Fig. 3). The latter mass exceeded also the mean mass of the pupae in the control variant (‘Podarok’) which was 390 mg. The mean body mass of the pupae was both in the variant with kale (var. *acephala*) and in the control significantly bigger than in the variants with rutabaga (var. *napobrassica*) (349 mg), Brussels sprouts (var. *gemmifera*) (347 mg), nasturtium (*Tropaeolum majus*) (345 mg) and horse-radish (*Armoracia rusticana*) (332 mg). The mean weight of pupae in the variants with red cabbage (var. *capitata* f. *rubra*) and cauliflower (var. *botrytis*) was approximately the same: 376 mg and 374 mg, respectively (Fig. 3).



1 – *Brassica oleracea* var. *capitata* f. *alba*, cultivar ‘Podarok’; 2 – *B. oleracea* var. *capitata* f. *rubra*; 3 – *B. napus* var. *napobrassica*; 4 – *B. oleracea* var. *botrytis*; 5 – *B. oleracea* var. *gemmifera*; 6 – *Tropaeolum majus*; 7 – *Armoracia rusticana*

**Fig. 2.** Emerged pupae of *Pieris brassicae* developed from caterpillars raised on different food plants. Different letters above vertical bars (standard deviation) indicate significant differences among columns.

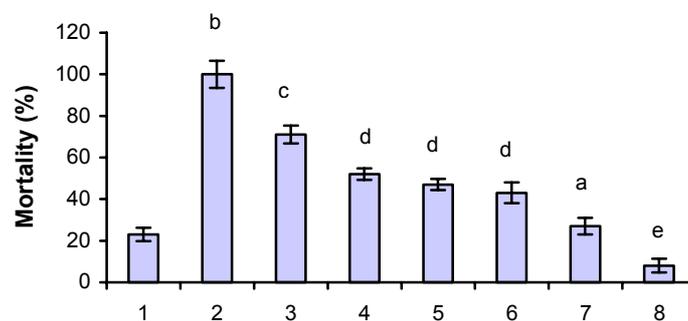


1 – *Brassica oleracea* var. *capitata* f. *alba*, cultivar ‘Podarok’; 2 – *Brassica oleracea* var. *acephala*; 3 – *Brassica oleracea* var. *capitata* f. *rubra*; 4 – *Brassica oleracea* var. *botrytis*; 5 – *Brassica napus* var. *napobrassica*; 6 – *Tropaeolum majus*; 7 – *Brassica oleracea* var. *gemmifera*; 8 – *Armoracia rusticana*

**Fig. 3.** Dependence of mean body mass of *Pieris brassicae* on the food plants shortly after pupation. Different letters above vertical bars (standard deviations) indicate significant differences among columns.

#### Winter Mortality Rate of Pupae of Large White Butterfly

The experiments showed that the pupae with the smallest mean body mass of the variants with horse-radish (*Armoracia rusticana*) and nasturtium (*Tropaeolum majus*) had a high winter mortality rate (100% and 71%, respectively), whereas in the variant with horse-radish all the pupae died already during the first twelve hibernation weeks.



1 – *Brassica oleracea* var. *capitata* f. *alba*; 2 – *Armoracia rusticana*; 3 – *Tropaeolum majus*; 4 – *Brassica oleracea* var. *gemmifera*; 5 – *Brassica napus* var. *napobrassica*; 6 – *Brassica oleracea* var. *botrytis*; 7 – *Brassica oleracea* var. *capitata* f. *rubra*; 8 – *Brassica oleracea* var. *acephala*

**Fig. 4.** Winter mortality rate of pupae of *Pieris brassicae*. Different letters above vertical bars (standard deviation) indicate significant differences among columns.

In the variants with Brussels sprouts (var. *gemmifera*), rutabaga (var. *napobrassica*) and cauliflower (var. *botrytis*), nearly half of the pupae died during the winter. In the control variant ('Podarok') and the variant with red cabbage (var. *capitata* f. *rubra*) winter mortality was 23% and 27%, respectively. The most resistant plant variant was kale (var. *acephala*) with the pupae of biggest body mass, where only 8% of them perished (Fig. 4).

## DISCUSSION

Considering the viability and fecundity of the offspring of LWB, caterpillar stage is the most important stage in its development cycle. During this period, energetic resources for all the following stages are collected and stored. The growth of body mass is the greatest in the last instar of caterpillars. According to Lafont et al. (1975) the body mass of the larvae increases about five times during the fifth instar. At that time caterpillars need the most of quality food. Food of poor quality forces the larvae to eat more food (Evans, 1940). The sufficient availability of food is also important. When other factors are favourable but food is limited the larval period increases. The moulting of larvae into a new instar or their pupation may occur only when the minimum amount of reserves required for the stage has been stored. In the case of unfavourable conditions, there develop individuals with smaller body mass than normal, whereas the worst feeding conditions are the main influencing factors. Our experiments showed that different foodplants have different effects on the growth of larvae. The most unsuitable food for larvae appeared to be both nasturtium and horse-radish. The caterpillars ate very little of nasturtium already in the first instars, however, the mortality rate became higher in the last instars. From the results it may be concluded that the plant contains toxic substances for the caterpillars. However, the amount of toxicants is so small that it does not cause immediate death. Instead, the caterpillars developed slow but constantly deepening intoxication. Toxic compounds force to invest resources in detoxification mechanisms that in turn incur growth and development costs (Kessler and Baldwin, 2002). The presence of a feeding deterrent in nasturtium has been demonstrated by Huang and Renwick (1995), and the major active compounds has been identified as chlorogenic acid (Renwick, 2001).

The amount and composition of nutrients containing in plants considerably influence the development of caterpillars. Vitamins containing in foodplants are very important for the development. For example, it is known that when larvae of LWB are fed on inner white leaves of cabbage, the larval stage lengthens and mortality rate increases. The reason here is considered to be the lack of vitamin A, or carotenoid pigments in the plant tissues that are essential for growth in insects (Feltwell, 1973, 1982). Also the lack of Fe and N induces the slowing of the development of caterpillars; the pupation period lengthens and the death rate increases (Allen, 1954). Chandra and Lal (1977), who studied the effect of several cabbage cultivars on caterpillars of LWB, did not find considerable differences in the growth of the caterpillars, but the food amounts eaten during the larval stage and the mortality rate of the caterpillars depended on the cultivars. Trying to rear caterpillars on plants not favoured by LWB as foodplants usually results in a high mortality rate. For instance, caterpillars fed on wallflower (*Cheiranthus cheiri*) died already during the second instar, and the cause of the death is believed to be cardiac glycosides in the plant

(Feltwell, 1982). Our experiments demonstrated that the suitable chemical composition of foodplants was important for the development of caterpillars of LWB. In the case of less suitable food, the caterpillar stage lengthened and the mortality rate increased.

Studies of feeding seldom measure the growth of body mass, as usually this does not give reliable results because of the rapid growth of body mass during eating and the reduction of the growth in the periods between eating and excretions. At prepupal stage body mass diminishes quickly, and, therefore, in a pupa it usually is only a half of the biggest weight of the larva of the last instar. Therefore, to obtain reliable data, the body mass of each pupa must be determined. Body mass is directly dependant on the amount of reserves stored at pupal stage, and pupae with small body mass appear when growing conditions in caterpillar stage are unfavourable (lack of food, great population density, etc.). In our experiments, caterpillars were provided with a sufficient amount of fresh food, and conditions in the growing chamber were near the optimum. That is why the different nutritive values of foodplants are reflected in different body mass of the pupae developed from caterpillars. It must be mentioned that there exists a correlative connection between the weight of the pupae and the ability of reproduction (Kramer, 1959), and the smaller the mass of a pupa, the less numerous its offspring.

A hibernating pupa loses its body mass mainly because of the loss of water. The water loss depends on the patterns of gas exchange, which, to a great extent, may vary individually. The more active the transpiration, the quicker the reduction of body mass. According to the data of K. Jõgar (1997), who has studied the gas exchange of hibernating pupae of LWB, most of the diapausing underweight pupae have continuous gas exchange; i.e. they do not have cyclical gas exchange. The pupae of this species of normal body mass show regular cycles of discontinuous gas exchange as a rule (Kuusik, 1977; Metspalu & Hiiesaar, 1980; Metspalu et al., 1982; Kuusik et al. 1995).

Discontinuous gas exchange cycles are often cited as being an adaptive mechanism for minimising respiratory water loss, especially for pupae in which opportunities for replenishing lost water are greatly limited (Hadley, 1994). Affected respiration may be one of the reasons why the water loss of underweight pupae often exceeds the critical limit (over 40% of the initial body mass) and causes the winter death of pupae. Pupae with great initial body mass were characterised by great endurance and an ability to survive hibernation. In our study the pupae reared on kale (*Brassica oleracea* var. *acephala*) had the biggest initial body mass (440 mg). In this variant, the winter mortality rate of pupae was the lowest.

It can be concluded from the study that one of the reasons for the great decrease in the abundance of LWB following the massive reproduction is the lesser viability of caterpillars growing on less valuable foodplants. The mortality rate of these caterpillars is high, the pupae are underweight and, in most cases, they perish during winter. In our experiments, we showed the high mortality rate of caterpillars feeding on nasturtium (*Tropaeolum majus*) and horse-radish (*Armoracia rusticana*), and their pupae were not able to survive winter conditions.

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