

Effect of Anaerobic Fermentation Temperature on Three Different Types of Low-Grade Saudi Dates for Producing Ethanol Fuel

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ABSTRACT

The present work aims to study the effect of anaerobic fermentation temperature on the production of ethanol fuel for three different types of sugar compositions in dates that commonly produced in Saudi Arabia. The fermentation temperature is considered as the most important factors affecting the production of ethanol. The obtained data referred that the ethanol yields from Sukkari dates were 107.6 cm³ L⁻¹ juice, 123.3 cm³ L⁻¹ juice, and 96.6 cm³ L⁻¹ juice, during anaerobic fermentation temperatures of 30°C, 35°C, and 40°C, respectively. Whilst, the ethanol yields from Khllass dates were 111.0 cm³ L⁻¹ juice, 122.0 cm³ L⁻¹ juice, and 128.0 cm³ L⁻¹ juice during the same fermentation temperatures. Ultimately, the Berhi dates produced 121.0 cm³ L⁻¹ juice, 136.0 cm³ L⁻¹ juice, and 136.0 cm³ L⁻¹ juice, under the same fermentation temperatures. The obtained results showed that the production of ethanol would directly proportional to the ratio of glucose and fructose composition in dates.

Keywords: Dates, Fermentation, Sugar, glucose and fructose

INTRODUCTION

Ethanol is a natural solvent combined with water and is called "Bioethanol" after its production through a fermentation procedure utilizing microorganisms. Transformation of sugars into ethanol is one of the most common anaerobic natural responses utilized by humankind (Benarji and Ayyanna, 2016). Ethanol can be made either artificially from petrochemical crude materials or naturally by the fermentation of sugar. When pure ethanol is produced from glucose or normal source by fermentation using distillation, the outcome is bioethanol. Bioethanol can be described as an inexhaustible and pure fuel for vehicles. However, it is typically utilized as a gasoline additive to raise the value of octane and enhance vehicle discharges.

Ethanol/bioethanol is an alcoholic transparent fluid that can be utilized as a raw material for derivatives of alcohol, in the synthetic industry, paint base industry, pharmaceutical industry, a blend of fuel for vehicles, that given the ethanol/bioethanol as a source of renewable energy to reduce the environmental pollution that occur from internal combustion engines. A 90-96% can be utilized in business and the industry, while ethanol having 96-99.5% can be utilized as a blend for fundamental modern materials and pharmaceuticals (Ghanim, 2013). Nowadays, the whole world countries have been taken the bioethanol as the most utilized biofuel instead of the fossil fuel. It is commonly created from sugar containing different farming productions eg., (sugar cane, corn, wheat, sugar beet, squander from sugar cane, or sweet sorghum). The dominating innovation for changing biomass to ethanol is the fermentation as a biochemical innovation (Prasad *et al.*, 2007).

Bioethanol is broadly investigated as a sustainable fuel source because of the numerous contemplations it is better than gasoline. Ethanol gives vitality of unlimited and less carbon in the gravity than oil (Jones *et al.*, 1994 ; Fakruddin *et al.*, 2013). Ethanol is an important chemical with excellent potential as a biofuel to replace petroleum derivatives (Ghassem *et al.*, 2012). Generation of ethanol from inexhaustible starchy materials has been drawing worldwide interest and research has been conducted for the creation of ethanol by immobilized microorganisms, through the utilization of consistent cultures (Goksungur, 2001). It is an essential natural compound utilized as a solvent in research work, businesses, and family units. Naturally, the

utilization of ethanol as a substitute fuel has acquired much attention as an answer to some issues caused by gasoline from exhaustible oil reserves. It is created from different substrates, such as saccharides, starches, and cellulose materials. Accumulation of appropriate and commercially available substrates is an imperative cost consideration for the manufacture of ethanol (Hagerdal *et al.*, 2006; Pinheiro *et al.*, 2008; Srinivasarao *et al.*, 2013).

Among biofuels, ethanol has extraordinary demand as it is widely acknowledged and it is ideal consumer. In many nations, ethanol is either utilized as a substitute fuel or mixed with petroleum or gasoline. Numerous specialists have contemplated on the generation of ethanol utilizing different raw materials. Organic product wastes such as papaya, mangoes (Reddy and Reddy, 2007), banana peels (Joshi, 2001), pineapple (Muttamara *et al.*, 1982), and grapes (Pramanik and Kinetic, 2005; Asli, 2010) were utilized for the manufacture of ethanol (Raikar, 2012). Technically, ethanol can be produced from a wide assortment of sustainable feedstock, which can be generally characterized into three primary categories; Firstly, those containing excessive amounts of promptly fermentable sugars (sweet sorghum, sugar beets, and sugar cane), secondly, starches and fructosans (corn, potatoes, rice, wheat, and agave), and thirdly, cellulosics (Stover, grasses, corncobs, wood, sugar reeds bagasse). Sugar reeds, beet, and sweet sorghum preserve the simple sugars such as sucrose, glucose, and fructose that can be promptly matured by yeasts (Amorim *et al.*, 2009; Basso *et al.*, 2011). Yeast and other similar microorganisms are normally utilized for the production of ethanol. Usually *saccharomyces cerevisiae* strains are utilized for the generation of ethanol.

Temperature is one of the most vital elements that influence ethanol generation by yeast utilizing molasses as a carbon source. The fermentation procedure is constantly accompanied with the development of warmth that raises the temperature of the fermenter (Jones *et al.*, 1994). Ethanol maturation at upgraded temperatures is a principle need for successful ethanol generation in tropical districts where ordinary daytime temperatures are commonly high everywhere throughout the year. The rewards of fast fermentation at optimal level of temperature were not only the danger of pollution regardless likewise diminishes the cooling costs. To accomplish upgraded temperature aging it is necessary to utilize a suitable yeast strain that can endure high temperature (Limtong *et al.*, 2007). For the bioethanol creation at higher temperatures, yeast cells die resulting in reduction in ethanol yield when the substrate is

concentrated, while the optimum temperature giving the best yield is 32°C for the most extreme strains. Therefore, it is important to choose the optimal level of temperature for anaerobic fermentation (Edgardo *et al.*, 2008; Yah *et al.*, 2010). Considering the strains for temperature variations, there is a need for thermo-tolerant strains that can withstand high temperatures in the experiments. For instance, K. Marxian's DMKU3-1042 is considered to have an ideal temperature of 40°C (Abdel-Banat, 2010). Moreover, since at 45°C no development was seen and no maturation was observed, it was inferred that *S. cerevisiae* have a range from 30–35 °C, which is known to be ideal for fermentation (Bollók *et al.*, 2000; Lin and Tanaka, 2006). Hence, temperature plays an important role. The dependence of the maximum ethanol production rate on the temperature was clarified by the superposition of initiation vitality for ethanol generation (Sánchez *et al.*, 2004; Gorsek and Zajsek, 2010). Thus, at an optimal level of temperature to the yeast acquires a higher ethanol production.

Dates, as a natural product, are a reasonable source for bioethanol production. They contain impressive amounts of transformed sugars (glucose and fructose); the two sugars are available in dates in a practically comparable amount. The fresh of dates contains around 70 to 75% sugars (Elleuch *et al.*, 2008). Low-grade dates demonstrated a similar sugar as dates of high grade (Besbes *et al.*, 2009). Maturation of sugars is an anaerobic natural process in which sugars are converted into ethanol by the activity of microorganisms, such as yeast (Demirbas, 2007). *Saccharomyces cerevisiae* is the most prominent modern microorganism utilized for sugar fermentation to create bioethanol, because it uses simple materials for growth and generation. This organism has already been accepted as a non-pathogenic, safe product that can easily be genetically manipulated and on simple and inexpensive media as compared with animal cell culture (Vrsalovic and Vasic-Racki, 2005). Low-quality dates are rich in sugars, mostly glucose and fructose, which can be converted into ethanol and can likewise source of basic carbon for yeast development. Different supplements, minerals, and vitamins are additionally present in dates, which enhance the maturation procedure to create bioethanol. Many factor, such as temperature and starting sugar fixations, can influence the entire procedure. The pH within a suitable range appeared to have minor effect on bioethanol generation. Ethanol yields >71% were obtained in controlled and uncontrolled pH tests. Ethanol yields of 91.3%, 68.7%, and 54.8% were obtained from 10%, 15%, and 20% starting sugar fixations, respectively. The decrease in ethanol production in the higher sugar ratios could be due to the Inhibition of sugar for ethanol (Suliman *et al.*, 2013).

Sucrose is broken down using fermentation and 14 enzymes with the water to two types of simple sugars (glucose and fructose). The glucose and fructose converted to ethanol by anaerobic fermentation can be represented by the following equation (Williamson *et al.*, 2007). The Sukkari dates contain a total sugar content of 67.42%, which can be divided into four types of fructose 19.8%, glucose 14.96%, sucrose 29.9%, and maltose 3.57%. The Khlass dates contain a total sugar content of 62.2%, divided into four types of fructose 34.97% and glucose 31.94% sucrose 0.1% and maltose 0.1% (Bhat and Sapna, 2003; Nanda, *et al.*, 2003; Ouchemoukh *et al.*, 2010; Hassan *et al.*, 2014; Elamshity, 2014). The Berhi dates contain a total sugar content of 57.6%, divided into two types of fructose 27.6% and glucose 29.7% (Ahmed *et al.*, 1995). This study aims to

investigate the effect of anaerobic fermentation temperature on the production of ethanol fuel from three different types of sugar compositions in dates that commonly produced in Saudi Arabia.

MATERIALS AND METHODS

Materials

- 1 - Three different types of Saudi date residues (Sukkari, Khlass, and Berhi) from the King Saud University Farm in Drab were utilized during this research work. These residues are commonly used as animal feeds. The moisture content of date residues is from 10 to 12% (w.b.).
- 2 - Nine glass fermenters were used with a fermented capacity of 2.5 L, containing a stirrer of stainless steel.
- 3 - Three similar water basins of stainless steel, each one comprising three glass fermenters. The water basin was function for heating the fermenters according to the desired level of fermentation temperature as shown in Fig (1).
- 4 - Device (ABBE 5 Refractometer, Bellingham & Stanley Ltd., Tun-bridge Wells UK) for measuring sugar contained.
- 5 - The bakery yeast (*Saccharomyces cerevisiae* yeast) was used during the fermentation experiments.

Total soluble solids (TSS)

Total soluble solids content in all treatments were determined before and after the fermentation processes using an Atago digital refractometer (Tokyo, Japan) which has a scale ranged between 0 and 30% Brix unit.

Sugar extraction of dates (syrup)

The sugars were extracted from the dates using distilled water at a rate of 1: 2.5 (w/v) of total dry dates, at 40 °C for four hours to prepare the syrup necessary for testing. The fiber and lingering material were removed using a set of filters to obtain pure syrup. The sugar content of the syrup was estimated to be approximately 23%, 23%, and 20 % in Sukkari, Khlass, and Berhi, respectively. The required amount of water to adjust the total sugar in the solution to be 20 % was calculated using the following formula (Liu and Chen, 1981):

$$Y = X \left[\frac{T_{s1} - T_{s2}}{T_{s1}} \right]$$

Where: Y = dilution volume (liter)

X = amount of raw material added (kg)

Ts1 = total solid of raw material (sugar in solution)

Ts2 = total solid of fermentation material.

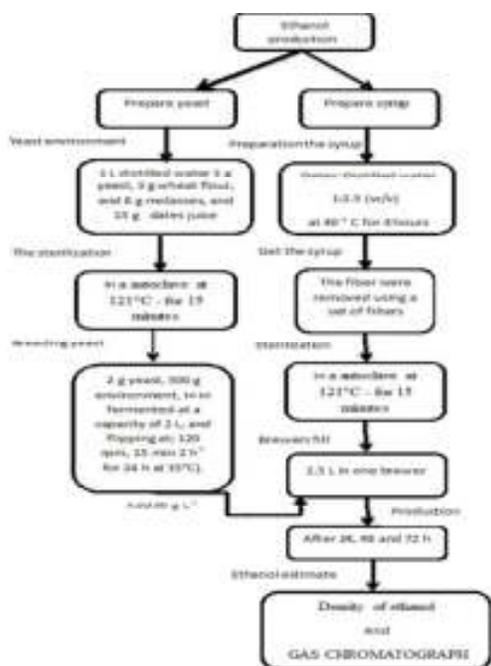
Preparation of yeast environment,

Fast-fermentation yeast which commonly used in bakeries (Wild strain *Saccharomyces cerevisiae* yeast, STAR brand), wheat flour, molasses has a concentration of 70%, and dates juice has 20% sugar, (3 g yeast, 3 g wheat flour, and 8 g molasses, and 15 g dates juice) were used for each liter of distilled water. The sterilization of the fermentation environment was carried out in an autoclave at temperature of 121°C for 15 minutes. Two grams of fast-fermentation yeast (*Saccharomyces Cerevisiae*) was added to 500 g of the fermentation environment, and were poured in fermented at a capacity of 2 -liter (anaerobic fermentation with flipping at; 120 rpm, 15 min. each 2 h for 24 h at 35°C).

Methods

Fermentation process

The experiments were executed by adding 50 ml of the activated yeast environment per liter of juice and placing it in a glass fermenter with 2.5-liter fermentation capacity. The glass fermenters were placed in a water basin to control the fermentation temperatures under study (30, 35 and 40°C).



(a)

(b)

Fig. 1. (a): Diagram of ethanol production process and (b) image of brewers

Estimation of Ethanol

Ethanol concentration was separated using two methods:

- 1- The density of ethanol is 0.78 g.cm^{-3} at 100% concentration and temperature of 25°C , while it was 0.8 g.cm^{-3} at 96% concentration and temperature of 25°C
- 2 - Using a device estimating the concentration of ethanol (GAS CHROMATOGRAPH – GC – 2025 – SHIMADZU – Japan)

Statistical analysis:

Data was analyzed for multiple comparisons by analysis of variance with least significant differences (LSD) between means at 5% significance level using SAS 9.2 software (SAS Institute, Cary, NC).

RESULTS AND DISCUSSION

The most important requirements for the anaerobic fermentation process were the yeast *Saccharomyces* used to produce ethanol, the presence of carbon, nitrogen, and basic minerals. Dates are an important source of all these

elements along the high sugar content. The fermentation temperature is one of the most important factors affecting production of ethanol from dates.

Effect of temperature on ethanol production

Ethanol production from the fermentation of three types of dates (Sukkari, Khlass, and Berhi) was measured at different fermentation times (24, 48, and 72 hours). The concentration of sugar was 20%, at three levels different levels of fermentation temperatures 30°C , 35°C , and 40°C . Table (1) lists the average ratio of ethanol production under different temperature levels. Figs (2), (3), and (4) refer the effect of fermentation temperatures on ethanol production. Fig (2) shows the increase in ethanol production with high temperature under 24h fermentation time. The ethanol production of dates (Sukkari) was on an average (v/v) 5.33%, 5.96%, and 6.4%, at 30, 35, and 40°C , respectively, whilst, it was on an average for the dates (Khlass) (v/v) 5.46 %, 6.6 % and 6.7% at the same temperature levels.

Table 1. Average ethanol production of three different species of dates under three different levels of fermentation temperatures

Ethanol%, (v/v)	Sukkari		Ethanol%, (v/v)	Khlass		Ethanol%, (v/v)	Berhi	
	¹ Concentrate of sugar	² Sugar ratio%		Concentrate of sugar	Sugar ratio%		Concentrate of sugar	Sugar ratio%
0 ^{Cdg}	20 ^{Aac}	0 ^{Cdg}	20 ^{Aaf}	0 ^{Cdf}	20 ^{Aag}	0 ^{Cde}	20 ^{Abg}	0 ^{Cde}
5.33 ^{Ccg}	16.6 ^{Abe}	17 ^{Ccg}	5.46 ^{Ccf}	16.3 ^{Abf}	18.1 ^{Ccf}	6.7 ^{Cce}	16.5 ^{Abg}	17.5 ^{Cce}
8.6 ^{Cbg}	14.36 ^{Ace}	28.2 ^{Cbg}	8.83 ^{Cbi}	14.14 ^{Act}	29.3 ^{Cbi}	10.7 ^{Cbe}	14 ^{Acg}	30 ^{Cbe}
10.76 ^{Cag}	13.7 ^{Ade}	31.5 ^{Cag}	11.1 ^{Ca}	12.6 ^{Adt}	36.8 ^{Cat}	12.1 ^{Ca}	12.8 ^{Adg}	36 ^{Ca}
0 ^{Adg}	20 ^{Cac}	0 ^{Adg}	0 ^{Adf}	20 ^{Caf}	0 ^{Adi}	0 ^{Ade}	20 ^{Cag}	0 ^{Ade}
5.96 ^{Acg}	15.66 ^{Cbe}	21.7 ^{Acg}	6.6 ^{Act}	15.6 ^{Cbt}	21.8 ^{Act}	6.8 ^{Ace}	15 ^{Cbg}	25 ^{Ace}
9.26 ^{Abg}	13.76 ^{Cce}	31.2 ^{Abg}	9.3 ^{Abf}	13.8 ^{Ccf}	30.7 ^{Abf}	13.4 ^{Abe}	12 ^{Ccg}	40 ^{Abe}
12.33 ^{Aag}	12.03 ^{Cde}	39.85 ^{Aag}	12.2 ^{Aaf}	11.9 ^{Cdf}	40.3 ^{Aaf}	13.6 ^{Aae}	10.5 ^{Cdg}	47.5 ^{Aae}
0 ^{Bdg}	20 ^{Bac}	0 ^{Bdg}	0 ^{Bdi}	20 ^{Bdf}	0 ^{Bai}	0 ^{Bde}	20 ^{Bag}	0 ^{Bde}
6.4 ^{Bcg}	15.5 ^{Bbe}	22 ^{Bcg}	6.7 ^{Bcf}	15.5 ^{Bbf}	22.3 ^{Bcf}	6.9 ^{Bce}	15 ^{Bbg}	25 ^{Bce}
8.23 ^{Bbg}	14.56 ^{Bce}	27.2 ^{Bbg}	9.85 ^{Bbf}	13.5 ^{Bcf}	32.5 ^{Bbf}	13.6 ^{Bbe}	13.4 ^{Bcg}	33 ^{Bbe}
9.66 ^{Bag}	14.13 ^{Bde}	29.35 ^{Bag}	12.8 ^{Bat}	11.5 ^{Bat}	42.3 ^{Bdt}	13.6 ^{Bae}	11.3 ^{Bdg}	43.5 ^{Bae}

Same upper letter (A,B and C) in column means no significant difference in temperature treatment , same lower letter (a,b,c and d) in column means no significant difference in time treatment and same lower letter (e,f and g) in raw means no significant difference in type of dates treatment. ¹Concentration of sugar in the dates juice during the fermentation process. ²The percentage of sugars converted during fermentations is attributed, to the ratio of primary sugars in date juice

The ethanol production of dates (Berhi) was (v/v) 6.7%, 6.8% and 6.9% at 30, 35, and 40°C, respectively. The ethanol production under fermentation time of 48 h is shown in Fig (3). It is referred that, the ethanol production of dates (Sukkari) was (v/v), 8.6% 9.26% and 8.23 % at 30, 35 and 40°C, respectively. While, the ethanol production of dates (Khlass) was 8.83% (v/v), 9.3% (v/v), and 9.85% (v/v), at the same temperature levels of fermentation. The average ethanol production for dates (Berhi) was (v/v) 10.70 %, 13.40 %, and 13.60 % at 30, 35, and 40°C, respectively. Fig (4) shows the ethanol production under fermentation time of 72 h. Under this circumstances the ethanol production of dates (Sukkari) was on an average (v/v) 10.%, 12.33%, and 9.66% at fermentation times of 30, 35, and 40°C, respectively, while, the ethanol production of dates (Khlass) was on an average (v/v) 11.13%, 12.2% and 12.8% at the same temperature levels. But it was for the dates (Berhi) on an average (v/v) 12.10%, 13.60% and 13.60 % at the same levels of fermentation temperature.

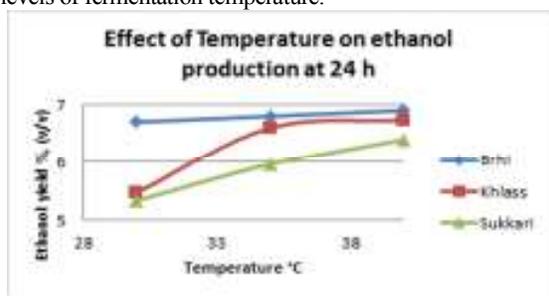


Fig. 2. Ethanol production for three different species of dates at three different fermentation temperatures and fermentation time of 24h.

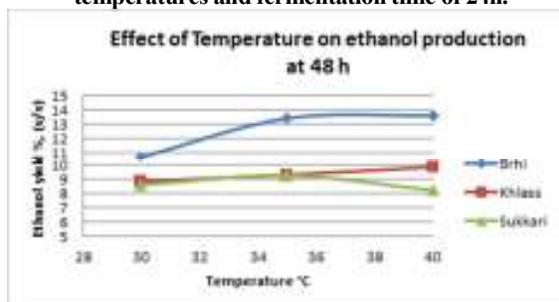


Fig 3. Ethanol production for three different species of dates at three different fermentation temperatures and fermentation time of 48h.

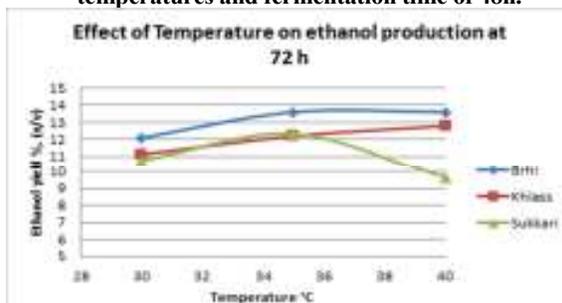


Fig 4. Ethanol production for three different species of dates at three different fermentation temperatures and fermentation time of 72h.

The obtained results referred that increasing the

fermentation times from 24h to 72h resulting in increasing the rate of ethanol production of Sukkari at fermentation temperature of 35°C by 55.37% and 106.88% as compared with 48h and 72h of fermentation time, respectively. The effect of the fermentation time on the production of ethanol at three different fermentation temperatures is shown in Figs (5), (6), (7). The ethanol production is usually increased with increasing the hydraulic retention time under all fermentation temperatures. The highest of ethanol production was achieved at hydraulic retention time (HRT) 72h and fermentation temperature of 35°C for Sukkari and Berhi, whilst it was achieved at 40°C for Khlass.

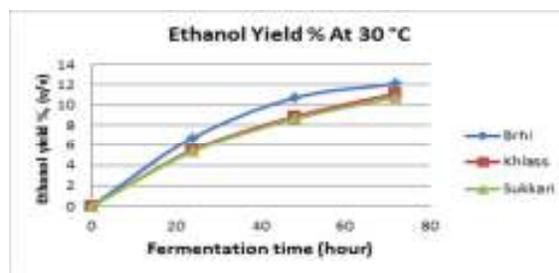


Fig 5. Ethanol production under different fermentation times and fermentation temperature of 30°C.

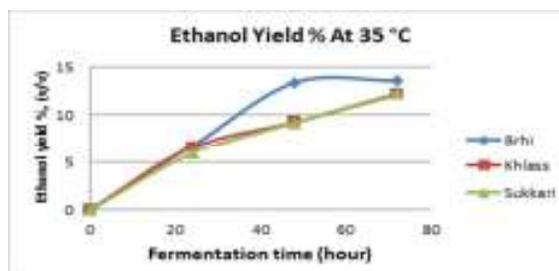


Fig 6. Ethanol production under different fermentation times and fermentation temperature of 35°C

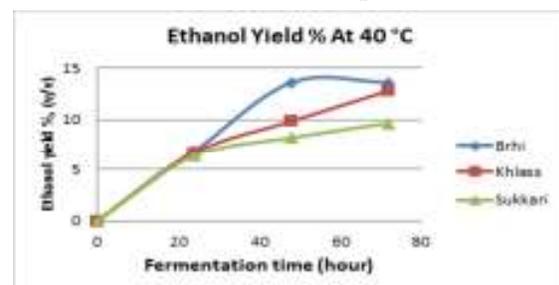


Fig 7. Ethanol production under different fermentation times and fermentation temperature of 40°C.

Effect of fermentation temperature on sugar consumption

Sugar consumption for the three different types of dates (Sukkari, Khlass, and Berhi) was measured at different fermentation times (24, 48, and 72 hours). The concentration of sugars was 20%, for the three different fermentation temperatures (30, 35, and 40°C). The effect of fermentation temperature on sugar consumptions are plotted in Figs (8), (9), and (10). Fig (8) refers an increase in sugar consumption with high level of temperature during the first fermentation time (24 h). For the duration of this experiment, the average sugar consumption of dates (Sukkari) was 17%, 21.7 %, and 22.3 % of total sugars at fermentation temperature of 30, 35, and 40°C, respectively. While, the average sugar consumption of dates (Khlass) was 18.1%,

21.8%, and 22.3% of total sugar at the same temperature levels, respectively. The sugar consumptions of dates (Berhi) was 17.5%, 25%, and 25% of total sugar at 30, 35, and 40°C, respectively. The sugar Consumptions for the fermentation time of 48 hours is shown in Fig (9). During this experiment the average sugar consumption of dates (Sukkari) was 28.2%, 31.2%, and 27.2% of the total sugar under three different fermentation temperatures of 30, 35 and, 40°C. While, the average sugar consumption of dates (Khllass) was 23.9%, 30.7%, and 32.5 % of the total sugar under the same temperature levels, respectively. The average sugar consumption of dates (Berhi) was 30%, 40%, and 33% of the total sugar under three different fermentation temperature levels, respectively. Fig (10) shows sugar consumption at fermentation time of 72 hours. The average sugar consumption for dates (Sukkari) was 31.5%, 39.85%, and 29.35% of the total sugar at three different fermentation temperature levels, respectively. While, the average sugar consumption for dates (Khllass) was 36.8%, 40.3%, and 42.3% of the total sugar at the same fermentation temperature levels, respectively. The average sugar consumption for dates (Berhi) was 36%, 47.5 %, and 43.5 % of the total sugar under the same fermentation temperature levels, respectively.

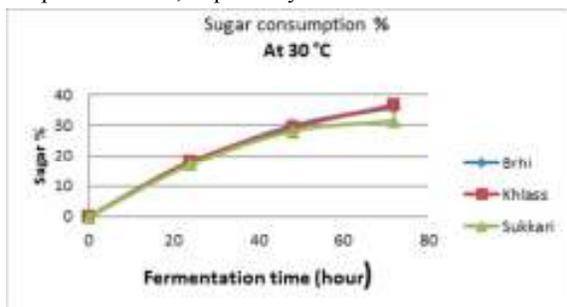


Fig 8. Sugar consumption versus fermentation time under fermentation temperature of 30°C.

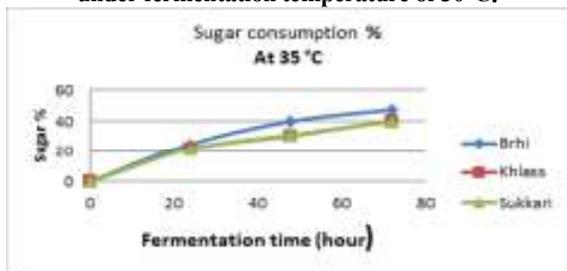


Fig 9. Sugar consumption versus hydraulic retention time under fermentation temperature of 35°C.

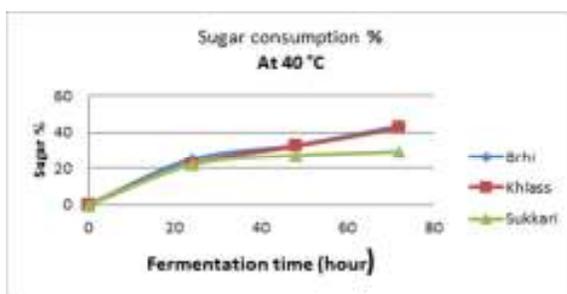


Fig 10. Sugar consumption versus hydraulic retention time under fermentation temperature of 40 °C.

The effect of fermentation times on the sugar

consumption at different temperatures is shown in Figs (11), (12), and (13). They clearly referred that the sugar consumptions increase as the fermentation time increases under all fermentation temperatures. They also referred that, the highest rate of sugar consumptions was achieved during the fermentation time of 72 hours at fermentation temperatures of 35°C for Sukarri dates, 40°C for Khllass and Berhi.

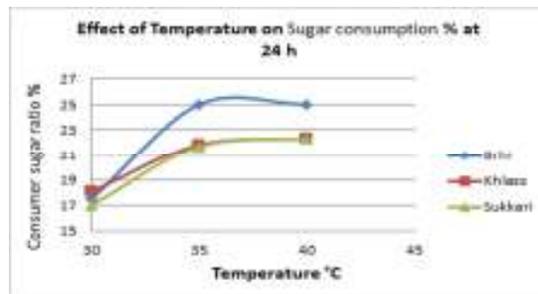


Fig 11. Effect of fermentation temperature on sugar consumption with fermentation time of 24 h.

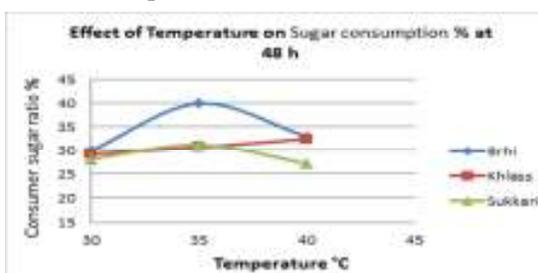


Fig 12. Effect of fermentation temperature on sugar consumption with fermentation time of 48 h.

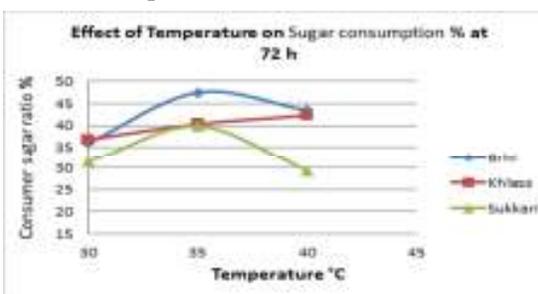


Fig 13. Effect of fermentation temperature on sugar consumption with fermentation time of 72 h.

Effect of fermentation temperature on ethanol production

The productivity of ethanol fuel ($\text{cm}^3 \text{kg}^{-1}$ dates) is summarized and listed in Table (2) and plotted in Fig (14). The productivity of ethanol for the three different dates species (Sukkari, Khllass, and Berhi) at fermentation temperature of 30°C and fermentation time of 24-hour was on an average 153.3, 157.16, and 167.5 $\text{cm}^3 \text{kg}^{-1}$ dates, respectively. While, the productivity of ethanol at fermentation temperature of 30°C and fermentation time of 48-hour was 247.2, 253.9, and 267.5 $\text{cm}^3 \text{kg}^{-1}$ dates, respectively. Finally, the productivity of ethanol at fermentation temperature of 30°C and fermentation time of 72-hour was 309.4, 320.1, and 302.5 $\text{cm}^3 \text{kg}^{-1}$ dates, respectively. On the other hand, the productivity of ethanol for the three different dates species (Sukkari, Khllass, and Berhi) at fermentation temperature of 35°C and fermentation

time of 24-hour was on an average 171.4, 189.75, and 170 $\text{cm}^3 \text{kg}^{-1}$ dates, respectively. While, the productivity of ethanol at fermentation temperature of 35°C and fermentation time of 48-hour was 266.5, 267.4, and 336 $\text{cm}^3 \text{kg}^{-1}$ dates, respectively. Finally, the productivity of ethanol at fermentation temperature of 35°C and fermentation time of 72-hour was 354.5, 350.8, and 340 $\text{cm}^3 \text{kg}^{-1}$ dates, respectively.

Table 2. Productivity of ethanol fuel ($\text{cm}^3 \text{kg}^{-1}$ date) form three different species of dates..

Temperature, °C	Time, h	Sukkari	Khlass	Berhi
30	0	0.0	0.0	0.0
	24	153.2	157.2	167.5
	48	247.3	254.0	267.5
	72	309.4	320.1	302.5
35	0	0.0	0.0	0.0
	24	171.4	189.8	170.0
	48	266.2	267.4	335.0
	72	354.5	350.8	340.0
40	0	0.0	0.0	0.0
	24	184.0	193.6	172.5
	48	236.6	283.2	340.0
	72	277.7	368.0	340.0

From the previous results, the heating process of dates juice during an anaerobic fermentation resulting in increasing the productivity of ethanol fuel from species; Sukkari by 5.33% and 12.33%, from Khlass dates by 5.5% and 12.8% and from Berhi dates by 6.7% and 13.6% due to change in fermentation time and fermentation temperatures. The obtained results also referred that the productivity of ethanol is directly proportional to the glucose and fructose in raw material. These data are in agreement with the data published by (Ahmed *et al.*, 1995; Bhat and Sapna, 2003; Nanda *et al.*, 2003; Williamson *et al.*, 2007; Ouchemoukh *et al.*, 2010; Sulieman *et al.*, 2013; Elamshity, 2014; Hassan *et al.*, 2014).

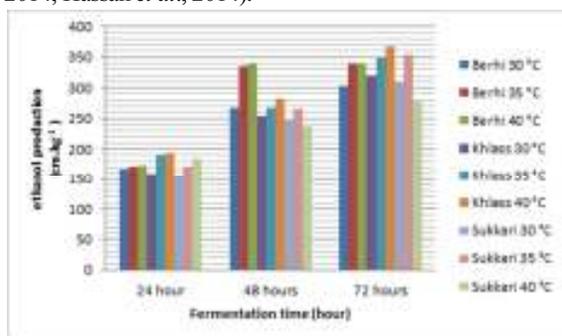


Fig 14. Productivity of ethanol ($\text{cm}^3 \text{kg}^{-1}$ date) at three different fermentation temperatures and fermentation times.

CONCLUSION

Sugars in Low-grade dates are mainly based on glucose and fructose, which can be converted into ethanol and can also serve as the main sources of carbohydrate for yeast growth. Other nutrients, minerals and vitamins are also presented in the dates components that promote fermentation to produce bioethanol. The most important factors affecting ethanol production are associated with; the initial sugar concentrations, fermentation temperatures and fermentation time. The ethanol yield greater than 300 cm^3 ethanol per one kg of dates was achieved during the experimental work. The productivity of ethanol (v/v) at three different fermentation temperatures (30, 35 and 40°C) from juice of Sukkari date, respectively,

was 10.76, 12.33 and 9.66 %. While the productivity of ethanol (v/v) from juice of Khlass at the same fermentation temperatures was 11.1, 12.2 and 12.8 %, respectively. Whilst, the productivity of ethanol (v/v) from juice of Berhi at the same fermentation temperatures was 12.1, 13.6 and 13.6%, respectively. Further experimental work are needed for more studying on the technology of bioethanol production using different sugars ratio and other types of yeasts that may enhance the ethanol revenues.

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REFERENCES

- Abdel-Banat, B.M.; Hoshida, H. A.; Ano, S. N, and Akada, R. (2010) "High-temperature fermentation: how can processes for ethanol production at high temperatures become superior to the traditional process using mesophilic yeast?" Appl. Microb. Biotech., 85 (4): 861 – 867.
- Ahmed, I. A.; Ahmed, A. W. K.; and Robinson, R. K. (1995) "Chemical composition of date varieties influenced by the stage of ripening, Food Chem., 54, 305 – 309.
- Amorim, H.; Basso, L.; and Lopes, M. (2009) "Sugar cane juice and molasses, beet molasses and sweet sorghum: composition and usage" The Alcohol Textbook, 5th Edition. W.M. Ingledew, G.D. Austin, C. Kluhsbies and D.R. Kelsall, Eds., Nottingham University Press: Nottingham, p. 39 – 46.
- Asli, M. S. (2010) "A study on some efficient parameters in batch fermentation of ethanol using *Saccharomyces cerevisiae* SC1 extracted from fermented siahe sardasht pomace" African J. Biotech., 9(20).
- Basso, L.C.; Basso, T.O.; and Rocha, S. N. (2011) "Ethanol production in Brazil: the industrial process and its impact on yeast fermentation" Biofuel production—recent developments and prospects. Rijeka, 85 – 100.
- Benarji, D. S. N. and Ayyanna, C. (2016) "Statistical Optimizations of Fermentation Factors on Bioethanol Production from Mahua Flower (*Madhuca indica*) with *Saccharomyces cerevisiae* by Response Surface Methodology in Batch Bioreactor" Microbioz Jouranls, J. Microbiol. Biomed. Res. 2 (1)
- Besbes, S.; Drira, L.; Blecher, C.; Deronne, C.; and Attia, H. (2009) "Adding value to hard date (*Phoenixdactylifera* L.): Compositional, functional and sensory characteristics of date jam" Food Chem., 112: 406 – 411.
- Bhat, K. K. and Sapna V. K. (2003) "Sensory properties, physical and chemical samples of commercial honey" Food Research International, Sensory Science Department, Central Food Technological Research Institute, Mysore-570 013, India, 36 (2): 183 – 191
- Bollók, M.; Réczey, K.; and Zacchi, G. (2000) "Simultaneous saccharification and fermentation of steam-pretreated spruce to ethanol" in Twenty-First Symposium on Biotechnology for Fuels and Chemicals, Springer
- Demirbas, A. (2007) "Progress and recent trends in biofuels" Progress in Energy and Combustion Sciences, 33 (1): 1 – 18.
- Edgardo, A. P.; Carolina, R.; Manuel, F. J.; and Baeza, J. (2008) "Selection of thermos-tolerant yeast strains < I > *Saccharomyces cerevisiae*< /I > for bioethanol production" Enzyme Microblog. Technology, 43 (2): 120 – 123.
- Elamshity, M. G. K. (2014) "Development of a Nutritional Drink from Cow's and Camel's Milk with Date Syrup" Department of Agricultural Engineering, Faculty of Food and Agricultural Sciences, King Saud University, Riyadh, Saudi Arabia.

- Elleuch, M.; Besbes, S.; Roiseux, O.; Blecher, C.; Deroanne, C.; Dirar, N.; and Attia, H. (2008) "Date flesh: Chemical composition and characteristics of the dietary fiber, Food Chem... 111: 676 – 682.
- Fakruddin, M. A.; Ahmed, M. M.; and Chowdhury, N. (2013) "Process optimization of bioethanol production by stress tolerant yeasts isolated from agro-industrial waste" Int. J. Renew. Sustain. Energy, 2(4)
- Ghanim, A. N. (2013) "Bioethanol Production from Iraqi Date Palm Resources" J. Babylon University/ Eng. Sci. 1 (21): 13 – 20
- Ghassem, T.; Sadat, D. A.; and Kulkarni, D. (2012) "Optimization of yeast for ethanol production" Int. J. Res. Ayurveda Pharm, 3(1): 95 – 97.
- Goksungur, Y.A. Z. N. (2001) "Production of Ethanol from Beet Molasses by Ca-Alginate Immobilized Yeast Cells in a Packed-Bed Bioreactor, Turk J. Biol, 265 – 275.
- Gorsek, A. and Zajsek, K. (2010) "Influence of temperature variations on ethanol production by kefir grains-mathematical model development" Chemical Engineering 20
- Hagerdal, H. B.; Galbe, M.; M. F.; Gorwa-Grauslund, G. L.; and Zacchi, G. (2006) "Bio-ethanol-the fuel of tomorrow from the residues of today" Trends Biotechnology, 24 (12): 549 – 556.
- Hassan, B. H.; Mohammed, A. A.; Khaled A.; and Ahmed, M. (2014) "Moisture sorption isotherms and hot air drying characteristics of Saudi dates (Phoenix dactylifera L.) and the effect of drying on dates color and mechanical properties" Project report, National Plan for Science and Technology. King Saud university project #10-AGR, 1194 – 1202.
- Jones, A.M.; Thomas, K.C.; and Ingledew, W.M. (1994) "Ethanol fermentation of blackstrap molasses and sugarcane juice using very high gravity technology" J. Agric. Food Chem. 42(5): 1242 – 1246
- Joshi, S.; Dhopeswarkar, R.; Jadhav, R.; Jadhav, U.; D'Souza, L.; and Dixit, J. (2001) "Continuous ethanol production by fermentation of waste banana peels using flocculating yeast" Indian J. Chem. Technol. 8 (3): 153 – 156.
- Limtong, S.; Sringiew, C.; and Yongmanitchai, W. (2007) "Production of fuel ethanol at high temperature from sugar cane juice by a newly isolated *Kluyveromyces marxianus*" Bio-resource Technol. 98 (17): 3367 – 3374.
- Lin, Y. and Tanaka, S. (2006) "Ethanol fermentation from biomass resources: current state and prospects" Appl. Microb. Biotech., 69 (6): 627 – 642.
- Liu, K.X. and Chen, C.Q. (1981) "Studies on the biogas fermentation of Chinese rural area" Global Impact of Applied Microbiology (GIAMVI). Sixth Int. Conf., 261- 272.
- Muttamara, S.; Nirmala, D.; Stuckey, D.C.; and Hamza, A. (1982) "Production of alcohol and acetic acid from pineapple wastes. in International Symposium on Management of Industrial Wastewater in Developing Countries" Pergamon Press
- Nanda, V.; Sarkara, B. C.; Sharma, H. K.; and Bawab, A.S. (2003) "Physico-chemical properties and estimation of mineral content in honey produced from different plants in Northern India" J. Food Comp. Anal. 16: 613 – 619.
- Ouchemoukh, S.; Schweitzer, P.; Bey, M. B.; Djoudad-Kadji, H.; and Louaileche, H. (2010) "HPLC sugar profiles of Algerian honeys" Food Chem. 121: 561 – 568
- Pinheiro, A. M.; Rocha, G. M.; and Gonçalves, L.B. (2008) "Evaluation of Cashew Apple Juice for the Production of Fuel Ethanol" Appl. Biochem. Biotech., 148: 227 – 234
- Pramanik, K. A. R. and Kinetic, D. E. (2005) "Study of ethanol fermentation of grape waste using *Saccharomyces cerevisiae* yeast isolated from toddy" J. of Inst. Eng., 85: 53 – 58.
- Prasad, S.; Anoop Singh, N. J.; and Joshi, H. C. (2007) "Ethanol Production from Sweet Sorghum Syrup for Utilization as Automotive Fuel in India" Division of Environmental Science, Indian Agriculture Research Institute, New Delhi-110012, India, April 4.
- Raikar, R. V. (2012) "Enhanced production of ethanol from grape waste" Int. J. Env. Sci., 3(2)
- Reddy, L. V. and Reddy, O. (2007) "Production of Ethanol from Mango (*Mangifera indica* L.) Fruit Juice Fermentation" Res. J. Microbiol., 2 (10).
- Sánchez, S.; Bravo, V.; Moya, A.; Castro, E.; and Camacho, F. (2004) "Influence of temperature on the fermentation of d-xylose by *Pachysolen tannophilus* to produce ethanol and xylitol" Process Biochem., 39 (6): 673 – 679.
- Srinivasarao, B.; Ratnam, B.; Subbarao, S.; Narasimharao, M.; and Ayyanna, C. (2013) "Ethanol production from cashew apple juice using statistical designs" J. Biochem. Microb. Technol., 18 – 15
- Suliman, A. K.; Gaily, M. H.; Zeinelabdeen, M. A.; Putra, M. D.; and Abasaeed, A. E. (June 2013) "Production of Bioethanol Fuel from Low-Grade-Date Extract" International Journal of Chemical Engineering Application, 4 (3)
- Vrsalovic, P. A. and Vasic-Racki, D. (2005) "Modeling of the alcohol dehydrogenase production in baker's yeast" Process Biochemistry, 40 (8): 2781 – 2791.
- Williamson, K. L.; Minard, R.; and Masters, K. M. (2007) "Macroscale and Microscale Organic Experiments" Houghton Mifflin Co., Boston, 5th ed., p 774.
- Yah, C.S.; Iyuke, S.E.; Unuabonah, E.I.; Pillay, O.; Vishanta, C.; and Tessa, S. M. (2010) "Temperature optimization for bioethanol production from corn cobs using mixed yeast strains" Online, J. Biolog. Sci., 10 (2): 103 – 112

تأثير درجة حرارة التخمر اللاهوائي على ثلاثة أنواع مختلفة من التمر منخفضة الجودة لإنتاج وقود الإيثانول

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يهدف هذا البحث إلى دراسة تأثير درجة حرارة التخمر اللاهوائي على إنتاج وقود الإيثانول من ثلاثة أنواع مختلفة من التمر ذات تركيبات مختلفة من السكر في التمر الذي تنتج عادة في مصر والمملكة العربية السعودية. وتعتبر درجة حرارة التخمر أهم العوامل المؤثرة على إنتاج الإيثانول وأشارت أهم النتائج التي تم الحصول عليها إلى أن متوسط إنتاج الإيثانول من تمر (السكري) كان 107.6، 123.3 و 96.6 سم³/ لتر عصير تمر، خلال درجات حرارة التخمر اللاهوائي 30 درجة مئوية، 35 درجة مئوية، و 40 درجة مئوية ، على التوالي. في حين أن متوسط إنتاج الإيثانول من تمر (الخلاص) 111.0، 122.0، و 128.0 سم³/ لتر أثناء نفس درجات حرارة التخمر على التوالي. في النهاية أنتجت تمر (البرحي) 121.0، 136.0، و 136.0 سم³/ لتر عصير، تحت نفس درجات حرارة التخمر. وأظهرت النتائج التي تم الحصول عليها أن إنتاج الإيثانول يتناسب طردياً مع نسبة الجلوكوز وتكوين الفركتوز في التمر وأفضل درجة للتخمر كانت 35 درجة مئوية. كما أظهرت النتائج إمكانية الاستفادة من متبقيات التمر في إنتاج الإيثانول كصناعة تحويلية للاستفادة منه في المجال الطبي أو كوقود بديل.