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# Utilization of Low Marketing Potential Libyan Dry Dates Cultivar "Tasfert" to Table Vinegar

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**ABSTRACT**

"Tasfert" is a common dry cultivars grown in the southern region of Libya, its characterized by; large fruit size, high sugar content, fruit firmness because of low moisture content caused by climate conditions and handling practices. This study investigated the potential of utilizing "Tasfert" cull dates to table vinegar. A fermentation process was carried out using a laboratory scale anaerobic fermenting setup equipped with an airlock. Whole dates were mixed with distilled water at 1:4 w/w ratio and commercial baker's yeast (Saccharomyces cerevisiae) was added at 2g /kg of dates. It was incubated at room temperature (winter time) averaging 22ºC, CO2 was released through the airlock, and the process was ended when CO2 bubbling stopped. Afterwards, juice was extracted and its properties were determined at; total soluble solids (TSS) was 11.3 (°Brix), Titratable Acidity (TA) was 1.3 v/v, pH was 4.3, and ethanol content was 5.1% v/v. Collected juice was aerobically acetified, whereas virgin date vinegar was added at 10% v/v, and incubated for 5 weeks at room temperature. TA was periodically measured throughout the aerobic reaction, and its stability was considered as a sign of completing acetic fermentation, then Residual Alcohol (RA) content was measured. The reaction was represented by second order polynomial equation. Final product properties were at; TSS 8.5 Brix, TA 5.09% v/v, pH 3.02, and RA 0.042% v/v. Such properties were in a good agreement with the Libyan vinegar standards. The study demonstrated the potential for producing good table vinegar from cull "Tasfert" dates.

## INTRODUCTION

Date palm (*Phoenix dactylifera* L.) is the most important tree to countries located in dry and semidry regions, including all Arab countries of the Middle East and North Africa. The world production of dates is estimated at 7 million metric tons and about 100million trees are grown worldwide (Zaid, 2002). Libya attains the tenth place among the world top date producers, it produces about 170 thousand metric tons/year, nearly ten million trees are cultivated, and most of its agricultural lands are suitable for date cultivation. About 400 varieties exist, yet only 95 cultivars are considered commercially important (Kader and Hussein, 2009; Racchi *et al.*, 2014). Libya has large area extending roughly between latitudes 18° and 33° North and 9° and 25° East, exceeding 1.75 million $km^2$ (Al-Idrissi *et al.*, 1996). Therefore, the three date cultivar groups exist; soft dates are grown in the coastal area, semidry dates are exist in the middle (Jufra region), while dry cultivars are grown in the southern oases of Fezzan region (Fennir *et al.*, 2014).

Dry cultivars are grown in the most undeveloped region, wherein harvesting, handling and storage practices are still very much traditional. Also, because of high temperature and dry weather in Fezzan region, dates are susceptible to over drying, reaching unmarketable conditions (can be classified as cull), and therefore becoming less appealing. It is worth mentioning that consumers in the northern populated region prefer dates rather soft (at Rutab stage). High losses in quantities and market value are generally common, however over dried dates are used in limited small scale date syrup ("Debis") making operations, and as animal feed. Exact figure of losses in dates is quite undetermined, yet it can be considered significantly high.

"Tasfert" is an important dry cultivar in Fezzan region, it is in fact one of the main date varieties in Libya (Mostafa and Ahmed, 1981). It is characterized by large fruit size and high reducing sugars that responsible for making dates firm at dry stage ("Tamr"). It has been reported that total sugar content in "Tasfert" dates about 67.2%, whereas less than 1.5% non-reducing sugars, also fruit moisture contents were reported at less than 13% in the three main Fezzan "Tasfert" cultivation areas (Al-Shurfa *et al.*, 1978).

In general, at the postharvest level, date production in Libya faces several difficulties; such as high losses due to traditional handling and storage practices, absence of proper processing and cull utilization, in addition to some socio-economical constrains.

Although dates at low quality are considered unmarketable, yet they are high in sugars, giving good potential for conversion to several valuable products with good marketing value, such as date syrup, date paste, liquid sugar, baker's yeast, bioalcohols (C2H5OH) and vinegar (CH3COOH) (Barreveld, 1993, Aleid, 2011). Historically, vinegar has been used almost in all cultures since its fermentation process was discovered. Natural vinegar is generally produced from bioethanol that is produced by microbial reactions using mostly agricultural products containing sugars and starch (Tan, 2003). While starch needs to be hydrolyzed to fermentable sugars, fruit sugars are directly converted to bioethanol (Lin and Tanaka, (2006), Onuki *at al.*, (2008). The fermentation process is mostly carried out by yeasts such as *Saccharomyces sp* under anaerobic conditions (Borah and Mishra, 2011, Simay *et al.*, 2013). For producing vinegar however, fermenting sugars takes place in a slow process similar to what is known as Orleans process, followed by an acetic fermentation process (oxidation) for converting ethanol to acetic acid using *Acetobacter aceti* bacteria. The process takes place in aerobic conditions, and adding non heated vinegar stock known as mother vinegar is a common practice.

Date vinegar is a popular food additive classified as condiment with several nutritional and health benefits (Matloob, 2014). Its main benefits in food industry related to retarding microbial activities and contributing to flavouring. Despite the importance of vinegar and the abundance of dates in their producing countries, very limited studies have dealt with date vinegar issues. Iraqi cultivars "Zahdi" and "Khastawi" were investigated for producing vinegar and its characteristics were reported by (Matloob and Hamza, 2013, Matloob, 2014). Also, making vinegar from Omani dates and investigating its aromatic compounds have been reported (AlShoaily, 2014). Traditional date vinegar preparation in Algerian Sahara community was investigated by Halladj *et al.*, 2016 using spontaneous alcoholic and acetic fermentation in one batch setup. In Libya however, date vinegar is produced traditionally in small scale homemade operations carried out by date farmers, mainly for their own uses and no commercial activities exist. In addition, no published studies addressing date vinegar production in Libya have been found in the literature. This work investigated applying a simple semi-spontaneous fermentation setup for producing vinegar from cull and over dried 'Tasfert' Libyan dry date cultivar.

## MATERIALS AND METHODS

### Dates

Dates cultivar 'Tasfert' were collected from the public market of Zintan town at about 145km south west of the capital Tripoli, sold in bulk, and packed in Jute bag of about 50kg weight. Dates were brought to the market in a non-cooled truck, and they were sold as low quality dates that commonly used for date syrup (Bebis) and to some extent used in homemade date paste that used in cookie making. Samples were transferred to the Postharvest Laboratory (PL) at the Department of Agricultural Engineering, Faculty of Agriculture, University of Tripoli. They were sorted, infested and undesirable fruits were removed and selected dates were kept in a walk-in cold room until experimental procedures were carried out.

### Anaerobic fermentation setup and process

The fermentation setup was built at the laboratory consisted of a 7L bottle equipped with an airtight lid. For securing airtightness, a brass tire valve equipped with threaded base was installed on the lid. The valve was connected to an airlock *via* flexible plastic tube installed on a threaded female connection that fitted on the valve from one end and submerged in water filled container from the other. Airtightness and anaerobic conditions were ensured by periodical measuring air contents inside the fermenter using portable gas analyzer (Model CANAL120 $O_2$ & $CO_2$ Gas Analyzer, EMCO Packaging Systems Ltd, Kent, CT14 0BD, UK). The instrument withdraws small air volume, analyzes its $CO_2$ and $O_2$ percentages and displays results on an LSD screen. The simplicity of the setup facilitated replication and applying multi treatments.

It worth saying that in this study whole dates were used for the reason of acquiring similarity with traditional vinegar methods and conditions similar to that reported by Halladj *et al.*, 2016. However, unlike traditional methods that are normally carried out in autumn time wherein room temperature is around 30°C, the current study was carried out at temperature averaging 22°C.

Dates were weighed, washed and dried using hot air, and whole dates were placed in the fermenter. Distilled water was added at 1:4 ratio (1.25kg of dates mixed with 5 litres of water) and commercial baker's yeast was used as a fermenting starter (2g of yeast per kg of dates), mixed with warm distilled water and sugar, stirred and incubated for one hour and

added to the soaked dates. The fermenter was closed, incubated at room temperature averaging 22°C with periodical inspection. The anaerobic reaction was ended when no $CO_2$ bubbling in the air lock was observed.

After the anaerobic fermentation was ended, fermenter contents were removed and the liquid part was extracted and filtered using a mechanical press.

**Properties of the anaerobic fermentation yield**

The collected juice was measured in volume and its Total Soluble Solid (TSS) was determined using digital handheld refractometer (Model PAL-α, ATAGO Co, Ltd, Tokyo, Japan). Acidity was also determined using standard titration method, 1.5ml of the juice was withdrawn and distilled water was added making 20ml solution, phenolphthalein as indicator was added, mixed and titrated with 0.1 Sodium Hydroxide. Acidity was determined and expressed as percentage volume (v/v). The pH was also measured using a pH meter (Inco Lab, WTW GmbH, Germany), all measurements were carried out in triplicates.

**Gas chromatography system**

A gas chromatography Model SRI 8610C, Pre-configured as a multiple gas analyzer configuration #3 (SRI instruments, Torrance, CA 90503-2162, USA) was used for measuring ethanol content. The GC is equipped with 2m HaysepD and 2m Molecular Sieve (MS 13X) columns, and the two columns were installed in series. The GC is also equipped with Valco valve system, facilitating directing sample to either or both columns via selectable event time settings and two valves (A and G) that timely activated by on-off software command. Also, it is equipped with Thermal Conductivity Detector (TCD) and Flame Ionized Detector (FID) detectors. It is operated by Helium as carrier gas and Hydrogen for igniting FID detector, also equipped with internal air compressor. The system is operated by PeakSimple software (ver. 3.93) installed on a Laptop computer. The software facilitates controlling oven temperature, sampling events, temperature program, chromatogram, eluted peaks labelling, calibration procedures using standards and components identifications.

**Calibration and measurement of ethanol**

Because of lack of information in the literature regarding the use of the above GC configuration for measuring ethanol, several combinations of hardware and software settings

were tested prior to carrying out the measurements. The best hardware settings were: Helium pressure 20psi (18ml.min$^{-1}$), Hydrogen pressure 20psi (20ml.min$^{-1}$), air flow pressure 8psi (250ml.min$^{-1}$), FID temperature 380°C. Software settings were: oven temperature at 180°C held for 2 minutes, followed by temperature increase (ramp) at 50°C.minute$^{-1}$ until 220°C, and holding at 220°C for 4 minutes.

Ethanol at high purity (99.9%) was obtained from the Libyan Authority of Food and Drug Inspection of Tripoli and used in the calibration process. First, Ethanol was diluted with distilled water, 2µL was injected in the GC *via* injection port using a 10µL syringe, ethanol peak was eluted using above mentioned GC setting at 220°C oven temperature and retention time starting at 5.6minutes. Afterwards, a calibration curve was developed using multiple dilutions of ethanol injection at 0.25, 0.5, 1.0 and 2% v/v, peak areas were recorded, concentrations and their peak areas were entered in PeakSimple software calibration protocol. At subsequent analyses, the software executed the calibration equation, and sample ethanol consentient were given as percentage (v/v) in a result window.

**Vinegar samples analysis**

Samples were withdrawn from the extracted juice at the end of the anaerobic fermentation and during the acidifying processes. In GC analysis, sample filtration and purification are generally of great importance, especially for fermented materials (Gerchman *et al.*, 2012). Therefore, special care was given to sample filtering. Samples were filtered twice using filter paper followed by micro filtering using Whitman 0.45µm syringe filter. A 2 µL sample volume was injected in the GC *via* the injection port. After every injection, the syringe was washed several times with distilled water. Ethanol contents in vinegar were measured right after the acetic fermentation, after three weeks (as a check), and at its end. All analyses were made in triplicates.

**RESULTS AND DISCUSSIONS**

**Ethanol standardization**

Figure 1 shows ethanol calibration curve. The calibration procedures were carried out according to the SRI instruments system supporting printed materials using (99.9%) ethanol as an external standard. Ethanol peak eluted starting in 5.6 minutes at 220°C oven temperature. Figure 2 shows peak eluted from injecting 2% ethanol concentration. As can be

noticed, peak has a normal shape. Peak area in relation to injected concentrations was linear and peaks were doubled in area as the injected standard concentration doubled. The relation was presented by a linear model with $R^2$ = 0.999. This was considered as a good sign for the sensitivity of the analyses. Comparative results for all injected samples within the calibration range exhibited standard deviation of (±0.04). This mainly attributed to the use of the same temperature setup, sample volume (2µL per injection) and enough time were given for warming up the GC. For the calibration curve obtained, four data points with four doubled known concentrations were used. This is in good agreements with standard calibration procedures recommended by (Mealy and Johnson, 1993), (Gowrisankar *et al.*, 2010). Therefore, the derived calibration was used with confidence for giving acceptable results in measuring ethanol content in vinegar samples.

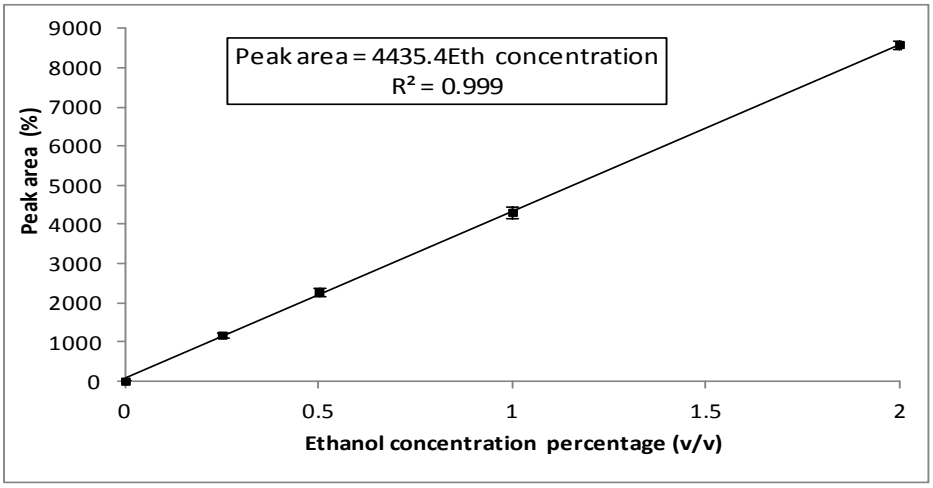


**Figure 1.** Ethanol calibration curve

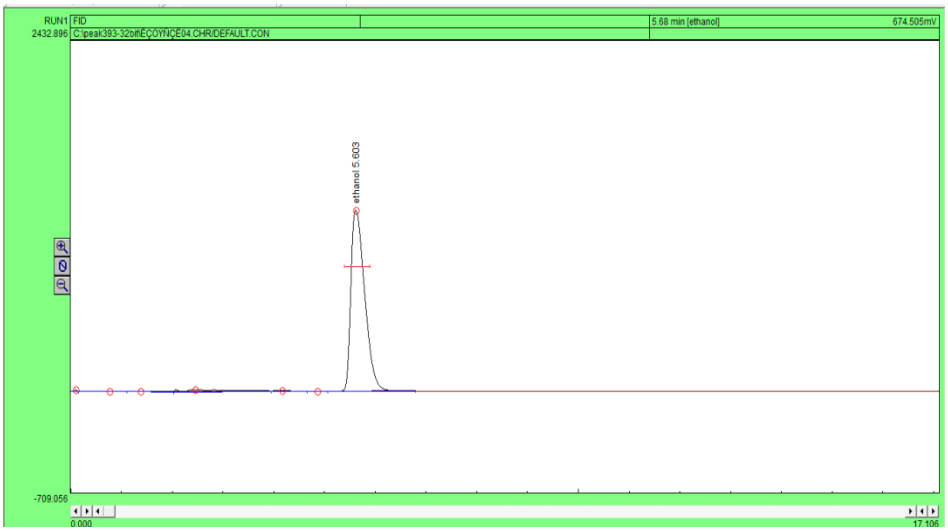


**Figure 2.** Ethanol eluted peak

Performance of anaerobic fermentation was monitored by inspection of released $CO_2$. The process lasted for two weeks at room temperature averaging 22°C. $CO_2$ production rate started at high rates for few days, followed by steady decline until it reached its end after 2 weeks. Such long time was quite anticipated due to relatively low temperature applied. Indeed, fermentation process at higher temperatures would remarkably shorten time by several days. However, in this study, room temperature was chosen for the purpose of convenience, and to bring the fermentation process close to traditional vinegar making that is normally made at room temperature. Also, in such fermentation using (date water mix), there was time taken for soaking dates and extracting their sugar content. However, the anaerobic fermentation led to digestion of dates, they formed a thick broth, some fruits settled down and others remained suspended. Once $CO_2$ release was halted, the juice was filtered and its TSS, TA, and ethanol contents were measured. Table 1 shows TSS and ethanol contents, TA and pH after the anaerobic fermentation and at the end of the acetifying process.

Although the initial TSS was not measured for the date-water mix at the start of the fermentation process, it was considered as high enough to produce good ethanol yield from such ratio. This was due to performing several preliminary trails prior to the current study. Nonetheless, based on the sugar contents of "Tasfet" cultivar reported by (Al-Shurfa *et al.*, 1978) at 67.4%, theoretically, the date-water mix (1:4) in this study contained about 170g.$L^{-1}$ of sugars. Considering solubility efficiency involved in the process, a 15% Brix is a quite fair estimate. However, after anaerobic fermentation, TSS mean value was 11. 3 (±0.11) Brix, and ethanol content was 5.1% (±0.08622), TA was 1.3% (±0.056) v/v and pH was 4.31 (±0.036). Thus, TSS remained high even after the anaerobic fermentation was completed. This may be attributed to sugar digestion efficiency by yeast at low temperature and the existence of other soluble compounds. Moreover, (Matloob and Hamza, 2013) reported sugar conversion efficiency of 53%, thus the current study can be considered in a good agreement with the literature. Moreover, in the present study, some sugar remained unfermented, giving TSS of 8.5% in vinegar. However, taking fermentation conditions and cultivar into account, again this is in a fair agreement with results reported by (Matloob and Hamza, 2013), they reported sugar content in their vinegar at 6.4%.

TA was determined right after the anaerobic fermentation at 1.3% v/v. However, acidity was used along the acetifying process as an indication of its progression. A TA of 1.3% after anaerobic fermentation might be due to some acetifying occurred in the process, those could

be not necessarily citric acid. In analyses of vinegar samples from several materials (Natera *et al.*, 2003) reported finding other organic acids (tartaric, malic, succinic) in several vinegar samples analyzed for the purpose of qualifying some aromatic and quality related compounds. However, the aim of this work was not performing qualitative evaluation of the fermentation rather investigating its potential.

Figure 3 shows performance of the acetic fermentation process within 35 days. The relation was fitted to a second order polynomial equation with an $R^2$ value of 0.967. It gave highest acidity in day 32 of the process (5.35%), followed by decline to (5.00%) in subsequent days. Similar behaviour was reported by Matloob and Hamza, 2013 in producing vinegar from Iraqi dates cultivar "Khastawi", also similar trend was reported by Matloob, 2014 in using another Iraqi date cultivar Zahdi. This gives the current study fair agreements with both studies.

Collected juice using the mechanical press was at an efficiency of 90%, and after acetifying the efficiency was 95%, giving an overall yield efficiency of 86%. This indicated that 1.25kg of low quality "Tasfert" dates gave 4.32L of vinegar with characteristics shown in Table 1. The Libyan standard (LNS 823: 2015) defined natural (bio) vinegar as: *"A product produced from double fermentation of sugary or starchy products. It must contain at least 4g acetic acid per 100ml plus other constituents, such as sugars, plant remains that sustain aroma, and salt. Free from any materials that are not from its source. Ethanol content must not exceed 1.0% for grape vinegar and 0.5% for others.TSS must be not less that 1.3g.L$^{-1}$ per 1% v/v acetic acid".*

It is quite evident that vinegar produced in this study meets the Libyan standard of table vinegar. Thus, the study demonstrated the process and its potential for producing quality vinegar from dates that unmarketable for table consumption. Further investigation using other cultivars and improving current methodology is needed.

**Table 1**. Properties of the date juice after anaerobic fermentation and acidification (means ± standard deviations)

| | Yield & Efficiency | TSS | Ethanol (%) | Acidity (%) | pH |
|---|---|---|---|---|---|
| Anaerobic | 4.56L (±0.01) (90%) | 11. 3 (± 0.11) | 5.105 (± 0.086) | 1.30 (±0.15) | 4.34 (± 0.01) |
| Vinegar | 4.32L (±0.02) (95%) | 8.5 (±0.076) | 0.052 (±0.027) | 5.00 (±0.06) | 3.89 (±0.08) |

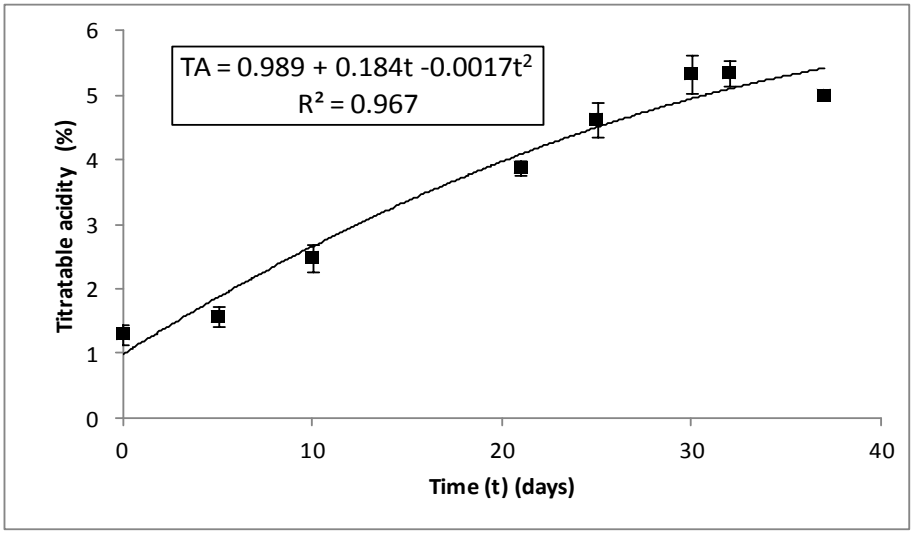


**Figure 3.** Acidifying process progression

## CONCLUSIONS

Libyan dry date cultivar "Tasfert" at low marketing quality was investigated for producing table vinegar. The investigation showed that both aerobic and anaerobic processes lasted for seven weeks at room temperature (averaging 22°C). The work showed that date to water ratio used was 1:4, date to water ratio gave 4.3L of table vinegar with 5% acidity (titratable), 8.5% TSS, 0.05% ethanol and 3.9 pH. The produced vinegar met the Libyan standards for natural and industrial vinegar (LNS 823-2015).

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