J. Entomol. Res. Soc., 19(3): 109-118, 2017 *ISSN:1302-0250*

Fatty Acid Composition of *Diplolepis fructuum* (Rübsaamen, 1895) (Hymenoptera: Cynipidae) During Its Developmental Stages

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ABSTRACT

In this study, fatty acid composition in lipid classes of *Diplolepis fructuum* (Rübsaamen, 1895) (Hymenoptera: Cynipidae), an insect species inducing gall formation on the rose hips (the genus *Rosa*), during its various developmental stages was analyzed. According to the results obtained, C 16:0, C 18:0, C 18:1 n-9, C 18:2 n-6 and C 18:3 n-3 were the most abundant fatty acids in lipid classes during developmental stages. In *D. fructuum*, the presence of C 20 polyunsaturated fatty acids could not be determined. The most important finding for the fatty acid analyses of the galls and the healthy fruits of the rose hips was that polyunsaturated fatty acid (PUFA) levels were higher in the healthy fruits.

Key words: Gall-inducing insect, *Diplolepis fructuum*, lipid classes, fatty acids, development.

INTRODUCTION

Insects, with perhaps over a million species, constitute the most diverse and crowded animal life form on the earth. Insects are useful tools for investigating biochemical pathways because they both share common metabolic processes with the vertebrate systems and are much simpler systems than vertebrates. For these reasons, lipid metabolism in insects is under a great investigation (Canavaso *et al.*, 2001).

Lipids are important molecules in all insect species during their life stages to organize their physiological processes such as growth, metamorphosis, energy production, reproduction, and flight (Beenakkers *et al.*, 1985; Ryan and Van der Horst, 2000). Fatty acids also take over crucial roles for insects such as biosynthesis of eicosanoids, waxes, pheromones, and various defensive secretions (Stanley Samuelson *et al.*, 1988; Başhan, 1996; Büyükgüzel *et al*., 2007; Büyükgüzel *et al.*, 2011). The roles of prostaglandins, which are synthesized from polyunsaturated fatty acids (PUFAs) (Büyükgüzel *et al.*, 2002; Tunaz *et al.*, 2003), in egg-lying behaviors of insects are noteworthy (Stanley Samuelson and Loher, 1986). Phospholipids constitute the roof of the biological membranes, with the incorporation of the PUFAs to the structure of cellular membranes (Tocher *et al.*, 2008). Fatty acid composition of

the biological membranes is dynamic and may change depending on the environmental factors and physiological state of the insect. This phenomenon is called homeoviscous adaptation (Çakmak, 2010). Triglycerides function as energy storage molecules due to their high energy capacities. At the same time, they are important for the formation of metabolic water, which is crucial for the terrestrial organisms, including insects (Kinsella, 1966).

Recently, important studies have been carried out on the gall making insects in the context of plant - insect interactions (Raman, 2011). One of these insect species is *Diplolepis fructuum* from Cynipidae family having approximately 1,400 species, which is one of the largest families of Cynipoidea (Ronquist, 1999; Katılmış and Kıyak, 2009). *D. fructuum* can cause extensive damage on the fruits of wild rose species, especially *Rosa canina*, resulting in the formation of plant galls, which can be defined as the host for the insect species (Lotfalizadeh *et al.*, 2009) and are abnormal growths in plant tissue (Raman, 2011). The genus Rosa has some important economic values. It is used as medicinal remedy and food because the fruits of the *Rosaceae* family's species have rich content in both vitamin C and the phenolic compounds, which are responsible for the physiological and biochemical functions, such as antimutagenic, antioxidant qualities (Ercisli, 2007; Lattanzio *et al.*, 2011). For these reasons, the present study has been focused on the determination of the fatty acid composition of *D. fructuum* during its developmental stages. Furthermore, the fatty acid compositions of the healthy fruits and the galls of *Rosa canina*, which is a given host for the insect species, were investigated.

MATERIALS AND METHODS

Insects

Individuals of *Diplolepis fructuum* used in the experiments were collected from the galls on *Rosa canina*, which is distributed in the different localities in city Sivas (Turkey). Field studies were carried out between November 2012 and November 2013. *R. canina* galls were collected from different localities of Sivas, including Gürün, Şarkışla, Gemerek, Kangal, Divriği, Zara, Suşehri, and Koyulhisar. The gall samples carried to the laboratory were divided two parts. The first part of the galls was used to provide the adult emergence by incubating the galls at room temperature in the bottle glasses. The second part of the gall samples was used to obtain the eggs, larvae, and pupae of *D. fructuum* according to the successive formation sequence of the embryological growth stages. With these processes, the eggs, larvae, pupae, and adult individuals of *D. fructuum* were obtained and used as experimental material. However, one limitation we faced was that we were not able to obtain sufficient amounts of the egg samples to perform the fatty acid analyses of the neutral and polar lipid studies of the egg samples of *D. fructuum*. We also collected the gall samples and the healthy fruits of *R. canina* to carry out fatty acid analyses during the field studies.

Lipid extraction and lipid class purification

The method which was developed by Folch *et al.* (1957) was used to extract the total lipids from the experimental materials used in the present study. To obtain the

total lipids, three 0.3 g replicates of the egg samples, three 1 g replicates of the larvae, pupae, and adult insect samples, three 3 g replicates of the gall and healthy fruit samples of *R. canina* were used. All the samples were homogenized in chloroform / methanol (2 / 1, v / v) with Ultra - Turrax T 25 homogenizer. The total lipids obtained from the larvae, pupae, and adult stages were applied to a silica gel (Davisil® grade 633, pore size 60A°, 200 –425 mesh) column chromatography to purify neutral and polar lipid fractions. The experimental procedures for obtaining total lipids and lipid class purification were the same as a previous published article (Görgün *et al.*, 2013). The method of Moss *et al.* (1974) was utilized in the saponification of total lipids and obtaining the fatty acid methyl esters (FAMEs) from all experimental materials.

Gas chromatography

Gas chromatographic analyses of FAMEs were performed with an HP Agilent 6890N model gas chromatograph (Hewlett Packard, Palo Alto, CA, USA) equipped with an HP - 88 capillary column (0.2 µm thickness, 0.25 mm ID, and 100 m length) and a flame ionization detector. In the gas chromatography conditions and studies, the method of Guler *et al.* (2010) was used.

Statistical analysis

Statistical analyses of the study were carried out with SPSS 15.0 (SPSS Inc., Chicago, IL, USA). Statistical evaluation of the fatty acid percentages obtained from gas chromatography was carried out with the analysis of variance (ANOVA). Tukey's test was used to assess the differences the means at $P \leq 0.05$ levels. Student's *t*-test was used to evaluate the differences between the fatty acid compositions of the galls and healthy fruits of rose hips.

RESULTS

The meteorological data which show the highest and lowest temperature (°C) and humidity (%) values during the collection of the experimental materials were reported in Table 1. During the field studies, we observed that the gall formation on rose species was at the lowest levels in the north areas (especially Suşehri and Koyulhisar regions) of Sivas when compared to the other regions, especially the south regions of Sivas that cover Gürün, Şarkışla, and Gemerek.

Table 2, 3, 4 show the fatty acid composition data of the total, neutral, and polar lipid fractions of *D. fructuum*, respectively. At the same time, fatty acid composition data of the healthy fruit and the gall samples of *R. canina* on which *D. fructuum* used as a host can be seen in Table 5. In the total lipids, egg samples had fourteen different fatty acids while eighteen fatty acids were identified in the larvae, pupae, and adult insect samples. C 11:0, C 12:0, C 13:0, C 14:1 n-5, and C 18:3 n-6 were absent in the egg sample of *D. fructuum*. Interestingly, the same data was observed for the fatty acid composition of the gall and healthy fruit samples of the rose hips (*R. canina*). The presence of C 20:0 were not determined in the larvae, pupae, and adult insect samples. In the neural and polar lipids, eighteen fatty acids were determined

in the developmental stages of *D. fructuum*. We observed that there are important quantitative differences in the fatty acid composition of *D. fructuum*, depending on the development. All the details for the results can be seen from the tables.

Table 1. Average temperature (°C) and humidity percentages (%) in the sampling sites*

Sampling period and the year (between 02 - 10 November)	South region of Sivas (Minimum - Maximum)	North region of Sivas (Minimum - Maximum)
Humidity (2012 year)	56.3 – 86.3	45 – 96
Humidity (2013 year)	53.0 – 78.3	35 – 84
Temperature (2012 year)	8 – 12.4	6.9 – 15.8
Temperature (2013 year)	5.7 – 8.9	6.5 – 12.1

*The data presented in the table was obtained from Turkish State Meteorological Service.

Table 2. Fatty acid composition (%) of the total lipid of *D. fructuum* during its developmental stages♯

Fatty Acids	Egg Mean ± SE	Larvae Mean ± SE	Pupae Mean ± SE	Adult Mean ± SE
C 8:0	0.27 ± 0.05^{a}	0.07 ± 0.01^{b}	0.17 ± 0.02^{c}	0.38 ± 0.07^{d}
C 10:0	0.34 ± 0.17^{a}	0.08 ± 0.02^{b}	0.19 ± 0.04^{c}	0.27 ± 0.03^{ac}
C 11:0	---	0.09 ± 0.02^{a}	0.19 ± 0.02^{b}	0.41 ± 0.04^{c}
C 12:0	---	0.12 ± 0.01^{a}	0.11 ± 0.01^{a}	0.13 ± 0.02^{a}
C 13:0	---	0.01 ± 0.00^{a}	0.14 ± 0.01^{b}	0.01 ± 0.00^{a}
C 14:0	0.42 ± 0.09^{a}	0.30 ± 0.02^{b}	0.32 ± 0.03^{b}	0.38 ± 0.06^{b}
C 15:0	0.17 ± 0.02^{a}	0.02 ± 0.00^{b}	0.02 ± 0.01^{b}	0.04 ± 0.01^{b}
C 16:0	8.73 ± 0.69^{a}	5.70 ± 0.11^{b}	6.90 ± 0.16^{c}	8.33 ± 0.23^{a}
C 17:0	0.14 ± 0.03^{a}	0.03 ± 0.01^{b}	0.06 ± 0.02^{ab}	0.09 ± 0.03^{ab}
C 18:0	2.38 ± 0.40^{a}	2.55 ± 0.10^{b}	2.54 ± 0.06^{b}	3.38 ± 0.04^{c}
C 20:0	1.28 ± 0.19	---	---	---
ΣSFA	**13.71 ± 0.44^{a}**	**8.95 ± 0.14^{b}**	**10.61 ± 0.12^{c}**	**13.46 ± 0.18^{a}**
C 14:1 n-5	---	0.07 ± 0.03^{a}	0.03 ± 0.01^{a}	0.35 ± 0.02^{b}
C 15:1 n-5	0.06 ± 0.01^{a}	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}	0.02 ± 0.00^{a}
C 16:1 n-7	0.14 ± 0.02^{a}	0.30 ± 0.06^{b}	0.78 ± 0.09^{c}	0.44 ± 0.11^{d}
C 17:1 n-8	0.28 ± 0.11^{a}	0.02 ± 0.00^{b}	0.03 ± 0.01^{b}	0.05 ± 0.01^{b}
C 18:1 n-9	11.08 ± 0.47^{a}	49.54 ± 0.38^{b}	42.58 ± 0.27^{c}	40.72 ± 0.53^{d}
ΣMUFA	**12.13 ± 0.37^{a}**	**49.93 ± 0.14^{b}**	**43.43 ± 0.10^{c}**	**41.58 ± 0.20^{d}**
C 18:2 n-6	50.68 ± 1.34^{a}	13.61 ± 0.13^{b}	26.23 ± 0.21^{c}	27.96 ± 0.26^{d}
C 18:3 n-6	---	0.04 ± 0.00^{a}	0.07 ± 0.03^{a}	0.18 ± 0.01^{b}
C 18:3 n-3	23.49 ± 0.71^{a}	27.47 ± 0.22^{b}	19.68 ± 0.10^{c}	16.80 ± 0.18^{d}
ΣPUFA	**74.16 ± 0.84^{a}**	**41.12 ± 0.11^{b}**	**45.97 ± 0.07^{c}**	**44.95 ± 0.13^{d}**

♯Each data represent the mean of three experiments with Standard Error (SE). Means with the same letter in each row do not significantly differ at P ≤ 0.05. ΣSFA: Total Saturated Fatty Acids. ΣMUFA: Total Monounsaturated Fatty Acids. ΣPUFA: Total Polyunsaturated Fatty Acids.

Fatty Acid Composition of Diplolepis fructuum

Table 3. Fatty acid composition (%) of neutral lipid fraction of *D. fructuum* during its developmental stages♯

Fatty Acids	Larvae Mean ± SE	Pupae Mean ± SE	Adult Mean ± SE
C 8:0	0.02 ± 0.00^{a}	0.01 ± 0.00^{a}	0.14 ± 0.01^{b}
C 10:0	0.08 ± 0.01^{a}	0.12 ± 0.01^{a}	0.48 ± 0.17^{b}
C 11:0	0.08 ± 0.03^{a}	0.13 ± 0.04^{a}	0.54 ± 0.09^{b}
C 12:0	0.06 ± 0.01^{a}	0.05 ± 0.02^{a}	0.29 ± 0.03^{b}
C 13:0	0.09 ± 0.03^{a}	0.13 ± 0.00^{a}	0.61 ± 0.11^{b}
C 14:0	0.15 ± 0.02^{a}	0.19 ± 0.07^{a}	0.37 ± 0.06^{b}
C 15:0	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}	0.02 ± 0.00^{a}
C 16:0	4.25 ± 0.30^{a}	5.93 ± 0.28^{b}	3.17 ± 0.21^{c}
C 17:0	0.05 ± 0.01^{a}	0.05 ± 0.01^{a}	0.01 ± 0.00^{a}
C 18:0	2.49 ± 0.17^{a}	1.98 ± 0.34^{b}	1.44 ± 0.20^{c}
ΣSFA	**7.27 ± 0.11^{a}**	**8.60 ± 0.20^{b}**	**7.05 ± 0.16^{a}**
C14:1 n-5	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}	0.13 ± 0.01^{b}
C15:1 n-5	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}	0.06 ± 0.02^{a}
C16:1 n-7	0.07 ± 0.01^{a}	0.44 ± 0.06^{b}	0.56 ± 0.18^{b}
C17:1 n-8	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}	0.06 ± 0.00^{a}
C18:1 n-9	58.60 ± 0.73^{a}	49.38 ± 0.69^{b}	36.21 ± 0.33^{c}
ΣMUFA	**58.70 ± 0.42^{a}**	**49.84 ± 0.37^{b}**	**37.01 ± 0.22^{c}**
C 18:2 n-6	14.17 ± 0.25^{a}	27.03 ± 0.19^{b}	32.18 ± 0.50^{c}
C 18:3 n-6	0.10 ± 0.01^{a}	0.03 ± 0.01^{a}	0.08 ± 0.01^{a}
C 18:3 n-3	19.77 ± 0.41^{a}	14.52 ± 0.24^{b}	23.70 ± 0.19^{c}
ΣPUFA	**34.03 ± 0.29^{a}**	**41.57 ± 0.17^{b}**	**55.95 ± 0.37^{c}**

♯See Table 2 for the explanations.

Table 4. Fatty acid composition (%) of polar lipid fraction of *D. fructuum* during its developmental stages♯

Fatty Acids	Larvae Mean ± SE	Pupae Mean ± SE	Adult Mean ± SE
C 8:0	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}
C 10:0	0.01 ± 0.00^{a}	0.12 ± 0.02^{b}	0.14 ± 0.03^{b}
C 11:0	0.04 ± 0.01^{a}	0.13 ± 0.01^{ab}	0.17 ± 0.02^{b}
C 12:0	0.14 ± 0.01^{a}	0.17 ± 0.01^{a}	0.12 ± 0.02^{a}
C 13:0	0.04 ± 0.01^{a}	0.10 ± 0.00^{a}	0.14 ± 0.02^{a}
C 14:0	0.39 ± 0.08^{a}	0.50 ± 0.07^{a}	0.47 ± 0.11^{a}
C 15:0	0.01 ± 0.00^{a}	0.02 ± 0.00^{a}	0.06 ± 0.01^{a}
C 16:0	6.25 ± 0.77^{a}	7.79 ± 0.80^{b}	8.77 ± 0.68^{c}
C 17:0	0.06 ± 0.01^{a}	0.08 ± 0.01^{a}	0.10 ± 0.02^{a}
C 18:0	2.13 ± 0.19^{a}	3.08 ± 0.18^{b}	3.51 ± 0.29^{c}

Table 4. Continued.

Fatty Acids	Larvae Mean ± SE	Pupae Mean ± SE	Adult Mean ± SE
ΣSFA	**9.07 ± 0.46^{a}**	**12.00 ± 0.44^{b}**	**13.47 ± 0.33^{c}**
C14:1 n-5	0.11 ± 0.02^{a}	0.08 ± 0.01^{a}	0.07 ± 0.01^{a}
C15:1 n-5	0.01 ± 0.00^{a}	0.02 ± 0.00^{a}	0.01 ± 0.00^{a}
C16:1 n-7	0.08 ± 0.02^{a}	0.40 ± 0.10^{b}	0.93 ± 0.14^{c}
C17:1 n-8	0.03 ± 0.00^{a}	0.06 ± 0.01^{a}	0.04 ± 0.01^{a}
C18:1 n-9	46.89 ± 0.88^{a}	36.63 ± 0.95^{b}	41.81 ± 1.44^{c}
ΣMUFA	**47.10 ± 0.80^{a}**	**37.19 ± 0.52^{b}**	**42.85 ± 0.60^{c}**
C 18:2 n-6	13.57 ± 0.70^{a}	26.27 ± 0.94^{b}	27.43 ± 0.50^{c}
C 18:3 n-6	0.08 ± 0.01^{a}	0.07 ± 0.01^{a}	0.13 ± 0.02^{a}
C 18:3 n-3	30.19 ± 0.61^{a}	24.47 ± 0.41^{b}	16.14 ± 0.56^{c}
ΣPUFA	**43.84 ± 0.57^{a}**	**50.81 ± 0.77^{b}**	**43.69 ± 0.37^{a}**

♯See Table 2 for the explanations

Table 5. Fatty acid composition (%) of the gall and the healthy fruit samples of the rose hips (*Rosa canina*) ♯

Fatty Acids	Gall Mean ± SE	Healthy fruit Mean ± SE
C 8:0	0.41 ± 0.08^{a}	0.07 ± 0.01^{b}
C 10:0	0.57 ± 0.15^{a}	0.09 ± 0.03^{b}
C 11:0	---	---
C 12:0	---	---
C 13:0	---	---
C 14:0	0.36 ± 0.03^{a}	0.10 ± 0.01^{b}
C 15:0	0.27 ± 0.04^{a}	0.06 ± 0.01^{b}
C 16:0	6.34 ± 0.62^{a}	4.23 ± 0.30^{b}
C 17:0	0.48 ± 0.07^{a}	0.10 ± 0.03^{b}
C 18:0	4.00 ± 0.31^{a}	2.99 ± 0.49^{b}
C 20:0	1.27 ± 0.12^{a}	1.20 ± 0.18^{a}
ΣSFA	**13.69 ± 0.44^{a}**	**8.83 ± 0.28^{b}**
C 14:1 n-5	---	---
C 15:1 n-5	0.07 ± 0.02^{a}	0.01 ± 0.00^{a}
C 16:1 n-7	0.14 ± 0.01^{a}	0.10 ± 0.01^{a}
C 17:1 n-8	0.13 ± 0.03^{a}	0.04 ± 0.01^{a}
C 18:1 n-9	22.20 ± 0.69^{a}	20.93 ± 0.86^{b}
C 20:1 n-9	0.40 ± 0.08^{a}	0.34 ± 0.06^{a}

Table 5. Continued.

Fatty Acids	Gall Mean ± SE	Healthy fruit Mean ± SE
ΣMUFA	**22.92 ± 0.34**[a]	**21.41 ± 0.43**[b]
C 18:2 n-6	46.55 ± 1.39[a]	50.32 ± 0.93[a]
C 18:3 n-6	---	---
C 18:3 n-3	16.84 ± 0.56[a]	19.45 ± 0.77[b]
ΣPUFA	**63.39 ± 0.91**[a]	**69.77 ± 0.82**[b]

♯See Table 2 for the explanations

CONCLUSIONS AND DISCUSSION

The meteorological data is a useful and important factor as the signals of physiological events in the livings. Regarding with the differences in the gall formation in the different localities, we thought that the complex interaction of the ecological events such as temperature and humidity might be an important factor both in the gall formation and in the insect invasion on the rose species which cause gall formation.

The major fatty acids in all lipid fractions during the growth stages of *D. fructuum* were C 16:0 (palmitic acid), C 18:0 (stearic acid), C 18:1 n-9 (oleic acid), C 18:2 n-6 (linoleic acid), and C 18:3 n-3 (linolenic acid). Previous published studies on the insect fatty acid composition confirmed the similar data. For example, in *Periplaneta americana*, approximately 95% of total fatty acids consisted of C 16:0, C 18:0, C 18:1 and C 18:2 (Kinsella, 1966). Another study focused on the fatty acid dynamics of *Gryllus campestris* during its various growth stages determined that the predominant fatty acids were C 16:0, C 18:0, C 18:1, C 18:2 and C 18:3 (Akpınar *et al.*, 2003).

Polyunsaturated fatty acids (PUFAs) were the main components in the total lipid (TL) of the egg samples, representing 74.16% of the total fatty acids due to the higher percentages of C 18:2 n-6 (50.68%) and C 18:3 n-3 (23.49%). During the developmental stages of *D. fructuum*, the highest percentage of the total saturated fatty acid fraction (ΣSFA) in total lipid was determined in the egg samples (13.71%) and adult stage (13.46%) ($P \geq 0.05$). In the neutral lipid fractions (NL) and polar lipid fractions (PL), the highest ΣSFA percentages were observed to be in the pupae (8.60%) (Table 3) and adult stages (13.47%), respectively. From these findings, generally fatty acids of TL, NL, and PL fractions during the growth stages of *D. fructuum* composed of monounsaturated fatty acids (MUFAs) and PUFAs fractions.

In the NL fraction (Table 3), C 18:1 n-9, showing a range between 58.60% (larvae stage) and 36.21% (adult stage), were the principal fatty acid within all fatty acids and different growth stages of *D. fructuum*. Other fatty acid members in the MUFAs fraction were the minor components during all of the growth stages. ΣMUFA percentages in the larvae (58.70%), pupae (49.84%), and adult stages (37.01%) of *D. fructuum* showed regular decreases in a stepwise manner. PUFAs percentages at different growth stages also showed in stepwise increases in the larvae (34.03%), pupae (41.57%), and adult stages (55.95%).

We observed similar fatty acid patterns in the PL fraction during different growth stages of *D. fructuum*. For example, C 18:1 n-9 was the predominant fatty acid within all fatty acids and growth stages (Table 4). However, fluctuations seen in this fatty acid in the PL fraction were not at a regular manner when compared to the NL fraction. At the same time, PUFAs plus MUFAs fractions were the predominant fatty acid classes throughout the growth stages. The highest C 18:1 n-9 percentage was found to be in the larvae stage (46.89%). ΣPUFA level in which the highest percentage was observed in the pupae stage (50.81%) also showed fluctuations between different growth stages.

The most abundant fatty acids of the gall and the healthy fruit samples of *Rosa canina* which are the host for the eggs of *D. fructuum* were C16:0, C18:0, C18:1 n-9, C18:2 n-6, C18:3 n-3 (Table 5). ΣSFA and ΣMUFA percentages were found to be higher in the gall samples when compared to the healthy fruit samples of *R. canina*. However, PUFAs percentage in the healthy fruits (69.77%) was higher in the gall samples (63.39%). Ercisli (2007) conducted a comprehensive study on the rose species and determined nine major fatty acid compounds in which the major fatty acid was C18:3 n-3. When compared to this study, we defined fifteen fatty acids. At the same time, C 18:2 n-6 was the dominant fatty acid both in the gall samples (46.55%) and in the healthy fruits (50.32%). With these findings, the present study has been suggested that *R. canina* samples appear to be rich in terms of n-6 PUFAs. Nowak (2005) investigated the fatty acid composition of the fruits of eleven wild rose species (including *R. canina* as well) from Poland and found to be that linoleic acid (44.4 %-55.7 %), linolenic acid (18.6 %-31.4 %), and oleic acid (13.5 %-20.3%) were the main compounds. Similar results with quantitative differences were published by Kazaz *et al.* (2009) in *R. canina* and *R. damascene* fruits from Turkey.

We were not able to identify C 20 long chain PUFAs both in the growth stages of the insect investigated and in the *R. canina* samples that include both the healthy fruit and the gall samples. One important generalization of the insect fatty acid pattern is that terrestrial insect species might have fewer amounts in C 20 PUFAs while aquatic ones might have high amount in these longer chain fatty acids (Hanson *et al.*, 1985). However, a comprehensive study which focused on the fatty acid dynamics of the lipid classes of five phytophagous insect species from chrysomelids showed that these insects did not accumulate C 20 PUFAs due to the deficiency of C 20 long chain fatty acids in their diets because the plant which is their diet was not able to synthesize C 20 PUFAs beyond C 18 PUFAs (Ogg *et al.*, 1993). In the present study, we also have showed that the host plant *R. canina* is devoid of C 20 long chain PUFAs. From this point of view, we suggest that *D. fructuum* which shows a phytophagous character might display similarities that of the fatty acid patterns of the species investigated in the study of Ogg *et al.* (1993) in terms of C 20 long chain PUFAs.

Finally, in this study, we carried out a detailed fatty acid composition study for both *D. fructuum* and *R. canina* which are found in Turkey. These results might be important in insect-plant interactions in terms of fatty acid composition data.

ACKNOWLEDGEMENTS

This research project was financially supported by the Scientific Research Found of Cumhuriyet University (CUBAP) with the project number F - 378. For this reason, the authors convey their special thanks to CUBAP.

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Received: June 02, 2017 *Accepted: October 12, 2017*