

Effects of Different Honeycomb and Sucrose Levels on the Development of Greater Wax Moth *Galleria mellonella* Larvae

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ABSTRACT

In this study, the effect of different honeycomb and sucrose amounts on the development of *Galleria mellonella* larvae has been investigated by using synthetic diet. These results showed that the *Galleria mellonella* larvae could tolerate different diets without any serious problem during their development. In a period of seven days maximum growth is determined on the larvae, which are fed the diet that contains 300 g honeycombs. In addition in ten days of experimental period maximum weight increase is 0.131 g and it is observed in the diet, which contains 50 g honeycombs. This increase in the diet that contains 300 g honeycombs is 0.118 g. At the end of the experimental period, the measures showed that the maximum weight increase appeared in diet of 15% sucrose as 0.0774 g. In the diet, which does not contain honey there was a weight loss as 0.0158 g.

Key Words: Honeycomb; Sucrose levels; *Galleria mellonella*

INTRODUCTION

The greater wax moth *Galleria mellonella* (Lepidoptera: Pyralidae) is an important pest of the honeybee. The larval stage of the *Galleria mellonella* feeds on the honey, pollen and wax produced by honeybees (Nurullahoglu & Susurluk, 2001). However, an effective method of control of this pest has not been developed. Physical, chemical and biological methods are imperfect (Cantwell & Smith, 1970; Ali *et al.*, 1973; Burges, 1977, 1978). Beekeepers see the wax moth as a pest. Today the wax moth *Galleria mellonella* can be named as the main negative factor on beekeeping and studies on increasing the production. In order to maintain the ecologic balance biologic control preferred rather than chemical control.

Galleria mellonella larvae development period is 40 days but this value can varies due to the environmental conditions. Optimal conditions embryonic development is completed in 3 - 4 days. But this period can extend to 25 days, because of the environmental conditions. The opening of the eggs is completed in 8 - 10 days. Egg incubation period can be affected by temperature, moisture and nutrient quality (Ozer, 1962). Each female lay approximately 500 eggs throughout its life cycle. The larvae emerged from the eggs distribute in the hive and create nutrition tunnels in honeycomb. Mature larvae can be 2.2 cm long. At the end of this phase they continue their life cycles by forming pupas in or out of the hive (Burges, 1978).

Galleria mellonellae is preferred in entomologic studies, because of its nutritional needs, ecologic adaptation and development characteristics. It is used as a host in favor of many hymenoptera species.

The solitary endoparasitoid wasps are *Pimpla*

turionellae and *Itopectis naranyae*, which attack various lepidopteran pupae and prepupae (Townes *et al.*, 1965; Ueno, 1999; Coskun *et al.*, 2005). Because of the natural host of *P. turionellae* are difficult to mass rear and because *P. turionellae* is a polyphagous parasitoid, it is possible to utilize alternative host that are easily cultured in the laboratory for mass rearing of the parasitoid. *G. mellonella* is a suitable of hosts for some parasitoids related to *P. parmarae* and *P. turionellae* (Sandlan, 1979a, 1982; Ueno, 1999).

Different kinds of synthetic nutrients are developed in order to produce wax moth. Mass propagation of many insects produced invented host *G. mellonella* has been accomplished by using diets development by (Balazs, 1958; Beck, 1960; Dutky *et al.*, 1962; King *et al.*, 1979) (Bratti & Costantini, 1991; Gross, 1994; Gross *et al.*, 1996). Gross (1994) reported that only about 60 - 65% of the *G. mellonella* larvae reared on the diet of King *et al.* (1979) produced adult parasitoids and that mature *G. mellonella* larvae weighing less than 100 mg. Our research was designed to determine the relative nutritional values of various ingredients in the Bronksill (1961) *G. mellonella* diet for producing strong *G. mellonella* larvae and consequent *Pimpla turionellae* adults. These kinds of studies are needed for biological control.

MATERIALS AND METHODS

The *Galleria mellonella* larvae, which are used in the experiment are produced with the food that is developed by Bronksill (1961). This food is kept in jars and its component is given in Table I.

Mature *G. mellonella* males and females, which are

Table I. Control diets Used for rearing *Galleria mellonella*

Ingredient	Amounts per Diet
Bran	500g
Glycerin	300 ml
Honeycomb	200g
Pure Water	150 ml
Honey	150 ml

taken from the beehives are produced in these jars with the conditions of 26 - 28°C, 50 - 60% relative moisture and darkness.

In order to show the effect of different wax ratios on the development of *G. mellonella* larvae the nutrient in Table I is used as the control diet. The honeycomb amounts are decreased by 100%, 87.5%, 75% and 50% and increased by 50%. So by this way six different kinds of diets are prepared with control. So the honeycomb amounts are appeared to be 0 g, 25 g, 50 g, 100 g, 200 g and 300 g. *G. mellonella*, which are taken from the stock culture are weighed and ten *G. mellonella* is put in each jar. On the 3rd, 7th and 10th day are weighed and the ratios of the larvae that did not pass the pupae phase are determined.

In order to study the effects of different sucrose ratios on the development of *G. mellonella* larvae the food in Table I is used as the control diet. Honey is taken out from diet and sucrose is added instead of honey with ratios of 0%, 5%, 10%, 15% and 20%. So six different diets took place.

The larvae, which are taken from the stock culture are weighed and ten larvae are put in each jar. On day 6, 8 and 13 these larvae are taken the jars and their weight changes are compared with the control group.

RESULTS

The effect of different honeycomb and sucrose amounts on the development of *Galleria mellonella* larvae are given (Table II & Table III).

In the larvae, which are fed with the diets that contain different amounts of wax, at the end of the 3 days experiment period, the greatest increase on weight is seen in the larvae, which are fed with the control diet as 0.029 g. In the insects, which are fed with diets that do not contain honeycomb a loss of weight such as 0.003 g is seen. There is not significant difference in weight increases of the larvae, which are fed with the diet that contained 25 g and 300 g honeycomb. The weight increase of the larvae, which are fed with diet that contained 100 g honeycombs is appeared to be very much lower than the control group.

In a period of seven days maximum growth is determined on the larvae, which are fed the diet that contains 300 g honeycombs. The weight increase in control, 100 g (honeycomb) 50 g (honeycomb) diets are 0.053 g, 0.051 g, 0.049 g in order. In also 25 g honeycomb diet it is 0.021 g and in the diets, which do not contain honeycombs the weight increase is 0.013 g.

In ten days of experimental period maximum weight increase is 0.131 g and it is observed in the diet, which contains 50 g honeycombs. This increase in the diet that contains 300 g honeycombs is 0.118 g; in control diet it is 0.096 g and the diet that contains 100 g and 25 g are 0.095 g and 0.051 g. Also this increase appeared to be 0.024 g in the diet, which does not contain honeycomb.

At the end of six days of experimental period the maximum weight increase in the insects, which are fed with diets that contain different amounts of sucrose is observed in the diet, which contained 20% sucrose as 0.0838 g. Weight loss appeared only in the diet, which contained 5% sucrose as 0.0348 g.

At the end of eight days of experimental period maximum weight increase appeared to be in the control group as 0.0705 g.

On the 13th day of the experimental period, the measures showed that the maximum weight increase appeared in diet of 15% sucrose as 0.0774 g. In the diet, which does not contain honey there was a weight loss as 0.0158 g and this loss was 0.0004 g in 5% sucrose diet. Increase in the control group was 0.0029 g and it was 0.0586 g and 0.0333 g in 10% and 20% sucrose diets.

DISCUSSION

The effects of different wax and sucrose ratios on the development of *G. mellonella* larvae are examined and it is seen that all of diets the larvae completed their growth. But especially in the ratios, which honeycomb level is lower than the control, because of the nutritional deficiency the insects made pupae earlier.

However the maximum increase in weight is seen in the diet with the ratio of 50 g and weight per larvae reached to 0.191 g from 0.060 g nevertheless, in the same diet at the end of the experiment nine larvae passed to the early pupa phase. The extra weight increase might mean that the larva that is survived is much stronger than the others. Gross *et al.* (1996) reported that the addition of 30 g of wheat germ to the standard diet was necessary to significantly increase the weight of mature *G. mellonella* larvae. In addition mature *G. mellonella* larvae that developed on the diet containing 5 - 10 g torula yeast and *Archytas marmoratus* adults consequently emerging from those larvae were considerably heavier than their counterparts reared on the standard diet lacking torula yeast.

In many parasitoids, positive relationships are found between parasitoid size and host size (King, 1987). In addition host size is one factor that influences the emergence sex ratio (Ueno, 1998; King, 2002).

The increase of honeycomb amount in the diet with a ratio of 50% effected the larvae development positively and decreased the transfers to early pupa phase than the other diets. It is seen that the increase in weight is directly proportional with the honeycomb amount and this data can be noted as very important one. It is very common situation

Table II. The Effects of Different Honeycomb Levels on the Development of Greater Wax Moth Larvae

Honeycomb (g)	Initial no. of larvae	Weight (g)	Day 3		Day 7		Day 10	
			Survival no. of larvae	Weight (g)	Survival no. of larvae	Weight (g)	Survival no. of larvae	Weight (g)
0,00	10	0,057	6	0,054	3	0,070	2	0,081
25	10	0,051	8	0,064	3	0,072	3	0,102
50	10	0,060	6	0,067	2	0,109	1	0,191
100	10	0,056	9	0,059	5	0,107	3	0,151
200 *	10	0,060	8	0,089	6	0,113	2	0,156
300	10	0,060	8	0,072	7	0,133	3	0,178

* Control diet

Table III. The Effects of Different Sucrose Levels on the Development of Greater Wax Moth Larvae

Sucrose %	Initial no. of larvae	Weight (g)	Day 6		Day 8		Day 13	
			Survival no. of larvae	Weight (g)	Survival no. of larvae	Weight (g)	Survival no. of larvae	Weight (g)
Control	10	0,0781	7	0,1729	3	0,1486	2	0,0810
0,00	10	0,1264	5	0,1313	3	0,1070	1	0,1106
5	10	0,1379	4	0,1031	3	0,1928	1	0,1375
10	10	0,0819	5	0,1621	4	0,1287	1	0,1405
15	10	0,0741	7	0,1091	4	0,1287	3	0,1515
20	10	0,0863	4	0,1701	3	0,1272	1	0,1196

to add or remove the diet compounds (Tsiropoulos, 1992) in the studies of the development of the insect reproduction, egg number and efficiency. Similar results were also obtained with *Cryptolaemus montroussieri* (Chumakova, 1962), *Dacus olea* (Tsiropoulos, 1977) and *Melanogryllus desertus* (Başhan & Balcı, 1994). Moreover, sex ratio of parasitic hymenopters has close relationship with host size (Sandlan, 1979a, b & 1980; Kazmer & Luck, 1995).

By this way we can show as if the diet is accepted by insect and as if it was prepared properly. The negative effect of the removal of the honeycomb from the diet (with different ratios) on the development shows us that these diets can not give the insects their nutritional needs properly. The diets should require the nutritional compounds such as carbohydrates, protein and lipids.

This is the reason of the loss weight in the diets, which do not include honey and sucrose. It is also shown that sucrose and honey have a negative effect on the development and the production system of the insects. However because of the decrease in the protein and glycogen amounts (with the absence of honey & sucrose) evident decreases in the lifetime of insects are observed. The reduced weight of parasitoids emerging from *Musca domestica* with heavy parasitoid loads was probably attributable to a scramble type competition for limited host resources, where each parasitoid larva received less than optimal quantities of food for their own development (Waage & Ng, 1984; Taylor, 1988; Harvey & Gols, 1998).

Endoparasite insect species need other insects as hosts in the part of their own life cycle. These features of the insects increase their economical importance and make it possible to use them for biological control programs. The control of pests by the predators depends on having healthy hosts for the predators and high number of populations.

Only an increase of sucrose amount in the diet as 15%

has a positive effect on larval development. For an optimum nutrition it is not enough to take all compounds that are needed. At the same time these compounds should be in balance for the metabolism. Nutritional balance is very important for insects in both the larval and mature levels. This study also shows the nutritional needs of *Galleria mellonella* and it is significant in the biologic control.

REFERENCES

- Ali, A.D., N.M. Barky, M.A. Abdelatif and S.K. EJ-Sawaf, 1973. The control of greater wax moth, (*Galleria mellonella* L.) by chemicals, *I. Z. Angew. Entomol.*, 74: 170–7
- Balazs, A., 1958. Nutritional and nervous factors in the adaption of *Galleria mellonella* to artificial diet. *Acta Biol. Acad. Sci. Hung.*, 9: 47–69
- Başhan, M. and K. Balcı, 1994. The effects of wheat germ oil, vitamin E and some fatty acids on the reproduction of *Melanogryllus desertus* Pall. *Tr. J. Zool.*, 18: 147–51
- Beck, S.D., 1960. Growth and development of the greater wax moth, *Galleria mellonella* (L.) (Lepidoptera: Galleriidae). *Trans. Wis. Acad. Sci. Arts Lett.*, 49: 137–48
- Bratti, A. and W. Costantini, 1991b. Effects of new artificial host diets on the host-parasitoid system *Galleria mellonella* L. (Lepidoptera: Galleriidae) - *Archytas marmoratus* Town. (Diptera: Tachinidae). *Redia*, 74: 445–8
- Bronskill, J.K., 1961. A cage to simplify the rearing of the greater wax moth, *Galleria mellonella* (Pyralidae). *J. Lep. Soc.*, 102–4
- Burges, H.D., 1977. Control of the wax moth *Galleria mellonella* on beecomb by H-stereotype V *Bacillus thuringiensis* and effect of chemical additives. *Apidologie*, 8: 155–68
- Burges, H.D., 1978. Control of wax moth: Physical and biological methods. *Bee World*, 59: 129–38
- Cantwell, G.E. and L.J. Smith, 1970. Control of the greater wax moth *Galleria mellonella* in honeycomb and comb honey. *American Bee J.*, 10: 141–3
- Chumakova, B.M., 1962. Significance of individual food components for the vital activity of mature predatory and parasitic insects. *Ecol. Kievsk.*, 8: 133–4
- Coskun, M., P. Ozalp, M. Sulanc and I. Emre, 2005. Effects of various diets on the oviposition and sex ratio of *Pimpla turionellae*. *Int. J. Agric. Biol.*, 7: 129–32

- Dutky, S.R., J.V. Thompson and G.E. Cantwell, 1962. A Technique for mass rearing the wax moth (Lepidoptera: Galleriidae). *Proc. Entomol. Soc. Wash.*, 64: 56–8
- Gross, H.R., 1994. Mass propagation of *Archytas marmoratus* (Diptera: Tachinidae). *Environ. Entomol.*, 23: 183–9
- Gross, H.R., C.E. Rogers and J.E. Carpenter, 1996. Development of *Archytas marmoratus* (Diptera: Tachinidae) reared in *Galleria mellonella* larvae (Lepidoptera: Pyralidae) feeding on selected diets. *Biological Control*, 6: 158–63
- Harvey, J.A. and G.J.Z. Gols, 1998. The influence of host quality on progeny and sex allocation in the pupal ectoparasitoid, *Muscidifurax raptorellus* (Hymenoptera: pteromalidae). *Bulletin Entomol. Res.*, 88: 299–304
- Kazmer, D.J. and R.F. Luck, 1995. Field tests of the size-fitness hypothesis in the egg parasitoid *Trihogramma pretiosum*. *Ecology*, 76: 412–25
- King, B.H., 1987. Offspring sex ratio in parasitoid wasps. *Q. Rev. Biol.*, 62: 367–96
- King, B.H., 2002. Offspring sex ratio and number in response to proportion of host sizes and ages in the parasitoid wasp *Spalangia cameroni* (Hymenoptera: Pteromalidae). *Environ. Entomol.*, 31: 505–8
- King, E.G., G.G. Hartley, D.F. Martin, J.W. Smith, T.E. Summers and R.D. Jackson, 1979. *Production of the Tachinid Lixophaga Diatraeae on its Natural Host, the Sugar Cane Borer and on its Un-natural Host, the Greater Wax Moth*. U.S. Department of Agriculture, Science and Education Administration, Advances in Agricultural Research Service Handbook No. 693
- Nurullahoglu, U.Z. and A.İ. Susurluk, 2001. Fecundity of Turkish and German strains of *Galleria mellonella* (L.) (Lepidoptera: Pyralidae) reared on two different diets. *S.U. Fen-Edebiyat Fakültesi Fen Dergisi*, 18: 39–44
- Ozer, M., 1962. Arı kovanlarında önemli zarar yapan balmumu güvesi (*Galleria mellonella* L.)'nin morfoloji, biyoloji ve yayılışı üzerine araştırmalar. *Bitki Koruma Bülteni*, 2: 26–36
- Sandlan, K.P., 1979a. Sex Ratio in *Coccygomimus turionellae* Linnaeus (Hymenoptera: Ichneumonidae) and its ecological implication. *Ecol. Entomol.*, 41: 365–78
- Sandlan, K.P., 1979b. Host feeding and its effects on the physiology and behavior of the Ichneumonidae Parasitoid, *Coccygomimus turionellae*. *Physiol. Entomol.*, 4: 383–92
- Sandlan, K.P., 1980. Host Location by *Coccygomimus turionellae* (Hymenoptera: Ichneumonidae). *Ent. Exp. Et. Appl.*, 27: 233–45
- Sandlan, K., 1982. Host suitability and its effects on biology in *Coccygomimus turionellae* (Hymenoptera: Ichneumonidae). *Ann. Entomol. Soc. America*, 75: 217–21
- Taylor, A.D., 1988. Host effect on larval competition in the gregarious parasitoid *Bracon hebetor*. *J. Animal Ecol.*, 57: 163–72
- Townes, H., S. Momoi and M. Townes, 1965. A catalogue and reclassification of the eastern Palearctic Ichneumonidae. *Mem. American Entomol. Inst.*, 5
- Tsiropoulos, G.J., 1977. Survival and reproduction of *Dacus olea* fed on chemically defined diets. *Z. angew. Entomol.*, 84: 192–7
- Tsiropoulos, G.J., 1992. Feeding and dietary requirements of the Tephritid Fruit Flies, advances in insect rearing for research and pest management. In: Thomas, E.A. and C.L. Norman (eds.), *Westview Press Chapter*, 7: 93–118
- Ueno, T., 1999. Multiparasitism and host feeding by solitary parasitoid wasps (Hymenoptera: Ichneumonidae) based on the pay-off from parasitized hosts. *Ann. Entomol. Soc. America*, 92: 601–8
- Ueno, T., 1998. Adaptiveness of sex ratio control by the pupal parasitoid *Itopectis naranyae* (Hymenoptera: Ichneumonidae) in response to host size. *Evolutionary Ecol.*, 12: 643–54
- Ueno, T., 1999. Host suitability and sex-ratio differences in wild-caught and laboratory-reared parasitoid *Pimpla parnarae* (Hymenoptera: Ichneumonidae). *Ann. Entomol. Soc. America*, 92: 609–14
- Waage, J.K. and S.M. Ng, 1984. The reproductive strategy of a parasitic wasp. I. Optimal progeny allocation in *Trichogramma evanescens*. *J. Animal Ecol.*, 53: 401–15

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