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Effect of Fungal Infection on Fatty Acid Contents of the Stored Green Coffee Beans

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ABSTRACT

Coffee beans exposed to infection by microorganisms during storage, especially fungal infection which cause great damage to beans. This research aimed at studying the bad effect of fungal infection of stored coffee beans at the eastern zone of Saudi Arabia. Result showed that coffee beans contain number of fatty acids, namely caprilic, lauric, miristic, palmitic, palmitoleic, margaric, stearic, Oleic, linoleic, alpha-linoleic and arachidic acids. Treatment of beans with fungi led to increase in the percentage of miristic, margaric and stearic acids. With increasing the duration period of storage% of oleic acid of Harari verity showed its level before treatment at 0, 8, 25°C to be 42.28, 40.04 and 42.97%, respectively . These levels showed decreasing after storage for three months to be 37.24, 36.59 and 38.98% at the same degrees with increasing the period of storage. In the Barry, the level of the oleic acid recorded 42.36 and 41.77% at 8 and 25°C. These records decreased after storage to be 38.71%, 38.65% in the infected beans. These levels changed during storage at 0°C from 35.87% in the control, to 39.1% in the infected ones. Storage for 9 months at 25°C decreased the acid level to 25, 19.93% in both control and treated beans storage at 0, 8°C led to increase in acid level from 19.81, 19.36%, in control beans, to be 27.84, 19.74% in infected beans, respectively.

Key words: Coffee beans, fatty acid, margaric, stearic, oleic, miristic, fungal infected

INTRODUCTION

Coffee was known during the 9th century BC when it was used in Ethiopia as nuts from Ethiopia at spread to Egypt and Aden. At the 15th century, it reached to Azerbaijan, turkey and Africa and from Islamic countries to Italy, Europe and America (Mekete, 2003). There are special criteria concerning the quality of coffee beans as the type of beans, environmental conditions during growing of the plant, preparation of the seeds and storage conditions. The method of bulb extraction is the main target in seed preparation: fermentation and removal of testa and its inner membrane are processes involved. Fermentation of coffee beans is by spreading beans in layers of 10 cm thick and left for 10-25 days exposed to sun for drying. A bout 754 microbial isolates are involved in fermentation through lysis of cellulose and pectin; these types of fermentations interfere with the quality and last of coffee (Silv *et al*., 2000). Ethiopia is considered one of the most important countries which exporting coffee. Betancourt and Frank (1983) assured that humidity should be less than 14.5% to prevent molding during storage. Ochratoxin was showed during storage with high relative humidity during 20-30 storage days. Humidity accompanied with

variation in physiological properties of fungi. High humidity and bad storage led to production of ochratoxin within 10 days (Ahmad and Magan, 2006). Shikimic acid was isolated and purified from *Aspergillus niger* by Tenkanen *et al.* (1991), Faulds and Williamson (1994), Kroon *et al.* (1996) and Christov and Prior (1993) mentioned that bacteria and fungi produced many enzymes as Hemocellulase, shikimic esterase to lyses cell wall and break down methyl ester bonds of the feriolic and kumaric acids which enzymes from hemocellulose increased during hydrolysis of cinnamic acid (Kroon and Williamson, 1996). Some researchers cleared that the structure and percentage of fatty acids deepened on the rate of lysis during storage. Mabbett (2002) mentioned that storage of coffee at high humidity raise the rate of fungal infection leading to increase in % of free Fatty Acids (FA) this phenomenon led to problems during processing. Morello *et al.* (2004) realized decrease in level of linoleic and linolinic acids and increase in oleic acid in stored olive oil. Fatty acids is the more important factor during storage which changed by oxidation, type of fatty acids and its dibonds restrict the type of chemical reactions happened during storage. Sisman and Delibas (2004) and Sisman (2005) proved that period of storage; humidity and temperature are the factors affecting the quality of the product. The level of oil in coli flower decreased after storage for 3 months; increase in temperature and humidity led to spoiling oil and increasing free fatty acids. The same results were recorded by Villiers *et al.* (1986), Martini and Anon (2005) and Neg and Anderson (2005) assured the same results on the kinoa plant. Vila *et al.* (2005) compared Between the level of fatty acids in two coffee samples, the first is pure Arabica processed coffee beans and the second was 80% Arabica +20% Robusta, both types were packaged under vacuum and stored at 25oc for 180 days. They recorded increase in fatty acid levels in the second sample only. Ghasemnez had and Honermeier (2007) realized decrease in oleic acid level after storage at 4 months from 90.6-88.2%, he explained that by the ability of this acid to react and change to free forms. He also recorded the same effect on palmalic and Linolic acids. The same results were recorded by Ghasemnez had and Honermeier (2007) when tested the effect of storage period on level of fatty acids in the spring flower. They explained the lower level of fatty acids to the activity of enzymes like lipase. Our research aimed at studying the effect of storage period, temperature and fungal infection on levels of fatty acids of coffee beans.

MATERIALS AND METHODS

Studies on fungal infected coffee beans: Coffee beans of 2 varieties were used (Harari and Barry) in our experiments, healthy, uninfected seed was chooses and sterilized using Arnold sterilizer. Relative humidity was estimated.

Seed were put in plastic bags (250 g beans in each) with% humidity 10%; each group is divided to 4 sub groups for treatment as follows:

- Control experiment without fungal infection
- Contamination of the 3 groups with *A. niger*, *A. melleus*, *A. alliacus*, each experiment is repeated 3 times and incubated at (0, 8, 25±2°C) with storage periods 3 and 9 months

Fatty acid analysis: Fatty acids were determined in the hexane extract of coffee beans by Gas-liquid Chromatographic technique (GLC) by Brockerhoff (1965) as follows:

Extraction of fatty acids: The coffee beans oil was extracted using hexane. The solvent was evaporated under reduced pressure and the residue was saponified by heating in a boiling water bath for 30 min in presence of alcoholic sodium hydroxide solution (15% w/v). After cooling to room

temperature, the solution was transferred into separatory funnel, then 20 mL. of petroleum ether (60-80°C) were added and the mixture was vigorously shaken. The aqueous layer was separated and acidified with HCl (1:1, v/v). The liberated fatty acids were extracted three times with petroleum ether (60-80°C) using 10 mL portion for each time and washed several ether (60-80°C) using 10 mL portion for each time and washed several times with distilled water until the washings were neutral to phenol phthalein.

Methylation of fatty acids: The combined petroleum ether extracts were dried over anhydrous sodium sulphate and the solvent was evaporated under reduced pressure. The fatty acids (1-2 mg) were dissolved in anhydrous diethyl, ether (0.5:1 mL) and methylated by drop wise addition of diazomethane solution until the yellow color persisted and the bubbles of nitrogen gas ceased. The mixture was then left at room temperature for 15 min and the solvent was removed by evaporation. Finally, the methyl esters of fatty acids were dissolved in chloroform and aliquots of this solution were subjected to gas-liquid chromatographic analysis for identification of the methylated fatty acids.

Separation of the methyl esters of fatty acids by gas-liquid chromatography: A GCV pye Unicam gas-liquid chromatography apparatus equipped with a flame ionization detector and a coiled glass column (1.5×4 mm) packed with 10% PEGA supported on acid washed diatomic C (100-120 mesh) was used. The sample was injected into the column using Hamilton micro syringe. The gas chromatographic conditions used for isothermal work: were temperatures column 190°C, detector 300°C and injection 250°C. Flow rates: hydrogen 33 mL min^{-1}, nitrogen 30 mL min^{-1} and air 330 mL min^{-1}. Range 32×10-2 and chart speed 1 cm/2 min.

Identification and determination of the fatty acids: A set of standard methyl esters of 10:0, 11:0, 12:0, 13:0, 14:0, 16:0, 18:0, 18:1, 18:2, 18:3 and 20:0 fatty acids were used as authentic materials to enable the characterization of the unknown fatty acids by reference to their retention times. Peaks identification and quantification were performed by using Philips PU 4810 computing integrator.

RESULTS

Effect of fungal infection and storage on fatty acids of coffee beans all tested beans (control and treated) proved to have groups of saturated and unsaturated fatty acids. Oleic, Miristic and margaric, stearic acids recorded high frequency the other types had no significant percentages (3%) as Fig. 1. Fatty acids proved to be affected by storage period or fungal infection. Variation in temperature also led to change in acid percentages.

Results in Table 1 proved increase in meristic acid level which recorded 36.1-38.9%, 36.29% after stored for 3 months at 25, 8, 0°C for harari beans as control experiments. After infection under the same conditions% of the acid were 38.9, 43.82 and 39.28%, respectively. Fatty acids of margaric acid in control experiments recorded 6.74, 7.58 and 6.9% at the same conditions of storage and temperature.

These values increased to be 7.49, 8.31 and 7.81% at the same conditions after fungal infection. Seatric acid also recorded increase in level of fatty acids after infection storage at the same period and varieties in temperatures while control experiment recorded levels of 9.29, 9.96 and 9.3% under the same conditions. In contract, oleic acid recorded decrease in levels of its fatty acids to be after infection under the same compared with the control to have 42.97, 40.04 and 42.28%.

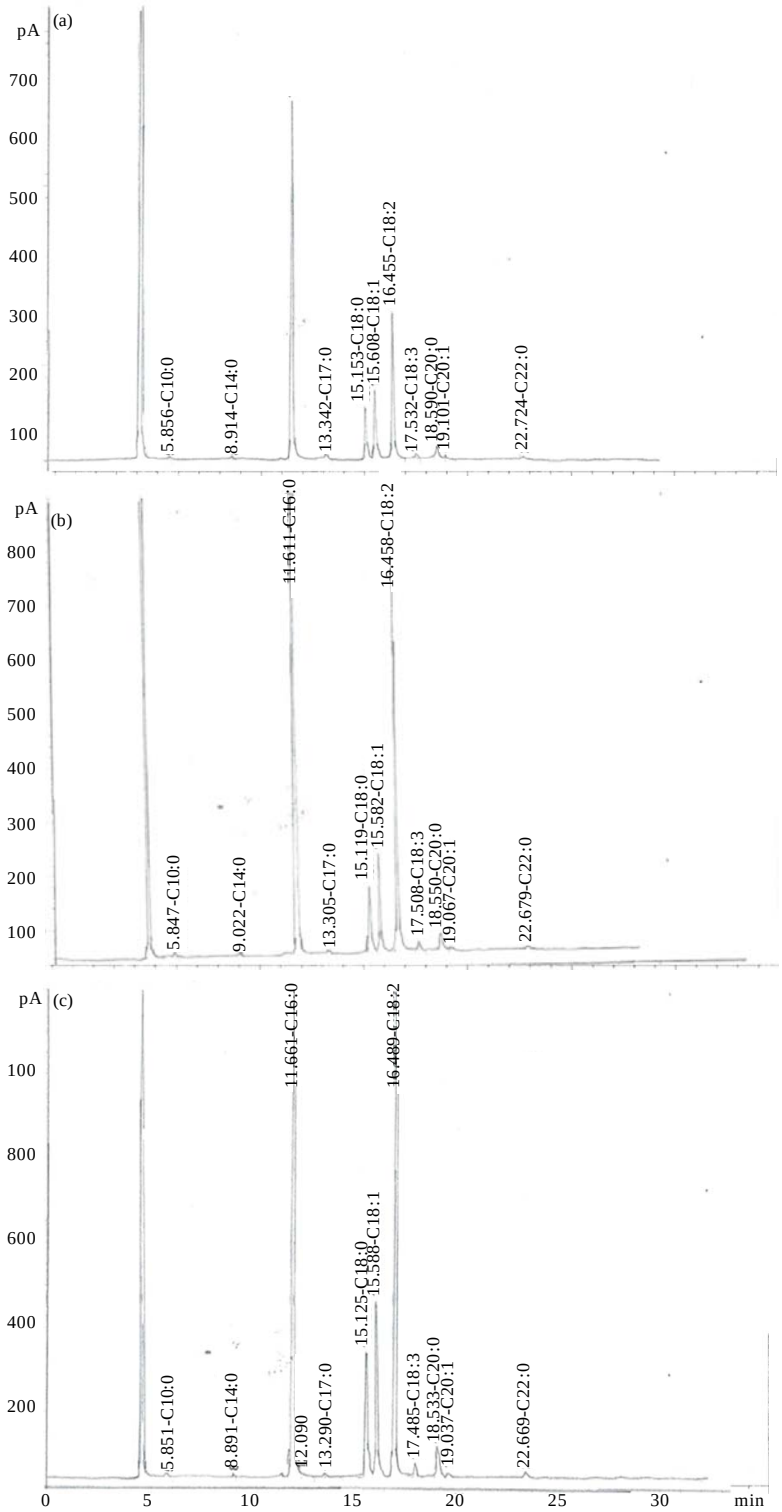

Fig. 1(a-d): Continue

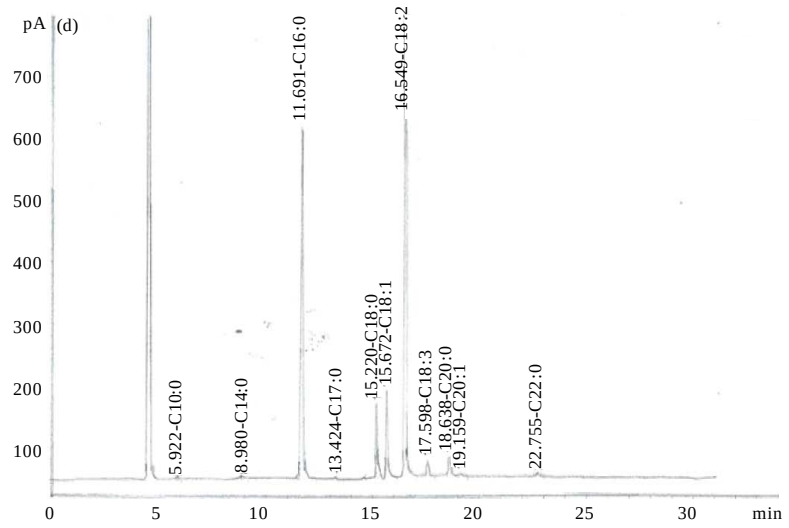

Fig. 1(a-d): Chromatogram of the fatty acids in Arabica coffee beans sample, (a) Coffee beans (Harari variety) infected with *A. niger*, (b) Coffee beans (Harari variety) infected with *A. melleus*, (c) Coffee beans (Barry variety) infected with *A. alliaceus*, (d) Coffee beans (Barry variety) infected with *A. niger,* was stored at a temperature of 25° C for a period of 9 months. (Caprilic acid = C 10: 0; Lauric acid = C 12: 0, Miristic acid = C14: 0, Palmitic acid = C 16: 0, Palmitoleic acid = C 16: 1, Margaric acid = C 17:0, Stearic acid = C 18: 0, Oleic acid = C 18: 1, Linoleic acid = C 18: 2, Alpha-Linoleic acid = C 18: 3, Arachidic acid = C 20: 0)

Table 1: Effect of artificial inoculation with tested fungi on fatty acids of stored Harari variety of coffee beans after 3 months

	Control			*A. niger*			*A. alliaceus*			*A. melleus*			Average of fatty acids		
Fatty acids	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°
Caprilic acid	0.02	0.030	0.07	0.50	0.040	0.01	0.04	0.15	0.03	0.07	0.04	0.02	0.05	0.07	0.02
Lauric acid	0.08	0.090	0.10	0.10	0.090	0.09	0.09	0.14	0.07	0.11	0.10	0.03	0.09	0.10	0.06
Miristic acid	36.13	39.500	36.29	40.52	38.660	43.4	36.46	56.34	36.77	39.33	36.47	37.67	38.90	43.82	39.28
Palmitic acid	0.21	0.050	0.08	0.18	0.230	-	0.11	-	0.06	0.09	0.06	-	0.12	0.13	0.02
Palmitoleic acid	0.09	0.100	0.10	0.10	0.100	0.10	0.10	0.14	0.10	0.10	0.10	0.09	0.10	0.11	0.09
Margaric acid	6.74	7.580	6.90	7.78	7.480	8.27	7.24	10.41	7.23	7.46	7.05	7.93	7.49	8.31	7.81
Stearic acid	9.29	9.960	9.30	10.33	9.710	10.10	9.81	12.2	10.09	9.80	9.95	10.51	9.47	10.62	10.38
Oleic acid	42.97	40.040	42.28	36.34	39.130	32.93	41.6	15.63	41.21	35.98	41.53	39.05	38.97	36.59	37.23
Linoleic acid	1.41	1.200	1.35	0.95	1.090	0.87	1.29	0.18	1.30	1.05	1.34	1.05	1.10	0.87	1.07
Alpha-Linoleic acid	2.21	2.530	2.55	2.57	2.500	2.7	2.32	3.41	2.39	2.21	2.36	2.64	2.48	2.75	2.58
Arachidic acid	0.35	0.360	0.48	0.41	0.370	0.41	0.44	0.41	0.36	0.36	0.37	0.40	0.40	0.38	0.38
Eicosenoic acid	0.45	0.570	0.54	0.59	0.550	0.62	0.48	0.73	0.36	0.57	0.53	0.58	0.54	0.60	0.51
LSD	8.65	0.336	0.758	0.257	0.333	0.248	3.53	1.50	7.34	0.431	3.77	0.136			

Table 2 showed results of measuring levels of fatty acids using (Harari or Barry) which stored for 9 months. Results showed that infection led to increase in% of Fatty Acids (FA). Miristic FA level changed from 55.26, 59.12 and 51.05% after storage at 25, 8 and 0°C to be after infection 56.22, 53.25 and 51.69%, respectively.

Table 2: Effect of artificial inoculation with tested fungi on fatty acids of stored Harari variety of coffee beans after 9 months

	Control			*A. niger*			*A. alliaceus*			*A. melleus*			Average of fatty acids		
Fatty acids	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°
Caprilic acid	0.09	0.07	0.04	0.07	0.07	0.07	0.09	0.02	0.05	0.08	0.05	0.06	0.07	0.04	0.06
Lauric acid	0.18	0.09	0.11	0.06	0.08	0.09	0.06	0.08	0.08	0.06	0.11	0.06	0.06	0.08	0.07
Miristic acid	55.26	59.12	51.05	58.61	55.97	49.53	63.18	49.68	53.90	52.88	54.11	51.64	56.22	53.25	51.69
Palmitic acid	0.17	-	0.10	0.21	0.13	0.31	0.15	0.08	0.41	0.15	0.19	0.11	0.17	0.13	0.27
Palmitoleic acid	0.07	0.14	0.13	0.13	0.16	0.08	0.08	0.12	0.09	0.13	0.13	0.13	0.11	0.14	0.13
Margaric acid	10.46	10.87	9.97	10.23	10.94	9.44	12.32	9.69	10.27	9.97	10.60	10.21	10.78	10.40	9.97
Stearic acid	11.78	10.59	11.73	11.76	11.86	11.54	11.50	12.50	11.61	11.65	12.80	12.32	11.63	12.38	11.82
Oleic acid	16.76	32.29	21.61	19.77	15.39	23.70	7.21	22.64	19.15	20.27	16.86	20.4	15.75	18.29	21.08
Linoleic acid	0.35	0.34	0.35	0.33	0.20	0.42	0.06	0.50	0.30	0.23	0.21	0.26	0.2	0.30	0.32
Alpha-Linoleic acid	3.58	3.67	3.47	3.08	3.79	3.32	4.21	3.36	3.03	3.38	3.58	2.37	3.55	3.57	2.90
Arachidic acid	0.36	0.44	0.48	0.50	0.47	0.46	0.61	0.46	0.49	0.49	0.48	1.31	0.53	0.47	0.75
Eicosenoic acid	0.9	0.75	0.93	0.65	0.91	0.96	0.52	0.83	0.54	0.88	0.83	0.65	0.68	0.85	0.71
LSD	3.6	7.50	4.30	5.70	5.20	2.60	5.90	6.60	2.93	4.50	2.30	1.80			

Table 3: Effect of artificial inoculation with tested fungi on fatty acids of stored Barry variety of coffee beans after 3 months

	Control			*A. niger*			*A. alliaceus*			*A. melleus*			Average of fatty acids		
Fatty acids	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°
Caprilic acid	0.05	0.02	0.03	0.05	0.040	0.03	0.02	0.02	0.03	0.05	0.04	0.04	0.04	0.03	0.03
Lauric acid	0.08	0.08	0.09	0.08	0.100	0.09	0.08	0.08	0.11	0.13	0.07	0.07	0.09	0.04	0.08
Miristic acid	36.42	37.01	40.75	35.51	44.880	39.55	36.43	36.40	40.58	44.61	35.03	36.63	38.85	38.77	38.92
Palmitic acid	0.11	-	-	0.25	0.160	-	0.10	0.09	-	-	-	0.24	0.11	0.08	0.08
Palmitoleic acid	0.095	0.08	0.095	0.09	0.120	0.09	0.08	0.09	0.09	0.11	0.09	0.08	0.09	0.09	0.09
Margaric acid	7.38	7.34	8.31	7.29	8.620	7.72	7.29	7.18	8.05	8.73	7.76	7.47	7.77	7.85	7.74
Stearic acid	9.37	9.55	9.99	9.08	10.010	9.56	9.26	9.13	9.52	10.68	9.48	9.22	9.67	9.54	9.43
Oleic acid	41.77	41.36	35.87	42.91	31.350	38.37	41.94	42.40	37.01	31.1	42.39	41.93	38.65	38.71	39.10
Linoleic acid	1.44	1.31	1.05	1.44	0.930	1.14	1.38	1.47	1.03	0.72	1.31	1.34	1.17	1.23	1.17
Alpha-Linoleic acid	2.43	2.36	2.77	2.49	2.860	2.5	2.38	2.3	2.62	2.91	2.80	2.33	2.59	2.65	2.48
Arachidic acid	2.33	0.38	0.36	0.34	0.380	0.36	0.34	0.37	0.27	0.4	0.26	0.33	0.36	0.34	0.34
Eicosenoic acid	0.53	0.51	0.61	0.55	0.610	0.57	0.48	0.49	0.57	0.64	0.63	0.48	0.56	0.58	0.54
LSD	0.09	0.39	1.997	1.72	0.152	0279	0471.00	020.00	0.16	0.43	032.00	017.00			

Percentage of margirc changed from 10.46, 9.44 and 9.19% after storage for 9 months at 25, 8 and 0°C to be 10.78, 10.4 and 9.97%, respectively after infection decreased the level of stearic acid from 11.78%, in the control, to 11.63% in the infected beans stored at 25°C. While storage at 8 and 0°C change the % from 10.59, 11.73-12.38 and 11.82%, respectively. Percentage of oleic acid also showed decrease after infection and storage. Control beans have acid percentage 16.76, 23.92 and 21.61% while after infection, percentages changed to be 15.75, 18.29 and 21.08% at 8, 0, 25°C.

Results in Table 3 showed results of treatment of Barry. Oleic acid in these beans changed from 16.76, 32.29 and 21.61%, in control experiment to 15.75, 18.29 and 21.68% after infection and storage for 3 months at 0, 8, 25°C, to be 38.85, 38.77% in infected beans after storage of or 3 months. At 0°C, the acid level decreased from 40.75-38.92% after infection. Margaric acid recorded percentages of 7.38, 7.34 and 8.31% in healthy beans; percentages increased to be 7.77, 7.85 when stored at 8, 25°C while infection and storage at 0°C altered this percentage to be 7.74%. Storage of control beans at 25°C recorded level of seatric acid to be 9.37%. The level in treated beans increased to be 9.67%; while storage at 0, 8°C showed slightly decrease in the level which changed

Table 4: Effect of artificial inoculation with tested fungi on fatty acids of stored Barry variety of coffee beans after 9 months

	Control			*A. niger*			*A. alliaceus*			*A. melleus*			Average of fatty acids		
Fatty acids	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°	25°	8°	0°
Caprilic acid	0.040	0.06	0.090	0.07	0.06	0.070	0.07	0.060	0.04	0.06	0.06	0.01	0.06	0.06	0.04
Lauric acid	0.110	0.06	0.080	0.13	0.11	0.080	0.12	0.060	0.08	0.14	0.07	0.12	0.12	0.08	0.09
Miristic acid	48.020	52.61	52.300	51.52	43.63	53.460	46.22	46.900	55.31	59.21	49.99	49.33	52.31	46.85	52.70
Palmitic acid	0.190	-	0.430	0.23	0.20	0.240	0.35	0.230	0.20	-	0.19	-	0.34	0.14	0.14
Palmitoleic acid	0.120	0.13	0.140	0.13	0.12	0.130	0.12	0.110	0.14	0.14	0.12	0.12	0.13	0.11	0.13
Margaric acid	9.940	10.45	10.620	10.49	8.70	10.530	9.53	9.300	11.15	11.96	9.95	9.76	10.66	9.31	10.48
Stearic acid	11.450	11.51	11.400	11.01	10.24	11.340	10.63	10.400	11.75	11.76	10.59	10.89	11.13	10.41	11.32
Oleic acid	25.640	19.81	19.360	21.08	31.72	18.770	27.75	28.130	15.85	10.97	23.68	24.59	19.93	27.84	19.74
Linoleic acid	0.420	0.27	0.310	0.35	0.93	0.330	0.51	0.540	0.19	0.11	0.59	0.39	0.32	0.68	0.3
Alpha-Linoleic acid	2.800	3.59	3.830	3.63	3.05	3.680	3.32	3.220	3.91	4.12	3.54	3.38	3.68	3.27	3.67
Arachidic acid	0.450	0.46	0.470	0.44	0.41	0.480	0.44	0.430	0.50	0.51	0.40	0.46	0.46	0.41	0.48
Eicosenoic acid	0.870	0.99	0.950	0.99	0.80	0.980	0.91	0.720	0.98	1.00	0.88	0.89	0.97	0.79	0.95
LSD	0.799	0.369	0.236	2.55	0.82	0.188	0.165	0.377	0.048	0.18	0.511	0.365			

from 9.55, 9.99 to 9.54, 9.43%. Oleic acid also showed a decrease in level, than normal, from 41.77, 41.36 in control to 38.65 and 38.71% after storage of infected seeds at 8, 25°C while storage at 0°C increased the level from 35.87%, in control to 39.1% in infected seed.

Table 4 showed results of measuring fatty acids for with or without infection. Storage under the same conditions increased level of fatty acid. Miristic acid increased from 48.02-52.31% at 25°C, after infection. Also at 0°C the level changed from 52.3% in uninfected seeds to 52.7%, in infected ones while keeping at 8°C, the acid recorded 52.61% for untreated and 46.85% for infected samples. The same results were recorded for margaric acid, storage to 25°C charged its level from 9.94-10.66% for control and infected beans. Storage at 0, 8°C recorded changes from 10.45 and 10.62% in control to 9.31 and 10.48% in infected beans. Stearic acid recorded decrease in activity at all tested temperatures, after infection. Healthy beans have levels of 11.45%, 11.51% and 11.4%, decreased after infection to be 11.3, 10.4 and 11.32%, respectively. Oleic acid recorded decrease in its level from 25-19.39% for healthy and infected beans at 25°C. Its percentage decreased at 0, 8°C to be 27.84, 19.74% in infected experiments and 19.81, 19.36% for healthy beans.

DISCUSSION

Results showed that for both varieties of coffee beans, level of fatty acid increased after infection and increasing storage periods and temperature; like stearic and margaric and meristic acid. While other fatty acids recorded decrease in level after infection and storage, like oleic acid. These phenomena may be due to transfer of fatty acid to other compounds, or from type to type. This finding is Agreed with other authors (Ahmad Khan and Shahidi, 2000; Muangkaeo *et al.*, 2005).

In all transactions that oleic fatty acid decreased in infected beans compared to control as well as increase the storage period, that decrease due to a tend to interact and change to other fatty acids (Ghasemnez had *et al.*, 2007; Ghasemnez had and Honermeier, 2007). With increasing the period of storage, the level of miristic, margaric and stearic acids increased in infected beans by increasing storage period. Increasing the lysis activity of certain enzymes like lipase share in degrading fatty acids (Ghasemnez had and Honermeier, 2007). They also mentioned that variation of storage period and temperature affect the activity of lipase enzymes in sunflowers. Ghasemnez had *et al.* (2007) had found the same results using the evening primrose and assured

by Ullah *et al.* (2003) when tested soy beans. It is realized that unsaturated fatly acids like oleic, palmetiolic and linoleic and alpha-linoleic acids are easily oxidized in the presence of oxygen, so increasing storage period increase the oxidation rate and vice versa. Metabolic processes of beans during storage and its physiological activity may be reasons of decreasing fatty acid level (Kindle, 1987) who explained that many enzymes are involved in metabolism of fatty acids like acyl oxidase, malate synthase, citrate synthase which need oxygen for its activity. Temperature has variant effect on fatly acid level in coffee beans (Ghasemnez had and Honermeier, 2007). It is obvious that storage conditions affect fungal enzymes its activity, growth and sporulation. These conditions help fungal hyphae to attack seed tissues led to its deterioration. These results agreed with the findings of Parenicova *et al.* (1997), Lang and Dornenburg (2000) and Silva *et al.* (2008), who proved that the deterioration of beans is directly proportional to the humidity, temperature and storage period. Batista *et al.* (2002) and Gerrit (2003) mentioned that increasing tissue coloration in corn beans. Coloration may include the testa or internal tissues. Cole and Milner (1953) explained coloration in beans to the reaction between nitrogenous compounds and reduced sugars, but Osman (1982) explained the brown color in internal tissue is due to oxidation of fats and fatty acids.

CONCLUSION

Coffee beans of two varieties were tested by artificial inoculation with each of *A. niger, A. alliances* and *A. melleus* were selected for further laboratory long storage tests under different temperatures and different periods of storage. the fatty acids to the seeds of Arabic coffee on the stored samples showed that they may contain a group of fatty acids which were Caprilic, Lauric, Miristic, Palmitic, Palmitoleic, Margaric, Stearic, Oleic, Linoleic, Alpha-Linoleic and Arachidic acids. Generally, the treatment of coffee beans with fungi resulted to increase of each of Miristic, Stearic, Margaric acid and the storage period whereas decreased the proportion of oleic acid.

REFERENCES

Ahmad Khan, M. and F. Shahidi, 2000. Oxidative stability of stripped and no stripped borage and evening primrose oils and their emulsions in water. J. Am. Oil Chem. Soc., 77: 963-968.

Ahmad, R. and N. Magan, 2006. Monsooned coffee: impact of environmental factors on fungal community structure, enzyme production profiles and ochratoxin. Aspects Applied Biol., 68: 161-168.

Batista, L.R., S.M. Chalffoun, G. Prado, R.F. Schwan and A.E. Wheals, 2002. Toxigenic fungi associated with processed (green) coffee beans (*Coffea arabica* L.). Int. J. Food Microbiol, 62: 1-8.

Betancourt, L.E. and H.K. Frank, 1983. Behavior of ochratoxin A during green coffee roasting and soluble coffee manufacture. J. Agric. Food Chem., 46: 673-675.

Brockerhoff, H., 1965. Stereo specific analysis of triglyciedes. J. Lipid Res., 6: 10-15.

Christov, L.P. and B.A. Prior, 1993. Esterases of xylan-degrading microorganisms: production, properties and significance. Enzyme Microbiol. Technol., 15: 460-475.

Cole, E.W. and M. Milner, 1953. Colorimetric and fluorometric properties of wheat in relation to germ damage. Cereal Chem., 30: 378-391.

Faulds, C.B. and G. Williamson, 1994. Purification and characterization of a ferulic acid esterase (FAE-III) from *Aspergillus niger*: Specificity for the phenolic moiety and binding to microcrystal I ine cellulose. Microbiology, 140: 779-787.

Gerrit, H.G., 2003. Enhancement of coffee quality by mould prevention. Food Control, 14: 245-249.

Ghasemnez had, A. and B. Honermeier, 2007. Influence of storage conditions on quality and viability of high and low oleic sunflower seeds. Int. J. Plant Prod., 3: 39-48.

Ghasemnez had, A., S. Cergel and B. Honermeier, 2007. The impact of storage time and storage temperature on the quality of the oil of evening primrose (*Oenothera biennis* L.). J. Med. Spice Plants, 12: 175-180.

Kindle, H., 1987. β-Oxidation of Fatty Acids by Specific Organelles. In: The Biochemistry of Plants. 9, Lipids: Structure and Function, Stumpf, P.K. and E.E. Conn (Eds.). Chapter 2, Harcourt Brace Jovanovich Publishing Company, San Diego, California, pp: 31-53.

Kroon, P.A. and G. Williamson, 1996. Release of ferulic acid from sugar-beet pulp by using arabinase, arabinofuranosidase and an esterase from *Aspergillus niger*. Biotechnol. Applied Biochem., 23: 263-267.

Kroon, P.A., C.B. Faulds and G. Williamson, 1996. Purification and characterization of a novel esterase induced by growth of *Aspergillus niger* on sugar-beet pulp. Biotechnol. Applied Biochem, 23: 255-262.

Lang, C. and H. Dornenburg, 2000. Perspectives in the biological function and the technological application of polygalacturonases. Applied Microbiol. Biotechnol., 53: 366-375.

Mabbett, T., 2002. Storing up problems? The crucial factors in the handling of Coffee and Cocoa beans before, during and after shipment from origin and the way they affect the final product. Coffee Cocoa Int., 5: 7-7.

Martini, S. and M.S. Anon, 2005. Storage of sunflower seed: Variation on the wax content of the oil. Eur. J. Lipid Sci. Technol., 107: 74-79.

Mekete, B., 2003. Coffee, in von Uhlig. In: Encyclopaedia Aethiopica, Siegbert (Ed.). Horrowitz, Weissbaden, Pages: 763.

Morello, J., M. Motilva, M. Tovar and M. Romero, 2004. Changes in commercial virgin olive oil (cv. Arbequina) during storage, with special emphasis on the phenolic fraction. Food Chem., 85: 357-364.

Muangkaeo, R., S. Srichuwong, E. Pawelzik and S. Verasilp, 2005. Influence of packaging material and storage time on seed viability and chemical component of rice seed. Proceedings of the Global Food and Product Chain Dynamics, Innovations, Conflicts, Strategies, oct. 11-13, Deucher Tropentag, Hohenheim, pp: 1-5.

Neg, S.C. and A. Anderson, 2005. Lipid oxidation in Quinoa (*Chenopodium quinoa*) as determinated through accelerated aging. J. Environ. Agric. Food Chem., 4: 1010-1020.

Osman, H.Y.A., 1982. Studies on fungi associated with sorghum grains during storage. Ph.D. Thesis, Plant Path. Faculty Agricultural Cairo University Egyptian.

Parenicova, L., J.A.E. Benen, R.A. Samson and J. Visser, 1997. Evaluation of RFLP analysis of the classification of selected black *Aspergilli*. Mycol. Res., 101: 810-814.

Silv, C.F., R.F. Scwan, E.S. Dias and A.E. Wheals, 2000. Microbial diversity during maturation and natural processing of coffee cherries of coffee Arabica in Brazil. Int. J. Food Microbiol., 30: 251-260.

Silva, C.F., L.R. Batista, L. Abreu, E. Dias and R.F. Schwan, 2008. Microbial interactions in fruits and beans of natural Brazilian coffee (*Coffea arabica* L.). Federal University of Lavras, Brazil.

Sisman, C.B. and L. Delibas, 2004. Storing sunflower seed and quality losses during storage. J. Central Eur. Agric., 4: 239-250.

Sisman, C.B., 2005. Quality losses in temporary sunflower seed stores and influences of storage conditions on quality losses during storage. J. Cent. Eur. Agric., 6: 143-150.

Tenkanen, M., J. Schuseil, J. Puls and K. Poutanen, 1991. Production, purification and characterisation of an esterase liberating phenolic acids from lignocellulosics. J. Biotechnol., 18: 69-84.

Ullah, J., M. Hamayoun, T. Ahmad, M. Ayub and M. Zafarullah, 2003. Effect of light, natural and synthetic antioxidants on stability of edible oil and fats. Asian J. Plant Sci., 2: 1192-1194.

Vila, M., S. Andueza, P. de Pena and C. Cid, 2005. Fatty acid evolution during the storage of ground, roasted coffees. JAocS, 82: 639-646.

Villiers, J.B.M., L.J. Toit and H.E. Pretorius, 1986. The behaviour of decorticated sunflower seeds stored under favourable and unfavourable conditions. Lebensrn-Wiss U- Technol., 19: 472-476.