

Physicochemical Properties of Single Cell Oil Extracted from Oleaginous Yeasts

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Abstract: The study aimed at extracting single cell oil (SCO) from some oleaginous yeasts living on wheat straw and sugarcane molasses media and to determine physicochemical properties of the produced oil for further assessment as edible oil. Thirty samples of yeasts were isolated according to the different methods based on the source of isolates. The effectiveness of oleaginous yeast to produce the single cell oil was studied, when the yeast was grown on two types of wheat straw media (detoxified liquid hydrolysate (DLH) and non-detoxified liquid hydrolysate (NDLH)), and molasse media. Single cell oil was extracted by hexane. *Saccharomyces cerevisiae* and *Pichia guilliermondii* were used to produce SCO. SCO produced from oleaginous yeasts in sugarcane molasses media was significantly ($P \leq 0.05$) higher than that produced in wheat straw media. DLH of wheat straw gave higher productivity of oil than NDLH wheat straw. Generally, *S. cerevisiae* gave higher oil productivity (84 %), compared to *P. guilliermondii* (52 %). The results indicated that most of the physicochemical properties of extracted oil were found to be within the recommended limit of the common edible oil reported, except for the iron content; it ranged from 21 to 24 mg/kg, which was 4-5 times more than the permissible level of that of the Codex Alimentarius Commission. In addition, single cell oil produced by

molasses contained omega-3 fatty acids such as eicosapentaenoic and docosahexaenoic which are not found in vegetable oil. Also the unsaturated fatty acids of SCO produced in wheat straw media were higher than that produced in molasses media. The fatty acid profile of SCO produced in molasses media was different from that of vegetable oils. The good physicochemical characteristics and fatty acids profile of SCO from wheat straw make it safe and a promising potential raw material for production of edible oil. The result of this study suggests the isolation of oleaginous yeast in order to be used in the edible oil production.

Keywords: Oleaginous yeast, *Saccharomyces cerevisiae*, *Pichia guilliermondii*, physicochemical properties, single cell oil

INTRODUCTION

A large number of microorganisms are known to accumulate 20 % or more of their biomass as lipid and have been termed oleaginous yeasts; they are known to utilize hydrolylates of rice hull, rice straw, wheat straw, sugarcane bagasse, fruit and vegetable wastes, and domestic wastes, and thus can be employed for the production of SCO from these waste streams (Ratledge and Cohen, 2008). Mostly yeasts, have been considered as oleaginous microorganisms that have lipid content higher than 20 % per dry weight of their biomass. These organisms are not confined taxonomically and phylogenetically to a closely related group. Some are ascomycetes, such as *Lipomyces starkeyi*, and *Yarrowia lipolytica*; others are basidiomycetes, such as *Cryptococcus curvatus*, *Rhodosporidium toruloides* and *Rhodotorula glutinis* (Ratledge and Cohen 2008). Some oleaginous yeasts have been reported to accumulate lipids up to almost 70 % of their cell dry weight when cultured under nitrogen limited condition. These yeasts belong to genera like *Rhodosporidium*, *Rhodotorula*, *Yarrowia*, *Candida*, *Cryptococcus*, *Trichosporon* and *Lipomyces* (Ageistos *et al.* 2011). The oil produced from those yeasts called single cell oil (SCO). The identification of the

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yeast is based on a combination of morphological and biochemical criteria.

Edible oils are important part of human diet produced from vegetable and animal source and used as ingredients in food products. Edible oil include liquid, semisolid and solid products. They have common properties of being insoluble in water, soluble in organic solvent, rich source of dietary energy, contain the essential fatty acids, contain vitamins and contribute to the food palatability. Recently, there are alternative resources to edible oil, the SCO (microbial oil) from low cost substrates of which composition is similar to traditional vegetable oils (Li *et al.* 2008). It is obvious that SCO will play a more critical role in the future, and low-cost substrates for SCO production will play a key role in the industrialization of SCO production. Several scientists have reviewed SCO and its production over the past decades (Li *et al.* 2008). The objective of this study is to isolate oleaginous yeasts from different sources (rotten fruits, fruit juices, milk, fish and air), to identify them and to study the possibility of production of SCO on wheat straw and sugarcane molasses media and to make a comprehensive characterization of the produced oil for further assessment as edible oil.

MATERIALS AND METHODS

Materials

Samples were collected from food processing factories, households and local markets. Liquid samples were kept in clean and sterile containers, and solid samples were kept in sterile polyethylene bags, then closed and immediately transported to the laboratory of the Department of Food Science and Technology, Faculty of Agriculture, University of Khartoum, Sudan. Oleaginous yeasts were cultured from rotten fruit (mango (M1-M2), grapefruit (Gf1-Gf2), apple (A1-A3) and banana (B1-B3)), air (Ai1-Ai2) and fruit juice (banana (Bj2-Bj3) and orange Oj2)).

Yeast isolation from Different sources

Oleaginous yeasts were isolated from rotten fruit samples of mango, orange and banana. Yeast isolation from rotten fruit samples was carried out, according to the method of Santosh *et al.* (2013). Yeast isolation from milk was carried out, according to the method of Papanikolaou and Aggelis (2002). Yeasts were isolated from air according to the method of Xiaochen *et al.* (2011). Yeasts were isolated from fish according to the method described by Chi *et al.* (2007). Yeast isolation from fruit juice, was done by the method described by Hu *et al.* (2009).

Single cell oil production using wheat straw and molasses media

The detoxified liquid hydrolysate of wheat straw (DLH), non-detoxified liquid hydrolysate of wheat straw (NDLH) and detoxified liquid hydrolysate of molasses (MDLH) media were prepared according to the method described by Chen *et al.* (2009) and then yeasts strains were cultivated in DLH, NDLH and MDLH media according to the methods described by Huang *et al.* (2009).

Dry cell weight determination

To determine the amount of biomass of oleaginous yeasts, a 5 ml cell suspension sample was centrifuged at 2500 rpm for 5 min. The cell pellet was then washed twice with distilled water, dried in a pre-weighed aluminium dish at 105°C for 3 h, and the final mass was expressed as dry cell weight (DCW) (Huang *et al.* 2009).

Oil extraction

Yeast cell were harvested from fermentation broth by centrifugation at 8000 rpm for 10 min then oil was extracted by shaking with hexane in separator according to the procedure of Li *et al.* (2010).

Oil characterization

The oil analysis comprised: physical characteristics (colour, viscosity, refractive index, specific density), and chemical characteristics (free fatty acids, iodine value, acid value, saponification number, peroxide value,

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polymer content, and toxicity). Physical and chemical characteristics were determined according to AOAC (2008).

Statistical analysis

Analysis of Variance (ANOVA) was carried out to test for significant differences among means of the treatments. Duncan's Multiple Range Test (DMRT) was used for further multiple comparisons among the means according to the method described by Montgomery and Douglas (2001).

RESULTS AND DISCUSSION

Oil production (%) of yeast samples using DLH and NDLH of wheat straw

The highest oil % (50.7 %) was found in sample A₁, followed by sample A₃ (48.5 %) in DLH, while the lowest mean (9.7 %) was found in sample Gf₁ in NDLH (Table 1). Variation in oil production value depends on the source of oleaginous yeast isolates and DLH, NDLH of wheat straw media. The data showed significant difference ($P \leq 0.05$) when the single cell oil producing samples were grown on DLH and NDLH. The single cell oil production was higher when using detoxified liquid hydrolysate as substrate to produce single cell oil than using non-detoxified liquid hydrolysate. The high percentage of oil produced in DLH oil sample indicates the importance of detoxification process, this is because the oleaginous strains were unable to efficiently produce lipids in the presence of the inhibitors in the hydrolysate. So a detoxification treatment was required prior to the fermentation (Chen *et al.* 2009). These results were not in agreement with the findings of Xiaochen *et al* (2011) who used this hydrolysate pre-treatment of wheat straw as substrates. Their results showed the lipid contents of 33.5 % and 27.1% in the NDLH and DLH, respectively. Moreover, Lipid concentrations in all yeast strains were slightly higher in the NDLH than in the DLH, indicating that the non-detoxified hydrolysate did not have a negative impact on lipid accumulation.

Oil production using sugar cane molasses substrate

The highest mean value of biomass (2.6 g/l) was detected in the sample Bj₂, while the lowest mean value (1.5 g/l) was detected in sample A₃. No significant difference in biomass was observed among oleaginous yeast samples (Table 1). The mean biomass values of this study was lower than the finding of Thidarat *et al* (2012) (8.2g/l), who used mixed cultures of the oleaginous yeast for microbial oil production using sugar cane molasses as carbon substrate. The highest oil content was found in sample A₁ (1.5 g/l), while the lowest one was found in sample Ai₁ (0.9 g/l). The mean oil content in this study was higher than the findings of Thidarat *et al.* (2012) (0.1 g/l). Yeast sample A₃ on molasses gave the highest % oil production mean value (84.1%), followed by sample A₁, (77.8 %). While the lowest one was found in sample Bj₂ (41. 7 %). There was significant difference ($P \leq 0.05$) among oleaginous yeast samples under this investigation. These results were higher than the findings of Gajdoš *et al.* (2015), who found that the *Saccharomyces cerevisiae* strains accumulated more than 40 % of lipids on sugar cane molasses.

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Table 1. Biomass (g/l), oil content (g/l) and %of oil production of SCO isolated from different sources and grown on wheat straw and molasses hydrolysate media.

| Sample | Biomass (g/l) | | | Oil content (g/l) | | | Oil % | | |
|--------|--------------------------|--------------------------|-----------------------|---------------------------|----------------------------|-------------------------|--------------------------|-------------------------|--------------------------|
| | DLH | NDLH | Molasses | DLH | NDLH | Molasses | DLH | NDLH | Molasses |
| M1 | 1.8±0.13 ^{abcd} | 1.9±0.02 ^{ab} | 1.7±0.06 ^a | 0.36±0.04 ^{fghi} | 0.32±0.01 ^{dehij} | 1.138±0.3 ^{ab} | 22.2±0.0 ^{fg} | 16.8±0.6 ^{jkl} | 67.0±16.7 ^{abc} |
| M3 | 1.8±0.0 ^{cd} | 1.9±0.0 ^a | 2.2±0.2 ^a | 0.47±0.03 ^{fgh} | 0.3±0.01 ^{ehi} | 1.0±0.06 ^b | 20.3±1.4 ^{gh} | 17.4±0.4 ^{ijk} | 46.4±6.2 ^{bcd} |
| Gf1 | 1.4±0.06 ^f | 1.8±0.06 ^{cd} | 2.3±1.1 ^a | 0.3±0.07 ^{ijk} | 0.2±0.01 ^k | 1.2±0.14 ^{ab} | 17.9±3.9 ^{hijk} | 9.7±0.7 ⁿ | 55.1±19.9 ^{abc} |
| Gf2 | 1.5±0.03 ^e | 1.8±0.04 ^{bcd} | 1.6±0.14 ^a | 0.3±0.03 ^{hij} | 0.3±0.06 ^{ijk} | 1.1±0.09 ^{ab} | 20.1±1.3 ^{ghi} | 14.1±2.9 ^{lm} | 68.9±0.9 ^{abc} |
| A1 | 1.3±0.01 ^g | 1.9±0.01 ^a | 2.0±0.8 ^a | 0.7±0.03 ^a | 0.3±0.0 ^{ghi} | 1.5±0.5 ^a | 50.7±1.8 ^a | 18.6±0.4 ^{hij} | 77.8±5.6 ^{ab} |
| A3 | 1.3±0.01 ^g | 1.9±0.03 ^{abc} | 1.5±0.14 ^a | 0.6±0.01 ^{abc} | 0.3±0.0 ^{hij} | 1.3±0.03 ^{ab} | 48.5±0.3 ^{ab} | 15.3±0.4 ^{klm} | 84.1±5.7 ^a |
| B2 | 1.3±0.02 ^g | 1.7±0.06 ^d | 2.4±0.48 ^a | 0.6±0.01 ^{abc} | 0.3±0.0 ^{ik} | 1.2±0.1 ^{ab} | 46.7±1.3 ^{bc} | 12.8±0.1 ^m | 52.8±16.6 ^{abc} |
| B3 | 1.2±0.0 ^g | 1.8±0.01 ^{abcd} | 2.6±0.6 ^a | 0.6±0.0 ^{cd} | 0.4±0.0 ^{rfg} | 1.1±0.1 ^{ab} | 44.3±0.2 ^{cd} | 24.4±0.0 ^f | 44.7±14.3 ^{bcd} |
| Ai1 | 1.2±0.1 ^g | 1.3±0.06 ^g | 1.7±0.35 ^a | 0.4±0.01 ^{efg} | 0.3±0.0 ^{hij} | 0.9±0.0 ^b | 36.4±1.1 ^e | 24.8±0.4 ^f | 58.1±12.2 ^{abc} |
| Ai2 | 1.3±0.01 ^g | 1.3±0.01 ^g | 1.9±0.8 ^a | 0.3±0.0 ^{hij} | 0.3±0.01 ^{hij} | 1.0±0.06 ^b | 23.1±0.0 ^f | 22.7±0.3 ^{fg} | 56.9±24.9 ^{abc} |
| Bj1 | 1.3±0.03 ^g | 1.2±0.02 ^g | 2.5±0.01 ^a | 0.5±0.0 ^{def} | 0.2±0.0 ^k | 1.1±0.19 ^{ab} | 35.8±0.99 ^e | 14.6±0.14 ^{lm} | 43.6±7.6 ^{bcd} |
| Bj2 | 1.3±0.01 ^g | 1.2±0.06 ^g | 2.6±0.48 ^a | 0.3±0.21 ^{hij} | 0.2±0.0 ^k | 1.0±0.13 ^b | 36.5±0.00 ^e | 14.5±0.07 ^{lm} | 41.7±12.7 ^c |
| Oj2 | 1.5±0.07 ^{ef} | 1.5±0.02 ^{ef} | 2.3±0.68 ^a | 0.6±0.05 ^{ab} | 0.5±0.01 ^{abcde} | 1.0±0.11 ^b | 42.6±1.27 ^d | 35.5±0.78 ^e | 46.6±18.24 ^{bc} |

Values with same superscripts along rows and columns are not significantly different at (P≤0.05) according to DMRT.

Oil Characterization

Physicochemical properties of extracted SCO using wheat straw and molasses as substrates:

Refractive index (RI):

Table 2 shows there is no difference in refractive index of SCO from DLH, NDLH and MDLH. These results are similar to the finding of Pearson (2002), who reported that RI of olive and sesame oils was 1.47. The RI of SCO produced (1.47) were found to be within the recommended limit of most edible oils as reported by Codex Alimentary (2008).

Table 2. Physicochemical properties of the single cell oil from wheat straw and molasses

| Parameter | DLH | NDLH | Molasses |
|-------------------------------------|-----------------------|--------------------------------------|--------------------------------------|
| Refractive index | 1.47±0.00 | 1.46±0.01 | 1.46±0.00 |
| Viscosity (cps) | 26.12±0.52 | 23.53±0.05 | 26.11±0.52 |
| Colour value | Blue Yellow Red | 0.07±0.06 27.50±0.20 1.33±0.06 | 0.07±0.06 27.50±0.20 1.33±0.06 |
| Specific density (cm ³) | 0.92±0.01 | 0.95±0.01 | 0.91±0.01 |
| Iodine value (g/100g) | 169.37±0.31 | 178.63±0.31 | 101.60±2.87 |
| Saponification (mg/g KOH) | 167.21±1.65 | 171.48±1.05 | 183.38±3.63 |
| Un-saponification (g/kg) | 8.44±0.16 | 6.40±1.23 | 3.30±0.52 |
| Acid value (mgKOH/g) | 0.80±0.05 | 0.72±0.04 | 0.70±0.12 |
| Peroxide value (m.Eq/kg) | 5.66±0.16 | 3.90±0.08 | 4.28±0.49 |
| Iron (mg/g) | 21.12±0.01 | 8.44±0.16 | 24.47±0.06 |
| Copper (mg/g) | 0.34±0.01 | 0.80±0.05 | 0.28±0.01 |
| Lead (mg/g) | 0.00±0.00 | 5.66±0.16 | 0.04±0.00 |
| Arsenic (mg/g) | 0.01±0.01 | 21.12±0.01 | 0.11±0.00 |
| Polymers (%) | 1.90±0.10 | 1.98±0.16 | 1.73±0.16 |

Values are means ±SD

Viscosity

The viscosity value of oil samples were 26.12, 23.53 and 26.11(cps) for DLH, NDLH and molasses, respectively (Table 2). These results are within the finding of Pearson (2002), who reported the normal value of viscosity for olive oil range from 20.4 to 31.0 (cps).

Colour

Colour value of oil samples were 0.10, 0.07 and 0.03 blue, 31.20, 27.50 and 20.73 yellow, and 1.57, 1.33 and 1.67 red for DLH, NDLH and molasses, respectively (Table 2). Colour value doesn't agree with the findings of Jar Elnabi (2001), who reported the colour of sunflower as 1.0 yellow and 0.1 red.

Specific density (SD)

Specific density of oil samples was 0.92, 0.95 and 0.91 (cm^3) for DLH, NDLH and MDLH, respectively (Table 2). The SD of DLH oil sample was found to be within most of edible oils (Codex 2008), which reported the specific density of different oils (sun flower, sesame and groundnut) varying from 0.91 to 0.93 (cm^3). The SD of NDLH oil sample under investigation was found to be higher than the recommended level, while the SD of MDLH was lower, which may be attributed to the different fatty-acids composition between DLH, NDLH and MDLH oils.

Iodine value (IV)

Table 2 shows (IV) of DLH, NDLH and MDLH was 169.37, 187.63 and 101.60 (g/100g), respectively. IV of DLH and NDLH are higher than the findings of Jar Elnabi (2001), who reported that, the iodine value of 129.4(g/100g) and 126.9(g/100g) for crude and refined sunflower oil, respectively. Also, these readings were higher than the threshold value of most edible oils as olive oil (75-94 g/100g) and sesame seed (104-120mg/g), which increase the probability of oxidation. The high number of iodine is an indication of high unsaturation in these oils and thus they become more vulnerable to oxidation, it may lead to the short storage life of DLH and NDLH oils. The IV of MDLH, however, falls below the

threshold value of sesame seed oil (104-120 g/100g) as reported by Codex (2008). These results indicated the long storage life of this oil.

Saponification number (SN)

The SN was higher in NDLH oil sample (171.48 mg/g KOH) than DLH oil sample (167.21 mg/g KOH) (Table 2), while the SN of MDLH was 183.38 mg/g KOH. These results are lower than Codex (2008) which indicates that, the SN range is 187-196 (mg/g KOH). These results do not affect the quality of produced oil if used as edible oil according to Garrett and Grisham (2012), who reported that the saponification number is only of interest if the oil is for industrial purposes, as it has no nutritional significance.

Unsaponification numbers (USN)

USN of oil samples under investigation are shown in Table 2. USN of DLH, NDLH and MDLH oil samples were 8.44, 6.40 and 3.29 (g/kg), respectively. They were below the recommended value of the most edible oils, which reported the USN of different oils (groundnut, olive and sesame) as varying from 10 to 20 (g/kg) (Codex 2008).

Acid value (AV)

AV of DLH, NDLH and MDLH oil samples were 0.80, 0.72 and 0.69 (mgKOH/g) respectively (Table 2). These results are in agreement with the findings of Pearson (2002), who reported an acid value range of 0.25-1.75 for sunflower oil. AV of oil samples falls below the maximum recommended value of olive oil (17 mg KOH/g) and groundnut (4 mg KOH/ g) (Codex 2008).

These results indicate the possibility of classifying this oil as edible oil, as the AV determination is often used as a general indication of the condition and edibility of the oil. This is because an increase in acid value is accompanied by development of objectionable flavours and odours (Nelson and Cox 2005).

Peroxide value (PV)

PV of DLH, NDLH and MDLH oil samples were 5.66, 3.90 and 4.28 (m.Eq/kg) respectively (Table 2). The PV of oil samples under investigation didn't agree with the finding of Robertson and Russell (1972), who reported that sunflower oil PV range was between 0.21 and 0.41m.Eq\Kg. The PV of both DLH and NDLH oil samples under investigation fell below the maximum limit of safe edible oils (10 m.Eq/Kg) as reported by Codex (2008). The low PV gives more stability to the oil and reduces the risk of rancidity, according to AOAC (2008), which indicates that the high peroxide value means high degree of unsaturation, which is in turn responsible for oxidative rancidity.

Heavy metals

The heavy metals in both of DLH, NDLH and MDLH oil samples *i.e.* Iron, copper, lead and arsenic are shown in Table 2. In DLH they were 21.12, 0.34, 0.000 and 0.0081(mg/g), respectively. While they were in NDLH oil sample 21.12, 0.33, 0.00 and 0.01(mg/g), respectively. Also, in MDLH they were 24.47, 0.28, 0.04 and 0.11 (mg/g), respectively. Iron reading in oil samples exceeded the permissible level of Codex (2008) (5 mg/kg), while copper, lead and arsenic were lower than the maximum values recommended by Codex (2008).

Polymer content

Polymers' content of single cell oil from wheat straw and molasses samples are shown in Table 2. Polymers' content of DLH, NDLH and MDLH oil samples are 1.90, 1.98 and 1.73 (%), respectively.

Fatty acids composition of DLH and NDLH single cell oil

The fatty acids composition data of produced SCO are shown in Table 3. DHL and NDHL oil samples were low in their saturated fatty acids composition, while unsaturated ones were higher. The data show low content of acetic, butyric, valeric, caproic and lauric acids as 2.33, 0.73, 0.60, 0.28 and 0.09 (%), respectively, for DLH oil sample, and 2.27, 0.87, 0.40, 1.67 and 0.09 (%), respectively for NDLH oil sample. There are no significant differences between DLH and NDLH oil samples in acetic,

butyric, valeric and lauric acids content. These results are not in agreement with Codex (2008) for fatty acids composition of sunflower and sesame oil where butyric, valeric, caproic and lauric acids were not detected. The values of myristic and stearic were 3.60 and 2.04 (%) for DLH oil samples and 2.60 and 2.25 (%) for NDLH oil samples, respectively. There were no significant differences between DLH and NDLH oil samples in palmitic, stearic and arachidic acids content. Caproic acid showed significant difference (($P \leq 0.05$) among oil samples. Myristic content of both DLH and NDLH (3.60 and 2.60, respectively) was higher than that set by the Codex (2008) for ground nut oil (< 0.1). The values of palmitic acid were 9.40 and 10.95 (%) for DLH and NDLH oil samples, respectively. Oil samples contain palmitic acid as the highest content of saturated fatty acids (Table 3). These results fall within the range of standard value (8.3-14.0 %) for groundnut and sesame oil 7.9-12.0 of Codex (2008). Data presented in Table 3 show that oleic acid content of the two types of oil samples were significantly different, Linoleic acid contents of the oil samples were 19.44 and 21.83 (%) for DLH and NDLH oil samples, respectively, indicating significant difference. These readings fall within the range of standard value of Codex (2008) standard for oleic acid (14.0-39.4 %) and linoleic acid (48.3-74.0 %) in sunflower oil. γ -linolenic acid contents of the oil samples were 3.54 and 2.8 (%) for DLH and NDLH oil samples, respectively.

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Table 3. Composition of fatty acids of extracted SCO produced using wheat straw

| Fatty acid (%) | DLH | NDLH |
|----------------------------------|------------------------------|------------------------------|
| a) Saturated fatty acid | | |
| Acetic | 2.33 ^a ±0.23 | 2.27 ^a ±1.01 |
| Butyric | 0.73 ^a ±0.23 | 0.87 ^a ±0.23 |
| Valeric | 0.60 ^a ±0.40 | 0.40 ^a ±0.23 |
| Caproic | 0.28 ^b ±0.00 | 1.67 ^a ±0.61 |
| Lauric | 0.093 ^a ±0.14 | 0.093 ^a ±0.05 |
| Myristic | 3.60 ^a ±0.69 | 2.60 ^b ±0.69 |
| Palmitic | 9.40 ^a ±1.06 | 10.95 ^a ±0.13 |
| Stearic | 2.04 ^a ±1.07 | 2.25 ^a ±0.05 |
| Arachidic | 0.09 ^a ±0.02 | 0.08 ^a ±0.04 |
| b) Unsaturated fatty acid | | |
| Vinylglycolic | 8.49 ^a ±0.19 | 8.46 ^a ±0.19 |
| Palmitoleic | 10.32 ^a ±0.09 | 10.41 ^a ±0.04 |
| Oleic | 32.80 ^a ±0.08 | 29.54 ^b ±0.10 |
| Linoleic | 19.44 ^b ±0.46 | 21.83 ^a ±0.58 |
| γ-linolenic | 3.54 ^a ±0.05 | 2.83 ^b ±0.12 |
| α-linolenic | 5.55^b±0.09 | 5.81^a±0.12 |

Values are means ±SD

Values bearing different superscripts in rows are significantly different at (P≤0.05) according to DMRT.

α-linolenic acid contents of produced SCO were higher than Codex (2008) standard for sunflower (0-0.3 %) and groundnut oil (0.1%) and sesame oil (0.3-0.4 %). It is observed that DLH and NDLH oil samples gave higher values of oleic acid than linoleic acid. In addition, a highly significant difference (P≤0.01) is observed among oil samples (Table 3). Fatty acids profile of produced oil is in agreement with other reported (Xin and Yang 2009 and Xiaochen *et al* 2009).

Fatty acids composition of molasses' SCO

The fatty acids composition data of produced SCO is shown in Table 4. Myristic content of oil sample (3.60 %) was higher than that set by the

Codex (2008) for ground nut oil (0.1). The content of palmitic acid was (6.80 %), this result fell below the standard value (8.3-14.0 %) for groundnut oil and sesame oil (7.9-12.0 %). Stearic acid content of oil sample was 4.90 %, and plamitoleic (7.07 %) was higher than the standard Codex (2008) content of sesame oil (0.1-0.2 %) and sunflower (0.0-0.3 %). Content of oleic acid was (16.60 %). These results are not in agreement with Husain *et al.* (2010), who used *Cryptococcus curvatus* for the production of SCO with a low cost cultivation medium containing beet molasses and corn gluten meal as carbon and nitrogen sources. α -linolenic acid content of the oil sample was (2.00 %), it was higher than the standard of Codex (2008) content of sunflower (0.00-0.30 %). It was observed that the produced SCO on molasses contain omega-6 fatty acids such as Eicosapentaenoic (0.51 %) and Docosahxaenoic (0.67 %), which were not present in vegetable sources as reported by Kankaanpaa *et al.* (2001). However, the fatty acids profile of produced oil by oleaginous yeast on using molasses as substrate was different from fatty acid profile of vegetable oil.

Table 4. Composition of fatty acids of extracted SCO using molasses as substrate

| Saturated fatty acid | % | Unsaturated fatty acid | % |
|----------------------|-----------------|------------------------|------------------|
| Acetic | 5.60 \pm 0.80 | Vinylglycolic acid | 10.80 \pm 0.69 |
| Butyric | 2.93 \pm 0.23 | Palmitoleic | 7.07 \pm 0.61 |
| Valeric | 7.07 \pm 0.61 | Oleic | 16.60 \pm 0.00 |
| Caproic | 5.47 \pm 0.61 | γ -linolenic | 19.27 \pm 0.46 |
| Lauric | 2.80 \pm 0.69 | α -linolenic | 2.00 \pm 0.40 |
| Myristic | 3.60 \pm 0.00 | Eicosapentaenoic | 0.51 \pm 0.26 |
| Palmitic | 6.80 \pm 0.80 | Docosahxaenoic | 0.67 \pm 0.23 |
| Stearic | 4.93 \pm 0.23 | -- | -- |

Values are means \pm SD

CONCLUSION

It can be concluded from this study that:

1. Oleaginous yeasts isolated from rotten fruits, juice, milk, fish and air, were positive for *Sacchromyces cerevisiae* and *Pichia guilliermondii*
2. The molasses medium is found to be more effective for promotion of the accumulation of substantial amount of lipids by *Sacchromyces cerevisiae*.
3. The productivity of SCO produced by *Sacchromyces cerevisiae* is different according to the sources of isolation.
4. The physicochemical characteristics and fatty acid profile of SCO from wheat straw make it potential raw material as edible oil.
5. The result indicates that molasses oil yield is high with good physicochemical properties and rich in polyunsaturated omega-6 fatty acid such as Eicosapentaenoic and Docosahexaenoic.
6. Generally, it may be concluded that SCO production from lignocellulosic matter and by product is a new source for edible oil.

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الخصائص الفيزيوكيميائية لزيت الخليه الواحدة المنتج من الخمائر الزيتية

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المستخلص: هدفت هذه الدراسة الى استخلاص زيت الخليه الواحدة من الخمائر الزيتية عند نموها في وسط قش القمح والمولاس و دراسة الخصائص الكيميائية الطبيعية لزيت المنتج لتقديره كزيت طعام . تم عزل ثلاثة عينات وفقا لطرق مختلفة للعزل بناء على مصدر العزل. فعالية هذه الخمائر الزيتية تمت دراستها عند نموها على قش القمح (سوائل مسمومة وغير مسمومة) والمولاس. واستخلص زيت الخليه الواحدة بالهكسان. استخدمت خميرة *Pichia* و *Sacchromyces cerevisiae* و *guilliermondii* لانتاج زيت الخليه الواحدة. أنتجت الخمائر الزيتية نسب عالية من الزيت عند إستخدام المولاس مقارنة مع استخدام قش القمح. أعطي قش القمح (السائل غير المسموم) انتاجية أعلى لزيت من قش القمح (السائل المسموم).

الخميرة *Sacchromyces cerevisiae* أعطت إنتاجية عالية لزيت 84٪ مقارنة مع *P. guilliermondii* والتي كانت 52٪. أشارت النتائج إلى أن الخواص الفيزيوكيميائية لزيوت المستخلصات كانت ضمن الحد الموصى به لمعظم الزيوت الصالحة للأكل التي حدتها الكودكس (2003)، عدا تركيز الحديد والذي ترواحت كميته بين 21 و 24 ملجم/كجم، حيث تجاوزت أربعة إلى خمسة أضعاف الحد المسموح به من الكودكس (2003). بالإضافة إلى أن الزيت المستخلص من المولاس تحتوي على أحماض دهنية أوميغا-6 مثل أيكوساتنويك ودووكوساهكسنويك والتي لا توجد في الزيوت النباتية. ايضا الأحماض الدهنية غير المشبعة لزيت الخليه الواحدة المنتج في وسط قش القمح كان اعلى من تلك المنتجه في وسط المولاس. يختلف تركيب

الاحماس الدهنية للزيت المنتج من الخمائر الزيتية عند استخدام المولاس عن تركيب الاحماس الدهنية للزيوت النباتية. الخصائص الكيميائية الطبيعية الجيدة وتركيب الاحماس الدهنية لزيت الخليه الواحد من قش القمح جعلته آمن وواعد في إمكانية استخدامه كمادة خام لتصنيع زيت الطعام. تشير نتائج هذه الدراسة الى امكانية عزل الخمائر الزيتية لاستخدامها في انتاج زيت الطعام.