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INFLUENCE OF ROASTING AND STORAGE CONDITIONS ON BIOCHEMICAL CONSTITUENTS OF ARABICA COFFEE BEANS

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ABSTRACT

The changes of biochemical compositions of arabica coffee beans were determined before and after roasting and storage. The caffeine content increased due to storage coffee beans under atmospheric air or nitrogen gas. The roasting of beans at 180°C for 15, 20 and 30 min to produce light, medium and dark coffee increased caffeine content in all samples. It was found that trigonelline content decreased as a result of both roasting and storage. The effect of roasting and storage of coffee beans for 3 and 6 months revealed a presence of new fatty acids as a degradable product. The beans stored under air or nitrogen, short chain fatty acids were appeared and linoleic was decreased, while palmitic and stearic were increased. The remarkable changes in the fatty acid composition showed especially when stored under atmospheric air, mean while storage under nitrogen resulted in less degradation in the unsaturated fatty acids.

INTRODUCTION

Coffee arabica is one of the most important variety within more than 80 species of genus *Coffea* L. (Rubiaceae) as stated by Casal *et al.* (2000, a). Arabica coffee is easily distinguished by their size, shape and color. Once roasted, these visual characters are eliminated. So, other criterion will be required to discriminate roasted coffee beans.

During roasting of coffee bean reads, dramatic and very complicated chemical changes take place. Castillo *et al.* (2002) demonstrated that physical and chemical changes take place during roasting of green coffee beans. Clifford (1987) and Macrae (1989) reported that caffeine is probably the single most analyzed chemical factor in coffee which has been frequently in discrimination of green coffee varieties. Michael (1979) reported that coffee beans contained 1% caffeine and 1% trigonelline. Trigonelline content decreased and nicotinic acid increased during roasting temperature > 180°C. The conversion of trigonelline to nicotinic acid being flavored by high temperature (Taguchi *et al.,* 1985).

Correia *et al.* (1995); Andrade *et al.* (1997) and Bicchi *et al.* (1995) suggested that chlorogenic acids either alone or in conjunction with caffeine can also be applied to this discrimination. Also Casal *et al.* (2000, a) concluded that trigonelline and caffeine can be used in the discrimination of pure roasted coffees.

Folstar *et al.* (1976) reported that coffee oil contained mainly palmitic, stearic, oleic and linoleic fatty acids. Higher proportions of the unsaturated fatty acids were found acids in the triglycerides in most vegetable oils.

Folstar (1989) and Speer *et al.* (1991) mentioned that the compositions of unsaponifiable matter represented the main difference between the two varieties: coffea arabica and coffea conephora.

Gianturco (1967) and Nakabayashi and Mansano (1986) found that the ratio of trigonelene to caffeine (T/C) indicated the degree of roasting bean. This value decreased proportionally with the progress of roasting during roasting of coffee beans, caffeine content increased gradually and trigonilline content decreased suddenly after the medium roasting period owing to thermal decomposition.

The main target of the present study is to evaluate the effect of roasting and storage under either air or nitrogen on the caffeine, trigonelline contents and fatty acids profile of arabica coffee bean seeds.

MATERIALS AND METHODS

The green arabica coffee bean seeds were purchased from the local market. The green beans were cleaned and roasted at 180°C for 15, 20 and 25 min to produce light, medium and dark coffee. Either green or roasted beans were stored for 3 and 6 months in aluminum bags under nitrogen in comparison with air.

Extraction of caffeine and trigonelline:

The ground coffee bean samples (2 g) were boiled in deionized water (50 ml) for 5 min with occasional shaking. After coaling to room temperature, it was filtered directly into 100 ml volumetric flask and made up to the mark with deionized water (Casel *et al.,* 1998).

Separation and determination of caffeine and trigonelline by high performance liquid chromatography (HPLC):

HPLC analysis was achieved as described by Casal *et al.* (1998); with an analytical HPLC consisting of two PU 980 pumps, A reversed-phase Spherisorb ODS2 (5 µm particle size, 25.0 × 0.46 cm) column was used. The solvent system used was a gradient of phosphate buffer pH 4.0 (0.1 M) and methanol performed at a constant flow rate of 1.0 mL min^{-1} at room temperature. Detection was accomplished with a diode-array detector, and chromatograms were recorded at 265 nm.

The compounds were identified by their retention times, chromatographic comparisons with authentic standards, and their UV spectra. Quantification was based on the external standard method. Under the assay conditions described, a linear relationship between the concentration and the UV absorbance was obtained at 268 nm for trigonelline and at 276 nm for caffeine.

Lipids extraction:

The lipids of the ground coffee bean samples were extracted with petroleum ether for overnight at room temperature.

Separation and identification of coffee bean oil fatty acids by Gas

chromatography:

The extracted coffee bean oil was saponified with KOH (20%, w/v) for overnight at room temperature. The unsaponifiable matters were extracted three times with hexane. The aqueous layer was acidified with H_2SO_4 (20%, v/v) and the liberated fatty acids were extracted with hexane, then washed several times with distilled water, and dried over anhydrous sodium sulfate.

The separated fatty acids were converted to methyl esters with diazomethane solution (Vogel, 1975). The fatty acid methyl esters were analyzed using a Hewlett Packard 5890 gas chromatograph equipped with flame ionization detector. The chromatograph was fitted with HP-INOWA × 30 m × 0.32 mm × 0.5 µm capillary column coated with polyethylene glycol. The column over temperature was programmed at 10°C/min from 100 to 220°C with nitrogen flow rate of 2 ml/min as carrier gas. Hydrogen and air flow rates were 30 and 300 ml/min, respectively (Farag *et al.,* 1985). Under the above mentioned conditions, all peaks from C_{10} to C_{20} homologous series were well identified. Peak identification was performed by comparison of the relative retention time (RRT) for such peak with those of standard chromatograms. The fatty acid percentage was performed using Hewlett Packard 5890 series recording integrator.

RESULTS AND DISCUSSION

Table (1) and Fig. (1) shows the caffeine content in both green and light, medium and dark roasted coffee bean as affected with either roasting or storage conditions. The data revealed that the green beans contained 1.1% of caffeine while light, medium and dark roasted beans presented 1.22, 1.32 and 1.32% for the same component, respectively. These results were in the line with those obtained by Nakabayashi and Mansano (1986).

Concerning the effect of storage on green coffee beans, it was found that storage exhibited gradual increase in caffeine content, either under atmospheric air or under nitrogen gas. Similar trend was observed when roasted beans were considered. However, storage under nitrogen gas showed a valuable increase in caffeine content comparable with those obtained if storage under atmospheric air was applied.

It could be concluded that the increase in caffeine content during storage may be due to the losses of some biochemical compounds or to the release of flavor ones. The obtained data are in agreement with those determined by Cloughley (1982) who mentioned that tea stored for 6 months contained more caffeine than freshly manufactured product. Also present data confirmed with those of Sayed (1997) who found that a gradual increase in caffeine content due to storage conditions and coffee bean variety.

Table (1): Effect of roasting and storage conditions on the caffeine

contents of arabica coffee beans (g/100 g d.w.).

Roasted coffee beans	**Storage period (months)**		
	Zero time	**3**	**6**
	Stored under atmospheric air		
Green	1.10	1.20	1.44
Light roasting	1.22	1.43	1.76
Medium roasting	1.32	1.62	1.91
Dark roasting	1.32	1.57	1.95
	Stored under N_2		
Green	1.10	1.11	1.13
Light roasting	1.22	1.52	2.04
Medium roasting	1.32	1.56	1.93
Dark roasting	1.32	1.64	2.00

fig1

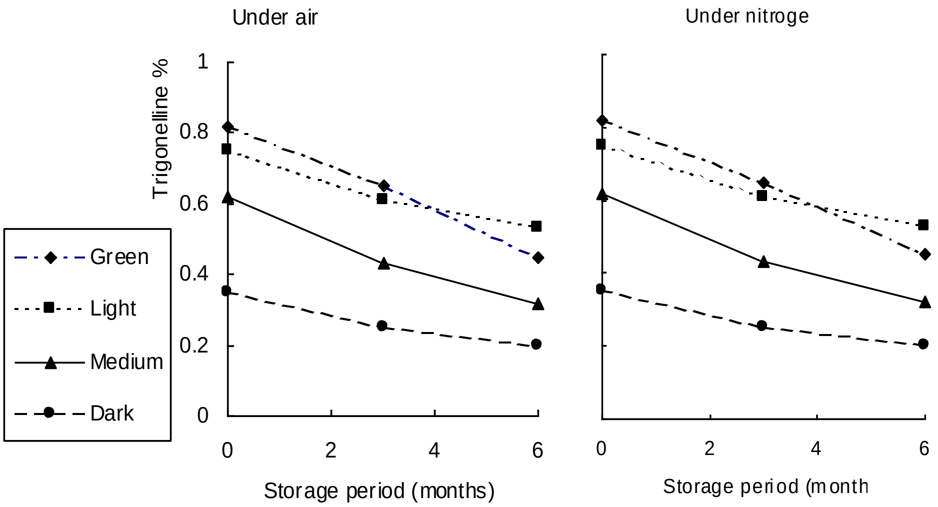
Fig. (1): Effect of roasting and storage conditions on the caffeine contents.

Table (2) and Fig. (2) indicated that trigonelline contents decreased after roasting. The green beans had a higher value of 0.82% trigonelline content. However, light, medium and dark roasted beans had lower values of 0.75, 0.60 and 0.35% for the same component, respectively.

These results are in agreement with those reported by Nakabayashi and Mansano (1986) and Castillo *et al.* (2002). In this connection, Trugo and Marae (1989) stated that loss of trigonelline was strongly dependent on the degree of roasting. The present data revealed that trigonelline decrease as affected by storage under air or nitrogen gas. Regarding these results, Trugo and Macrae (1989) and Cassal *et al.* (2000,b) indicated that decrease in trigonelline content may be explained by an increase in nicotinic acid.

Table (2): Effect of roasting and storage conditions on trigonelline contents of arabica coffee beans (g / 100 g d.w.).

Roasted coffee beans	Storage period (months)		
	Zero time	3	6
	Stored under atmospheric air		
Green	0.82	0.65	0.45
Light roasting	0.75	0.61	0.53
Medium roasting	0.62	0.43	0.32
Dark roasting	0.35	0.25	0.20
	Stored under N_2		
Green	0.82	0.75	0.65
Light roasting	0.75	0.60	0.55
Medium roasting	0.62	0.45	0.35
Dark roasting	0.35	0.30	0.20



(2): Effect of roasting and storage conditions on the trigonelline content.

Table (3) and Fig. (3) shows the fatty acids composition of green coffee bean stored for 3 and 6 months either under atmospheric air or under nitrogen gas. These data revealed the presence of some new fatty acids as a degradable product which may be attributed to storage conditions. The total new fatty acids composed 5.78 and 6.83% due to storage under atmospheric air for 3 and 6 months respectively, while it was 3.23 and 4.37% for the same period when nitrogen gas was used for storage.

From the above mentioned data, it could be demonstrated that the appearance of new medium chain fatty acids during green coffee beans storage may be explained as a biochemical activities during storage.

Table (3): Effect of storage conditions on the fatty acids composition of green arabica coffee beans (%).

Fatty acids	Storage conditions

	Zero time	Under air		Under Nitrogen	
		3	6	3	6
$C_{12:0}$	–	1.06	2.66	1.38	1.41
$C_{13:0}$	–	0.83	0.91	–	–
$C_{13:1}$	–	0.77	0.78	–	–
$C_{14:0}$	–	1.17	2.00	1.85	2.10
$C_{14:1}$	–	1.11	0.48	–	0.86
$C_{15:0}$	–	0.84	–	–	–
$C_{16:0}$	9.58	9.66	9.71	9.61	9.63
$C_{18:0}$	12.56	13.11	13.21	12.61	12.72
$C_{18:1}$	10.91	11.33	11.38	11.06	11.15
$C_{18:2}$	66.95	60.12	58.88	63.50	62.13
TU	77.86	73.33	71.52	74.56	74.14
TS	22.14	26.67	28.49	25.45	25.86
TU/TS	3.52	2.75	2.51	2.93	2.87

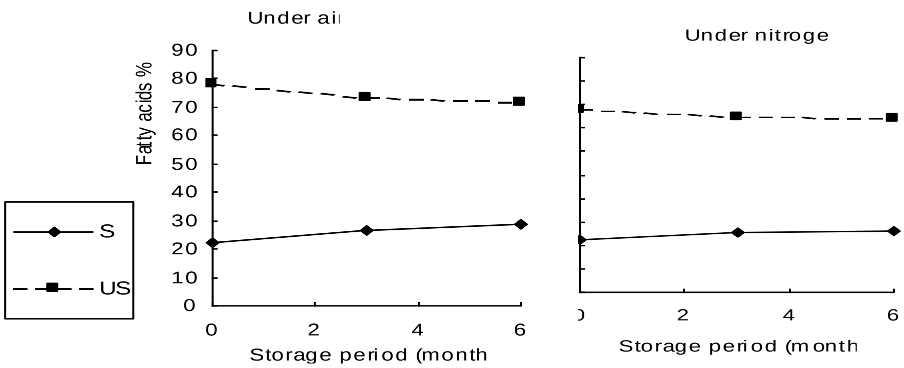


Fig. (3): Effect of storage on the fatty acids content of coffee beans.
TU = Total unsaturated fatty acids. **T S = Total saturated fatty acids.**
TU/TS = The ratio between total unsaturated and total saturated fatty acids.

On the other hand, it can be observed that the total amount of the saturated fatty acids increased, while that of the unsaturated ones decreased during storage, either under atmospheric air or under nitrogen gas. Results which are in accordance with those determined by Simonova and Solov'eva (1980). They found that coffee during storage in air or CO_2 for less than 8 months revealed a decrease in unsaturated fatty acids and an increase in saturated fatty acids.

Roasting of arabica green beans to produce light, medium and dark roasted beans exhibited a dramatic changes in the fatty acids composition as illustrated in Table (4) and Fig. (4). The variation in the fatty acids percentage may be due to both time and temperature of roasting procedure, which catalyze the autoxidation of the unsaturated fatty acids to produce the short and medium chain fatty acids.

Table (4): Effect of roasting on the fatty acids composition of arabica coffee beans (%).

Fatty acids	Green coffee beans	Roasted coffee beans		
		Light	Medium	Dark

$C_{12:0}$	–	2.15	3.00	4.86
$C_{13:0}$	–	0.50	1.70	2.13
$C_{13:1}$	–	0.81	0.91	1.17
$C_{14:0}$	–	6.17	7.12	7.52
$C_{14:1}$	–	4.31	5.00	5.80
$C_{15:0}$	–	0.70	1.13	1.22
$C_{16:0}$	9.58	13.13	14.00	14.17
$C_{18:0}$	12.56	12.87	13.51	13.88
$C_{18:1}$	10.91	10.11	9.87	9.51
$C_{18:2}$	66.95	49.25	43.76	39.74
TU	77.86	64.48	59.54	56.22
TS	22.14	35.52	40.46	43.78
TU/TS	3.52	1.82	1.47	1.28

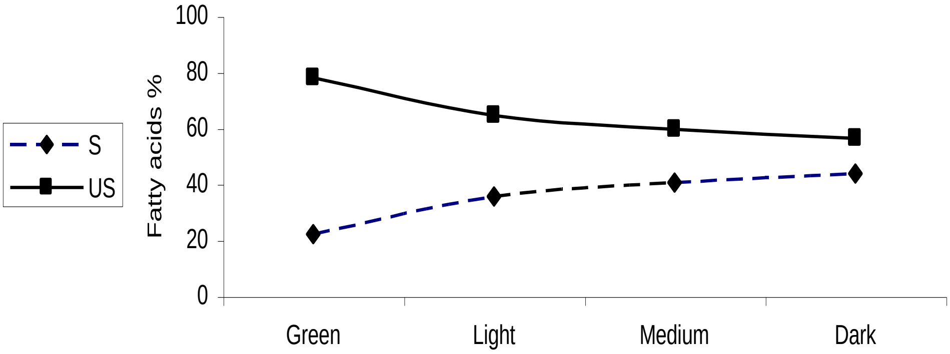


Fig. (4): Effect of roasting on the fatty acids content of coffee beans.

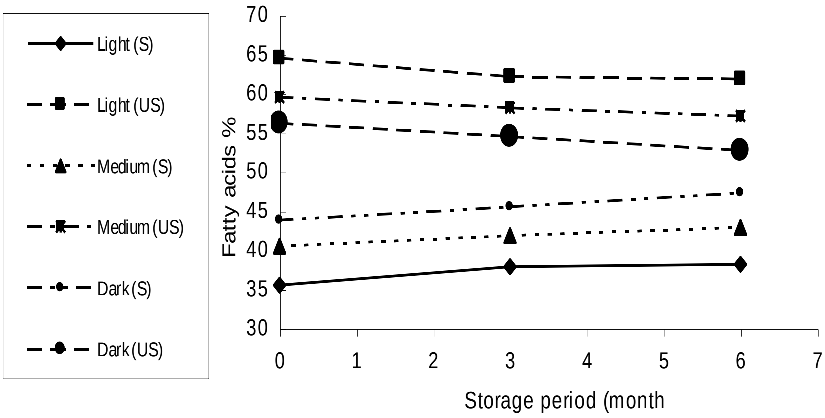
TU = Total unsaturated fatty acids. TS = Total saturated fatty acids.
TU/TS = The ratio between total unsaturated and total saturated fatty acids.

Regarding the stored roasted coffee beans, it could be observed a remarkable changes in the fatty acids composition, especially when storage under atmospheric air (Table 5 and Fig. 5). In the same time, storage under nitrogen gas resulted in less degradation of the unsaturated fatty acids to produce short or medium chain fatty acids (Table 6 and Fig. 6).

Table (5): Effect of storage conditions on the fatty acids composition of the roasted ground arabica coffee beans under atmospheric air (%).

Storage	Storage periods (months)

conditions Fatty acids	Light roasting			Medium roasting			Dark roasting		
	Zero	3	6	Zero	3	6	Zero	3	6
$C_{12:0}$	2.15	2.35	2.41	3.00	3.22	3.38	4.86	4.98	5.21
$C_{13:0}$	0.50	1.32	1.37	1.70	1.91	2.11	2.13	2.51	2.87
$C_{13:1}$	0.81	1.10	1.10	0.91	1.22	1.41	1.17	2.20	2.27
$C_{14:0}$	6.17	6.22	6.26	7.12	7.44	7.52	7.52	7.70	7.91
$C_{14:1}$	4.31	4.37	4.42	5.00	5.18	5.32	5.80	6.00	6.42
$C_{15:0}$	0.70	0.81	0.87	1.13	1.27	1.41	1.22	1.47	1.85
$C_{16:0}$	13.13	13.23	13.25	14.00	14.32	14.55	14.17	14.72	15.10
$C_{18:0}$	12.87	13.92	14.00	13.51	13.66	13.91	13.88	14.11	14.37
$C_{18:1}$	10.11	10.00	9.78	9.87	9.80	9.72	9.51	9.40	9.33
$C_{18:2}$	49.25	46.68	46.54	43.76	41.98	40.67	39.74	36.91	34.67
TU	64.48	62.15	61.84	59.54	58.18	57.12	56.22	54.51	52.69
TS	35.52	37.85	38.16	40.46	41.82	42.88	43.78	45.49	47.31
TU/TS	1.82	1.64	1.62	1.47	1.39	1.33	1.28	1.98	1.11



ig. (5): Effect of storage conditions on the fatty acids content of the roasted coffee under air.

TU = Total unsaturated fatty acids. TS = Total saturated fatty acids.
TU/TS = The ratio between total unsaturated and total saturated fatty acids.

From the above mentioned data, it could be concluded that the appearance of new medium chain fatty acids during green coffee bean storage may be due to the biochemical activities during storage. Concerning roasting, the variation in the fatty acids percentage may be due to the roasting time and heat temperature which help or catalyze the autoxidation of the unsaturated fatty acids to produce the short and medium chain fatty acids. On the other hand, storage under nitrogen gas may prevent or delay the oxidation of the unsaturated compounds. Whereas storage under atmospheric air may enhance that as a result of the presence of some transitive elements such as

Fe^{2+}. The obtained data were in the line with the findings of Sayed (1997) who found a variable changes in the fatty acids composition during roasting and storage of the coffee beans.

Table (6): Effect of storage conditions on the fatty acids composition of the roasted ground arabica coffee beans under nitrogen gas (%).

Storage conditions Fatty acids	Storage periods (months)								
	Light roasting			Medium roasting			Dark roasting		
	Zero	3	6	Zero	3	6	Zero	3	6
$C_{12:0}$	2.15	2.18	2.20	3.00	3.15	3.27	4.86	4.93	5.16
$C_{13:0}$	0.50	0.57	0.60	1.70	1.77	1.91	2.13	2.22	2.37
$C_{13:1}$	0.81	0.88	0.90	0.91	0.93	1.00	1.17	1.22	1.51
$C_{14:0}$	6.17	6.23	6.27	7.12	7.17	7.22	7.52	7.63	8.10
$C_{14:1}$	4.31	4.33	4.38	5.00	5.11	5.31	5.80	5.88	6.17
$C_{15:0}$	0.70	0.75	0.75	1.13	1.15	1.27	1.22	1.27	1.31
$C_{16:0}$	13.13	13.17	13.27	14.00	14.20	14.30	14.17	14.25	14.86
$C_{18:0}$	12.87	12.91	12.94	13.51	13.58	13.61	13.88	14.12	14.38
$C_{18:1}$	10.11	10.10	10.10	9.87	9.85	9.82	9.51	9.48	9.38
$C_{18:2}$	49.25	48.88	48.58	43.76	43.10	42.29	39.74	39.00	36.76
TU	64.48	64.19	63.96	59.54	58.99	58.42	56.22	55.58	53.82
TS	35.52	35.81	36.03	40.46	41.02	41.58	43.78	44.42	46.18
TU/TS	1.82	1.79	1.78	1.47	1.44	1.41	1.28	1.25	1.17

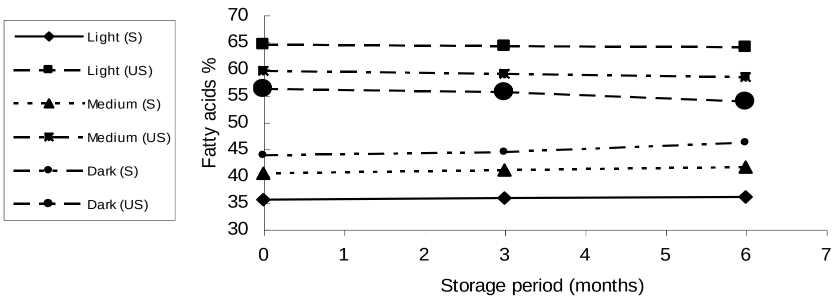


Fig. (6): Effect of storage conditions on the fatty acids content of the roasted coffee under nitrogen.

TU = Total unsaturated fatty acids. TS = Total saturated fatty acids.

TU/TS = The ratio between total unsaturated and total saturated fatty acids.

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تأثير التخزين والتحميص على المكونات الكيميائية لحبوب القهوة العربى

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الهدف من هذا البحث هو دراسة تأثير التحميص على حبوب القهوة على درجة ١٨٠°م لمدة ١٥ ، ٢٠ ، ٣٠ دقيقة وأيضاً التخزين لمدة ٣ ، ٦ شهور فى الجو العادى وتحت غاز النيتروجين على المكونات الكيميائية فى بذور القهوة العربى .

وقد وجد من الدراسة أن :

- زيادة فى محتوى الكافين كنتيجة للتحميص على درجة ١٨٠°م كما أدى إلى تكوين قهوة بيضاء اللون ، متوسطة اللون ، غامقة اللون على التوالى .
- زيادة في محتوى الكافين كنتيجة للتحميص والتخزين لمدة ٣ ، ٦ شهور .
- أدى التحميص أيضاً إلى نقص فى محتوى الترايجونيلين ، كما أدى التخزين لمدة ٣ ، ٦ شهور فى الظروف الجوية العادية وتحت غاز النيتروجين إلى نقص فى محتوى الترايجونيلين.
- أدى التحميص والتخزين إلى إنخفاض نسبة الأحماض الدهنية الغير مشبعة (أوليك – لينوليك) وظهور أحماض دهنية مشبعة قصيرة السلسلة ، بينما زادت نسبة الأحماض الدهنية المشبعة (الأستياريك – البالمتيك) .
- كان تأثير التخزين تحت ظروف الهواء الجوى أكثر وضوحاً فى إنتاج أحماض دهنية مشبعة عنه فى حالة التخزين تحت غاز النيتروجين .