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# The Effect of Some Sugar Supplemented Diet with Various Sucrose Ratios on the Life Cycle and Egg Productivity of the Adult Females of *Pimpla turionellae* L. (Hymenoptera: Ichneumonidae)

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## ABSTRACT

This study aimed to investigate the effect of nutrition which consisted of 7% of ribose, rhamnose, arabinose, and xylose supplemented with 0, 7, and 14% of sucrose respectively on the life cycle and egg production of the adult females of *Pimpla turionellae.* In all experimental combinations, the nutritions containing 7% of ribose, rhamnose, arabinose, and xylose had an effect on the insects to which lived approximately 9 days and the addition of sucrose could extend this period in some way. The time extension was significantly at low level compared to the control (except for the combination of 7% ribose and 14% sucrose). All nutrient combinations caused remarkable decreases in egg productivity of the insect. The nutrition, except 7% arabinose and 7% sucrose combination, did not affect egg hatching.

Results of this study suggest that *P. turionellae* require sucrose rather than ribose, rhamnose, arabinose, and xylose to optimise the life cycle and egg production. We conclude that future researchers should investigate the effect of sugar at these levels, but also that we need a better understanding of which proportion of toxic sugar will reduce pest populations in the field.

*Key words: Pimpla turionellae,* toxic, sugars, life cycle, egg number, egg hatching.

## INTRODUCTION

Biological control studies are one of the leading factors to avoid the limitless growth of insect populations in nature. Many hymenopteran species are commonly used against lepidopteran species. Parasitoids have to be produced in large numbers and fed with synthetic diet under laboratory conditions to be able to use them in biological control studies. Hence more practical and synthetic diets and techniques are required for this purpose.

Parasitoids, like other insects, need various sources of nutrition at their different growth stages and the components of the nutrition for egg productivity at the adult stage are closely related to the feeding regimes (Murugan & George, 1992; Zucoloto, 1992; Jing *et al*., 2001).

The feeding of parasitic hymenopter species with nutritions highly rich in carbohydrates such as plant sap gives rise to the nutrient ingredient (Haydak, 1970).

In general, the sugars such as ribose, rhamnose, arabinose, and xylose are not useful and have a toxic effect in some insect species (Nettles, 1972; Barker & Lehner, 1974; Ozalp & Emre, 2001). Moreover, it is important to remember that the toxicity level may vary according to age, sex, type and amount of usable carbohydrate in nutritions although there is no certain evidence. For example, toxic effects of arabinose in the *Anthonomus grandis* adults (Nettles, 1972) and xylose in the *Tribolium castaneum* adults (Applebaum, 1972) were diminished by the addition of sucrose and glucose in the diet, respectively.

Carbohydrate contents of the diet should be bioavailable to consider the diet as useful. Carbohydrates should be used together with a highly phagostimulant to be able to understand if they are to be useful. For example; it is hard to define the useful sugars if sucrose is not present in the study where *Myzus persica* is common (Mitler *et al*., 1970).

In this study, the effect of synthetic nutritions, individually prepared with 7% of ribose, rhamnose, arabinose, and xylose in which 0, 7, and 14% of sucrose having high phagostimulant were added, was investigated on the life cycle and egg productivity of the *P. turionellae* adults. The experiments were also designed to search the effect of two kinds of nutritions, prepared with four different sugars known as toxic and mixed with various concentrations of sucrose as highly phagostimulant, on the life cycle and egg productivity of the *P. turionellae* adults.

## MATERIALS AND METHOD

In this study, maintenance of the stock culture, on which test insects were reared for this study, was obtained by feeding insects on 50% honey and haemolymph of *Galleria mellonella* under laboratory conditions with 50% relative humidity at 24±2 °C under 12:12 hrs light: dark photoperiod. Adult *P. turionellae* used in this test was obtained from parasited *G. mellonella* pupae under same laboratory conditions. The present study was conducted with 100 ml of synthetic diet complex (Emre, 1988) with known chemical composition, which consisted of 3.000 g L- amino acid mix, 0.284 g water soluble vitamin mix, 0.075 g inorganic salt mix, 0.075 g RNA, 14.000 g sucrose, was used as the control nutrition. The chemical structure of the synthetic diet is given in Table 1.

The sucrose concentration was arranged as 0, 7, and 14% in the control diet and 7% of D(-) arabinose, D(+) xylose, D(-) ribose, L(+) rhamnose were individually added into three nutrition combinations to feed the *P. turionellae* adults. Non-fed, its juveniles of *P. turionellae* individuals of the same day were used in each experiment and its replication. Four females and two male individuals were placed in a 1000 cc-glass jar tightly covered by two layers of cheesecloth and they were fed through this system. Experimental diet preparations were equally divided with thin pipette dispensers onto aluminum foils (3x3 cm) and the insects were fed for one hour daily at the same time of the day during the experiments.

The data collected was evaluated for the effect of both control and experimental nutritions on the insect life cycle, egg productivity, and egg hatching. The insects under the experimental circumstances were parasitized starting on the 10$^{th}$ day of the experiment and this was repeated every 3 days until the females died. The pupae of *G. mellonella* were used to get more eggs from the *P. turionellae* females, by parasiting them. The pupae were given into the environment wrapped by double layers of window screen to force the *P. turionellae* egg production and to prevent the insects feeding on hemolemph of the pupae. After they were parasitized, the pupae were taken out from glass jars and placed in a petri dish for 24 hours to complete their embryonic development in darkness under laboratory conditions.

Table 1. Composition of the chemically defined diet for feeding of the adults of *P. turionellae.*

| Constituent | mg/100 ml diet | Constituent | mg/100 ml diet | Constituent | mg/100 ml diet |
|---|---|---|---|---|---|
| L-Amino acid mixture | 3000.00 | Water soluble vitamin mixture | 284.38 | Inorganic salt mixture | 75.00 |
| Alanine | 210.00 | Ascorbic acid | 10.6105 | $FeCl_3$ $6H_20$ | 2.1583 |
| Arginine-HCl | 150.00 | Biotin | 0.0379 | $K_2HPO_4$ | 45.0129 |
| Aspartic acid | 195.00 | Ca-Panthothenate | 2.8042 | $Na_2HPO_4$ $12H_2O$ | 6.2201 |
| Cysteine | 39.00 | Choline chloride | 246.3158 | $MgSO_4$ $7H_2O$ | 15.7853 |
| Glutamic acid | 315.00 | Folic acid | 0.1137 | $MnSO_4$ $H_2O$ | 0.0479 |
| Gycine | 192.00 | Inositol | 17.0526 | $CoCl_2$ $6H_2O$ | 0.5798 |
| Histidine | 120.00 | Nicotinic acid | 5.6842 | $CuSO_4$ $5H_2O$ | 0.6721 |
| Hydroxproline | 57.00 | Pyridoxine-HCl | 0.2842 | $CaCl_2$ | 3.6684 |
| Isoleucine | 156.00 | Riboflavin | 1.3263 | $ZnCl_2$ | 0.8552 |
| Leucine | 231.00 | Thiamine-HCl | 0.1516 | | |
| Lysine | 159.00 | | | Miscellaneous | |
| Methionine | 90.00 | Lipid mixture | 540.96 | Ribonucleic acid | 75.00 |
| Phenylalanine | 165.00 | Cholesterol | 138.8430 | Sucrose | 14.000.00 |
| Proline | 246.00 | Linoleic acid | 8.0331 | 2N KOH | 280.00 |
| Serine | 195.00 | Linolenic acid | 25.5537 | 2N $K_2HPO_4$ | 14.03 |
| Threonine | 165.00 | Oleic acid | 10.5950 | Distilled water to 100 ml | |
| Tryptophane | 60.00 | Palmitic acid | 0.6777 | | |
| Tyrosine | 120.00 | Stearic acid | 0.2314 | | |
| Valine | 135.00 | Tween 80 | 357.0248 | | |

The *G. mellonella* pupae were dissected in a petri dish with 0.8% NaCl. The eggs were gently removed with a soft brush, counted in the presence of 0.8% NaCl. They were then left for 24 hours under laboratory conditions. Afterwards, the numbers of hatched eggs was determined by microscopy observations. All experiments were repeated three times for each of the nutrition complex with different sugar concentration. The total number of eggs was divided by the number of female individuals in the experiment to calculate the number of eggs per female. The percentage of the hatched eggs according to the total number of eggs was used for the ratio of egg hatching. Moreover, the life cycle of the insects was calculated according to the number of days starting from the first day on which the insect were placed into the glass jars until the insect deaths occurred. The insects which escaped from the jars and/or died due to experimental mistakes were not taken into consideration for the data evaluation. The "t" test for the comparison of dual groups (Sokal & Rolhlf, 1969) and "Student-Newman Keul's test" (SNK) for the comparison of other groups were used for the statistical analyses of the data collected from the experiments. The differences among means were accepted as significant at $p< 0.05$.

## RESULTS

The effect of four different toxic sugars on the life cycle and egg productivity of the *P. turionellae* adult females are individually presented in Table 2. The nutrition complexes except 7% ribose + 14% sucrose significantly decreased the insect's life cycle compared with the control group. It was about 9 days when sucrose was not added into the nutrition media and was significantly longer with the sucrose added diet; however, this was still quite a shorter life when compared to the control. No results were obtained from the nutrition without sucrose in egg productivity and hatching experiences due to the insects having died 10 days prior to egg lying. The number of eggs showed a remarkable decrease in all the diets compared to the control. The lowest egg number was about 2.72 in 7% xylose + 7% sucrose whereas it was around 43.59 in 7% arabinose + 14% sucrose fed group (Table 2). Hatching ratio was significantly decreased only in %7 arabinose + %7 sucrose fed individuals. The other diets also caused some fluctuations in egg hatching ratio but the differences were not statistically important.

Table 2. Effects of different toxic sugars and sucrose ratios on the life cycle, egg number and egg hatching of the *P. turionellae* adult females.

| | Life cycle (day) | | Egg number | Egg hatching (%) |
|---|---|---|---|---|
| Nutrition | Min-Max | $\overline{X} \pm s\overline{x}$ * | $\overline{X} \pm s\overline{x}$ * | $\overline{X} \pm s\overline{x}$ * |
| Control | 44-58 | 51.66 ± 2.12 a | 53.66 ± 1.33 a | 69.46 ± 1.81 a |
| 7% ribose | 9-10 | 9.66 ± 0.33 c | - | - |
| 7% ribose + 7% sucrose | 33-37 | 34.66 ± 1.20 b | 19.88 ± 0.46 c | 76.67 ± 2.73 a |
| 7% ribose + 14% sucrose | 49-52 | 50.33 ± 0.88 a | 28.84 ± 0.51 b | 72.73 ± 1.10 a |
| Control | 42-56 | 50.24 ± 1.98 a | 52.24 ± 1.33 a | 68.38 ± 1.35 a |
| 7% rhamnose | 8-10 | 9.00 ± 0.57 c | - | - |
| 7% rhamnose + 7% sucrose | 19-20 | 19.66 ± 0.33 b | 7.03 ± 0.39 b | 58.94 ± 2.74 a |
| 7% rhamnose + 14% sucrose | 17-19 | 18.00 ± 0.57 b | 8.73 ± 0.28 b | 71.79 ± 6.39 a |
| Control | 45-61 | 55.33 ± 2.12 a | 54.42 ± 1.33 a | 67.80 ± 1.81 a |
| 7% arabinose | 8-9 | 8.66 ± 0.33 d | - | - |
| 7% arabinose + 7% sucrose | 20-23 | 21.66 ± 0.88 c | 7.60 ± 0.44 c | 53.48 ± 4.36 b |
| 7% arabinose + 14% sucrose | 41-44 | 42.66 ± 0.88 b | 43.59 ± 0.19 b | 73.23 ± 1.75 a |
| Control | 48-57 | 54.33 ± 1.70 a | 56.24 ± 1.78 a | 70.24 ± 2.02 a |
| 7% xylose | 9-10 | 9.33 ± 0.33 c | - | - |
| 7% xylose + 7% sucrose | 25-29 | 26.33 ± 1.33 b | 2.72 ± 0.14 b | 60.00 ± 2.50 a |
| 7% xylose + 14% sucrose | 28-35 | 33.33 ± 2.02 b | 4.56 ± 0.34 b | 75.94 ± 4.90 a |

* Values in columns not followed by the same letter are significantly different at level P<0.05.

$\overline{X} \pm s\overline{x}$ * Arithmetic mean ± Standard deviation.

## DISCUSSION

In this work, a synthetic diet with known chemical composition (Emre, 1988) was used as the control diet and twelve kinds of nutrition complexes prepared by mixing toxic sugars (7% of ribose, rhamnose, arabinose, and xylose) and three kinds of diets containing sucrose, actual sugar of this nutrition, at various concentrations (0, 7, and 14%) were investigated for their effects on the life cycle, egg productivity and egg hatching of the *P. turionellae* adult females.

The major conclusions of this study are that the sucrose significantly increases the life cycle, egg productive of *P. turionellae* in favour of females, and that it also

significantly increases egg hatching of the individuals. Thus this study improves our understanding of the role of sugars in continuance of insects required for biological control. It is also the first study to report of sucrose with toxic sugars for the adult stages of *P. turionellae* on meridic diets.

The data obtained from our study shows that there is a positive relationship between sucrose and lifetime. Previous studies have shown that the development, growth, reproduction, and behavior of insects are closely related to the quantity and quality of food, and it is fundamental that dietary ingredients are well balanced (Heimpel *et al.,* 1997; Idris & Grafius, 1997; Chang, 2004; Magro *et al*., 2006).

The *P. turionellae* adult females died within 10 days when they were fed with the mix of 7% ribose, rhamnose, arabinose, and xylose as carbohydrate source. On the other hand, the insects fed with the diet lacking carbohydrate lived about 9 days (Emre & Yazgan, 1990) and these results showed that *P. turionellae* females were not capable of using four of these sugars like other insect species (Fraenkel, 1940, 1955; Chippendale & Reddy, 1974; Friend *et al*., 1988; Mitler *et al*., 1970).

It was suggested that the reasons fort the unavailability to use these pentoses by various insects are the lack of availability in absorbing them through the intestine membrane, even if they are absorbed their unavailability to metabolic processes or their inhibition effect feeding (Fraenkel, 1955; Gordon, 1974; Tsiropoulos, 1980; Emre, 1984). Moreover, the toxic effect of arabinose, xylose, and ribose in some insect species can be explained by their activity as ant metabolites during sugar transport (Nettles, 1972). In the present study addition of these pentoses with sucrose into the diet had extended the life cycle of the insect whereas the one with 7% ribose + 14% sucrose brought the life cycle to control levels. Hence, our results also suggested that the tested pentoses had an inhibition effect on feeding. Similarly, rhamnose had inhibiting effects on the growth and survival of the black cricket *Melanogryllus desertus* Pall (Mehmetoğlu & Başhan, 1996). In one of the very few inclusive evaluations of sugar consumption by parasitoids, Wäckers (2001) tested the effects of 14 sugars on longevity of *Cotesia glomerata.* The longest lifespan was recorded with sucrose, fructose and glucose. Furthermore some sugars, such as lactose and raffinose were not utilized by parasitoid whereas rhamnose significantly reduced longevity of wasps. These studies supported our data.

In conclusion, these sugars had a negative impact in the number of eggs and life cycle of *P. turionellae* which also caused unfertile egg production. There are some other studies supporting these findings (Tsiropoulos, 1980; Özalp & Emre, 2001) which mention that the shortest life span was 5.33 days observed with arabinose followed by ramnose, xylose, sorbose, ribose and mannose. These results support that our findings. The best effect on egg productivity was obtained from the sucrose added-nutrition with arabinose which also resulted in long life cycle. The best results among the carbohydrates tested was obtained with sucrose which was also employed as a control (Özalp & Emre, 2001 ). Similar results were obtained from other studies for instance; sucrose prolonged the lifespan in *Cotesia glomerata* females by more than 35 hours (Hausmann *et al*., 2005). There was no significant difference on egg hatching between 7% ribose + 7% sucrose and 7% rhamnose + 7% sucrose fed

group compared to the control. Previous studies have shown that diet, particularly carbohydrate, affects insect lifetime (Fadamiro & Heimpel, 2001; Özalp & Emre 2001; Wäckers, 2001; Jacob & Evans, 2004; Onagbola *et al*., 2007). In the synthetic diet we used for *P. turionellae,* sucrose, which is a strong phagostimulant (Wäckers, 1999), was used as a carbohydrate source because Özalp & Emre (2001) have shown sucrose to be the most beneficial among the 23 different carbohydrates tested for their effect on *P. turionellae* lifetime. We suggest that future studies be conducted to find out the optimal quantity of sucrose to eliminate the toxic sugar effects, as the carbohydrate resource, in meridic diets for *P. turionellae*. It is concluded that these sugars with descending sucrose concentration can be used in *P. turionellae*'s diet.

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## REFERENCES

Applebaum, S. W., 1972, The carbohydrate nutrition of *Tribolium. In*: Rodriguez, Y. G. (Ed.). The insect and mite nutrition. North- Holland-Amsterdam. 287-297.

Barker, R. J., Lehner, Y., 1974, Acceptance and sustenance value of naturally occurring sugars fed to newly emerged adult workers of honey bees (*Apis mellifera* L.). *Journal of Experimental Zoology*, 187: 277-285.

Chang, L. C., 2004, Effect of amino acids on larvae and adults of *Ceratitis capitata* (Diptera:Tephritidae). *Annual Entomological Society of America*, 9: 529-535.

Chippendale, G. M., Reddy, G. P. V., 1974, Dietary carbohydrates: role in feeding behaviour and growth of the southwestern corn borer, *Diateria grandiosella*. *Journal of Insect Physiology*, 20: 751-759.

Emre, I., 1984, *Locusta migratoria migratorioides* R ve F nimflerinin besin almasına karbohidratların etkisi. *Doğa Bilim Dergisi*, 8: 56-60.

Emre, I., 1988, Meridik bir besinin *Pimpla turionellae* L. (Hymenoptera: Ichneumonidae) ergin dişilerinin yumurta verimine etkisi. *Doğa Bilim Dergisi*, 12: 101-105.

Emre, I., Yazgan, S., 1990, Effects of Dietary Components on Reproduction of *Pimpla turionellae* L. (Hymenoptera: Ichneumonidae). *Doga Turkish, Journal of Biology*, 14: 94-104

Fadamiro, H. Y., Heimpel, G. E., 2001, Effects of partial sugar deprivation on lifespan and carbohydrate mobilization in the parasitoid *Macrocentrus grandii* (Hymenoptera: Braconidae). *Annals of the Entomological Society of America*, 94: 909-916.

Fraenkel, G., 1940, Utilization and digestion of carbohydrates by the adult blowfly. *Journal of Experimental Biology*, 17: 18-29.

Fraenkel, G., 1955, Inhibitory effects of sugars on the growth of the Mealworm,*Tenebrio molitor* L. *Journal of Cell Biology*., 45: 393-408.

Friend, W. G., Schmidt, M. J., Smith, J. J. B., Tanner, R. J., 1988, The effect of sugars on ingestion and diet destination in *Culiseta inornata*. *Journal of Insect Physiology*, 34: 955-961.

Gordon, H. T., 1974, Carbohydrate utilization and phagostimulation in *Oncopeltus fasciatus*. *Entomologia Experimentalis et Applicata*, 17: 450-451.

Hausmann, C., Wäckers F. L., Dorn, S., 2005, Sugar convertivility in the parasitoid *Cotesia glomerata* (Hymenoptera: Braconidae). *Archives of Insect Biochemistry and Physi*ology, 60: 223–229.

Haydak, M. H., 1970, Honey bee nutrition. *Annual Review of Entomology*, 15: 143-156.

Heimpel, G. E., Rosenheim, J. A., Kattari, D., 1997, Adult feeding and lifetime reproductive success in the parasitoid *Aphytis melinus*. *Entomologia Experimentalis et Applicata*, 83: 305-315.

Idris, A. B., Grafius, E., 1997, Nectar-collecting behavior of *Diadegme insulare* (Hymenoptera: Ichneumonidae), a parasitoid of Diamondback Moth (Lepidoptera: Plutellidae). *Environmental Entomology*, 26: 114-120.

Jacob, H. S., Evans, E. W., 2004, Influence of different sugars on the longevity of *Bathyplectes curculionis* (Hym., Ichneumonidae). *Journal of Applied Entomology*, 128:316-320.

Jing, S. Hu., Gelman, D. B., Bell, R. A., 2001, In vitro rearing of *Edovum puttleri* an egg parasitoid of the Colorado potato beetle from egg to pupal stage in artificial diets devoid of insect sources: effects of dietary amino acids and carbohydrate levels. *BioControl,* 46: 43-60.

Magro, S. R., Dias, A. B., Terra, W. R., Parra, J. R., 2006, Biological, nutritional, and histochemical basis for improving an artificial diet for *Bracon hebetor* Say (Hymenoptera:Bracanidae). *Neotropical Entomology*, 35: 215-222.

Mehmetoğlu, R., Başhan, M., 1996, The carbohydrate requirements of *Melanogryllus desertus* Pall. (Orthoptera: Gryllidae). *Turkish Journal of Biology*, 23: 91-100.

Mittler, T. E., Dadd, R. H., Daniels, S. C. Jr., 1970, Utilization of different sugars by the aphid *Myzus persicae*. *Journal of Insect Physiology*, 16: 1873-1890.

Murugan, K., George, A., 1992, Feeding and nutritional influence on growth and reproduction of *Daphnis nerii* (Linn.) (Lepidoptera: Sphingidae). *Journal of Insect Physiology*, 18: 961-967.

Nettles, W. C. Jr., 1972, Toxic sugars and longevity of *Anthonomus grandis*. *Journal of Insect Physiology*, 18: 1107-1113.

Onagbola, E. O., Fadamiro, H. Y., Mbata, G. N., 2007, Longevity, fecundity, and progeny sex ratio of *Pteromalus cerealellae* in relation to diet, host provision, and mating. *Biological Control*, 40: 222-229.

Özalp, P., Emre, I., 2001, The effect of carbohydrates upon the survival and reproduction of adult female *Pimpla turionellae* L (Hym.,Ichneumonidae). *Journal of Applied Entomology.* 125: 177-180.

Sokal, R. R., Rohlf, F. J., 1969, *Biometry*. W.H. Freeman and Company, San Francisco. 776 pp.

Tsiropoulos, G. J., 1980, Carbohydrate utilizatıon by normal and y-Sterilized *Dacus olea*. *Journal of Insect Physiology*, 26: 633-637.

Wäckers, F. L., 1999, Gustatory response by the hymenopteran parasitoid *Cotesia glomerata* to a range of nectar and honeydew sugars. *Journal Chemical Ecology*, 25: 2863-2877.

Wäckers, F. L., 2001, A comparison of nectar- and honeydew sugars with respect to their utilization by the hymenopteran parasitoid *Cotesia glomerata*. *Journal of Insect Physiology*, 47: 1077-1084.

Zucoloto, F. S., 1992, Egg production by *Ceratitis capitata* (Diptera,Tephritidae) fed with different carbohydrates. *Revista Brasileira de Entomologia*, 36: 235-240.

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