

Phytochemical Screening of Methanolic Extract of Five Libyan Date Varieties (*Phoenix Dactylifera* L.) And Evaluation of Their Antimicrobial Activity

Nahla Labyad ^{a*}, Basma Doro^b, Fadia Gafri^c, SafaElamaari ^a, Namarq Almusrati^a,

^aDepartment of Pharmacognosy, Faculty of Pharmacy, University of Tripoli, Tripoli, Libya

^bDepartment of Microbiology and Immunology , Faculty of pharmacy,
University of Tripoli, Tripoli, Libya

^cDepartment of Pharmacology and Clinical Pharmacy , Faculty of Pharmacy,
University of Tripoli, Tripoli, Libya



Abstract – More than 2000 cultivars of *Phoenix dactylifera* L. are known to grow around the world. Date is an essential fruit in North Africa, and in Libya in particular, it represents an important national food resource. Recently, researchers discovered the medicinal value of different parts of palm tree beside the well known nutritional value of the fruit edible part. In this study five Libyan date seeds (Bestian, Deglet, Abel, Khadrai and Hamrai) were collected from Aljofra region and were screened for phytochemical components and their antimicrobial activity. The phytochemical analysis of methanolic extract of five different Libyan date palm seeds showed the presence of aflavonoids, glycosides, phytosterols, phenolic compound, tannins, and fixed oil, absences of alkaloids and saponine. The susceptibility of bacterial species to five methanolic extracts of *Phoenix dactylifera* L seeds by cup-cut agar method exhibited Inhibition zones (IZD). That inhibition zone diameters are vary from 15.0 ± 0.3 to 3.02 ± 0.01 against *MRSA*; 16.04 ± 0.1 to 6.0 ± 0.7 against *Pseudomonas aeruginosa*; 14.2 ± 0.1 to 6.0 ± 0.7 against *Proteus vulgaris*; 18.3 ± 0.2 to 6.0 ± 0.1 against *Staphylococcus aureus*; 14.04 ± 0.2 to 2.0 ± 0.02 against *E. coli*; and 18.0 ± 0.1 to 3.2 ± 0.02 against *Klebsiella pneumonia*. The results of current study shows the antibacterial properties of five Libyan samples of date seed extract against a wide spectrum of bacteria.

Keywords – *Phoenix Dactylifera*; Libya; Seeds, Antimicrobial.

I. INTRODUCTION

The *Phoenix dactylifera* L. (date palm) belongs to the Palmae (Arecaceae) family is known as one of the oldest tree cultivated by a human for its edible fruit [1, 2]. Millions of people across the world are using date palm for its nutritional value in several countries. Lately, the health promoting properties of different parts of the palm tree (fruit, leaves, seeds and pollen grains) have been recognized for many medicinal applications [3].

In last decades, researchers have **worked** on identification and isolation of phytochemical constituents of date palm and

investigated their therapeutic value [4-6]. Most of the identified biologically active compounds were relatively similar in different palm tree parts. Of these dominant constituents; phenolic acids (e.g: galic acid, cinnamic acid , ferulic acid [7] and syringic acid) and flavonoid glycosides (e.g: quercetin, luteolin, apigenin, chrysoeriol, procyanidins, catechin, epicatechin, cartenoids, kaempferol[8-11] and p-coumaric acid [12]). In addition, fatty acids such as oleic, lauric, palmitic, myristic and linoic[13] and phytosterols (sitosterol, iso fucosterol, stigmasterol, campesterol)[14] also identified. Recently, malonyl derivatives were identified for

the first time as one of *Phoenix dactylifera* (Medjool palm) constituents[15].

In many regions several experimental studies; have tested the biological activities of different parts of date palm extracts and reported their therapeutic value[16, 17]. Among these effects, the antibacterial[18-20], antifungal[21], antiviral[22] anti-diabetic[23, 24], antioxidant[25-30] and anti-tumor [31-33], neuropharmacological, analgesic[34], anti-psoriasis[35], anti-Inflammatory[36, 37], hypolipidemic, cardioprotective[38], and anti-allergic properties[39] were found.

The diversity of date palm cultivars in North Africa is attributed mainly to the climate of the region. Libya in particular, still cultivates more than 400 different date varieties. This richness make Libya valuable producer of such highly effective nutritive product. Thus, the International Union for protection of New Varieties of Plants cooperative program between Italy and Libya, where focusing on 18 Libyan date palm cultivars in the oasis of Al Jufrah (Hun, Waddan, Sokna, Zellah and Al fughah)[40, 41].

Most of previous research studies were concentrated on the fruit part of Date palm *P. dactylifera* because as it is the edible portion. However, it has been found to be a good source of phytochemicals that are interesting for their antimicrobial effects. Discovery of new antimicrobials become a challenging field; especially, after the alarming increase in number of resistant pathogenic microorganism to an existing antibiotics. Therefore, in the present study seed

part will be investigated to discover the biological activity of Libyan dates.

Aim and objective

The aim of this study was to investigate the phytochemical screening and to evaluate antimicrobial activity of five types of date palm seeds which are produced by Libyan oasis (Aljufrah).

II. MATERIALS AND METHODS

2.1 Collection of samples:

Five types of dates (fruits) at the *tmar* stage which were locally known as: *Abel*, *Bestian*, *Deglet*, *Hamrai*, and *Khadrai* from region of Aljufrah are collected. The dates were deseeded by removing the seeds and each type of seeds was kept separately.

2.2 Sample preparation:

Seeds were sun dried for one week during summer then heated in oven at 100 °C for 2 hours then roasted. After samples cooled down, each seeds type was grinded into fine powder with the help of grinder. The powders were stored in glass an airtight container and kept at dry place for the experiment (figure 1).

2.3 Extract preparation:

The 100mg powders of roasted dried seeds were immersed in 300 ml methanol. After filtration, the filtrate remained under hood until get dry completely. The extract residues were stored in cool place for study experiments.

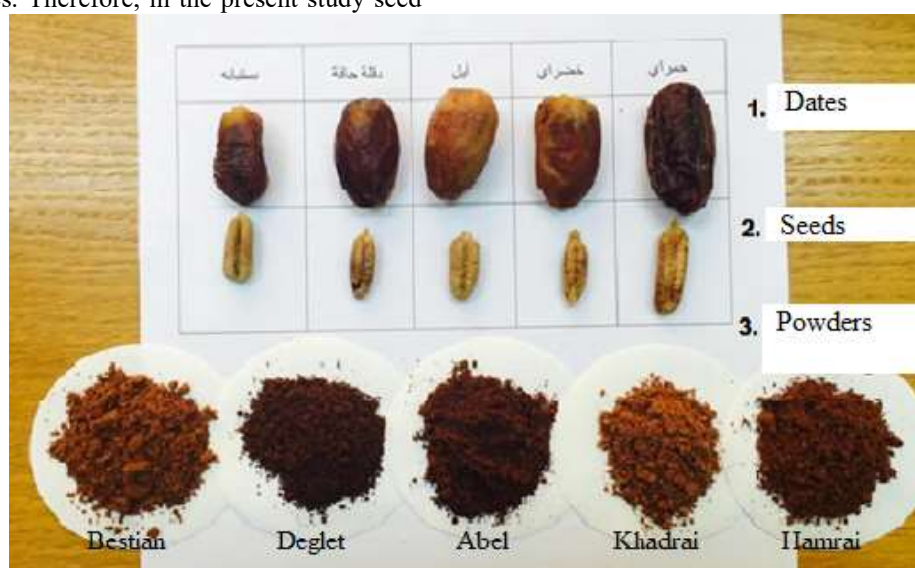


Figure 1: The five samples of Libyan date varieties (Bestian, Deglet, Abel, Khadrai and Hamrai) in their different stages of preparation.

2.4 Qualitative phytochemical screening of crude methanolic extracts:

All the extracts were qualitatively examined for their phytochemicals by following standard procedure in the literature. [42, 43]

Detection of flavonoids:

Alkaline Reagent Test:

1g of powdered material was boiled in distilled water and 5 ml sodium hydroxide solution. It yields of intense yellow color, which becomes colorless on addition of dilute acid, indicates the presence of flavonoids.

Detection of alkaloids:

The methanolic extracts were dissolved individually in dilute Hydrochloric acid and filtered. The filtrates were treated with Mayer's reagent (formation of a yellow colored precipitate indicates the presence of alkaloids) and Dragendroff's reagent (formation of red precipitate indicates the presence of alkaloids).

Detection of phytosterols

Salkowski's Test: Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of Conc. Sulphuric acid, shaken and allowed to stand. Appearance of golden yellow color indicates the presence of triterpenes.

Detection of phenols

Ferric Chloride Test: Water extracts of samples were treated with 3-4drops of ferric chloride solution. Formation of bluishblack color indicates the presence of phenols.

Detection of tannins

Gelatin Test: water extract of samples were treated with, 1% gelatin solution containing sodium chloride. Formation of white precipitate indicates the presence of tannins.

Detection of glycosides:

Aqueous extract were hydrolyzed with dil.HCl, and then subjected to test for glycosides. Modified Borntrager's Test: Extracts were treated with Ferric Chloride solution and immersed in boiling water for about 5 minutes. The mixture was cooled and extracted with equal volumes of benzene. The benzene layer was separated and treated with ammonia solution. Formation of rose-pink color in the ammoniacal layer indicates the presence of anthraquinone glycosides.

Detection of saponine

Foam test: 2 ml of aqueous filtrate was taken in a test tube and shaken vigorously. Formation of froth of more than 1cm long indicates the presence of saponins.

Detection of fixed oil

Spot test: A small quantity of extract was pressed between two filter papers.

Oil stain on the paper indicated the presence of fixed oils.

2.5 In vitro antibacterial activities of plant extract

Determine antibacterial activity of five seeds extract against G+ ve bacteria and G- ve bacteria. The extracts activity was tested by using cup- cut diffusion method.

Microorganism:

Standard bacterial strains used in this study were *Pseudomonas aeruginosa* NCTC 12903/ATCC, *Escherichia coli* NCTC 12241/ATCC 25922, Methicillin resistant *Staphylococcus aureus* (MRSA) NCTC 13373/ATCC 43300, *Staphylococcus aureus* NCTC 12973/ATCC, *Proteus vulgaris* NCTC 4175/ATCC 13315, *Klebsiella pneumoniae*, NCTC 9633/ATCC, the standard bacterial strains were activated and cloned three successive times in nutrient agar and stored on nutrient agar slants at 4°C.

Screening of antimicrobial activity:

Antimicrobial susceptibility was tested using Cup-cut agar method[44] was used throughout this study to find out if the extracts have the ability to inhibit growth of bacteria. This method was performed using freshly prepared Mueller Hinton agar with overnight culture of bacteria. Inoculum were prepared by suspending the freshly grown bacteria in sterile normal saline and adjusted to a 0.5 McFarland standard. On each plate wells (5 mm) were made by sterile cork borer. Each well was filled with 100µL of seeds extract and the plates were then re-incubated for 24 h at 37°C. Then diameters of zones of inhibition were measured in millimeter. Inhibition zones were indicated by a lack of microbial growth due to inhibitory concentrations. The antibiotics Cephalexin (5µg) and a 5% (v/v) phenol were used as standards to compare the activity of seed extracts in inhibiting the growth of bacteria. Each experiment was carried out three times.

2.6 Statistical Analysis of Data

The data was tested for normality using a QC Analyses/K-S Normality Test. Normally distributed data was analyzed by student's t-test using the Statview® version 5.0.1 software package (SAS Institute Inc, Abacus Concept, Inc., Berkeley, CA, USA).

III. RESULTS

The phytochemical analysis of methanolic extract of five different Libyan date palm seeds (Bestian, Deglet, Abel, Khadrai and Hamrai) showed the presence of aflavnoids, glycosides, phytosterols, phenolic compound, tannins, and fixed oil absences of alkaloids and saponine (Table 1).

The effect of seed methanolic extracts of date plam on different types of bacteria strains is shown in table 2 and figures 2 -3) with clear valuable antimicrobial activity. That most potential date seed extracts were found to be Deglet then followed by Bestain and Hamrai.

The susceptibility of bacterial species to five methanolic extracts of *Phoenix dactylifera* L seeds by cup-cut agar method exhibited Inhibition zones (IZD). That inhibition zone diameters are vary from 15.2 ± 0.1 to 3.02 ± 0.01 against *MRSA*; 16.04 ± 0.1 to 6.0 ± 0.7 against *Pseudomonas aeruginosa*; 14.2 ± 0.1 to 6.0 ± 0.7 against *Proteus vulgaris*; 18.3 ± 0.2 to 6.0 ± 0.1 and against *Staphylococcus aureus*; 14.04 ± 0.2 to 2.0 ± 0.02 against *E. coli*; and 18.0 ± 0.1 to 3.2 ± 0.02 against *Klebsiella pneumoniae* (Table 2).

Ultimately, maximum inhibition zone diameter (IZD) was achieved by the methanolic extract of Deglet; 19.2 ± 0.1 and 18.3 ± 0.2 mm against *Proteus vulgaris* and *Staphylococcus aureus* respectively. The Bestain seed extract showed potential antimicrobial activity as it demonstrated an inhibition zone diameter (IZD) of 18.0 ± 0.1 and 17.5 ± 0.3 against *Klebsiella pneumonia* and *Staphylococcus aureus* respectively. In addition, the Hamrai seed extract showed moderate antimicrobial activity as it demonstrated inhibition zone diameter (IZD) of 16.5 ± 0.2 and 14.5 ± 0.2 against *Staphylococcus aureus* and *MRSA* respectively. On the other hand, Khadrai seed extract showed neglected antibacterial effects compared to other varieties.

Table 1. Phytochemical screening of five different seeds of *Phoenix dactylifera* L

Methanolic extract of five seed samples					
Samples Tests	Abel	Deglet	Bestian	Hamrai	Khadrai
Flavonoids	+	+	+	+	+
Tannins	+	+	+	+	+
Alkaloids	-	-	-	-	-
Glycosides	+	+	+	+	+
Saponine	-	-	-	-	-
Fixed oil	+	+	+	+	+
Phytosterol	+	+	+	+	+

Table 2. Inhibition zones of five different seeds types of date plam (*Phoenix dactylifera* L) against various bacteria using 100µL of methanolic seed extract.

Diameter of inhibition zone $\pm$ SE						
	<i>MRSA*</i>	<i>Pseud. aeruginos</i>	<i>Proteus vulgaris</i>	<i>Staph. aureus</i>	<i>E.Coli</i>	<i>K.Paneumoni ae</i>
Abel	7.8 ± 0.4	6.0 ± 0.7	8.2 ± 0.2	8.9 ± 0.03	5.0 ± 0.03	10.2 ± 0.1
Deglet	15.0 ± 0.03	15.02 ± 0.1	19.2 ± 0.1	18.3 ± 0.2	14.04 ± 0.2	13.8 ± 0.2
Bestian	15.2 ± 0.1	16.04 ± 0.1	16.3 ± 0.2	17.5 ± 0.3	6.00 ± 0.1	18.0 ± 0.1
Hamrai	14.5 ± 0.2	12.02 ± 0.1	12.3 ± 0.2	16.5 ± 0.2	2.5 ± 0.01	12.3 ± 0.1
Khadrai	3.02 ± 0.01	8.2 ± 0.1	7.2 ± 0.1	6.0 ± 0.1	2.0 ± 0.02	3.2 ± 0.02
Phenol	13.5 ± 0.1	19.5 ± 0.1	19.3 ± 0.1	28.0 ± 0.0	29.0 ± 0.0	19.1 ± 0.01

Cephalexin	3.2 ±0.01	1.5 ±0.01	3.0±0.0	3.3 ±0.09	3.0 ±0.01	3.0± .02
------------	-----------	-----------	---------	-----------	-----------	----------

**MRSA*= Methicillin resistant *Staphylococcus aureus*

The values expressed as mean of triplicates ± SE

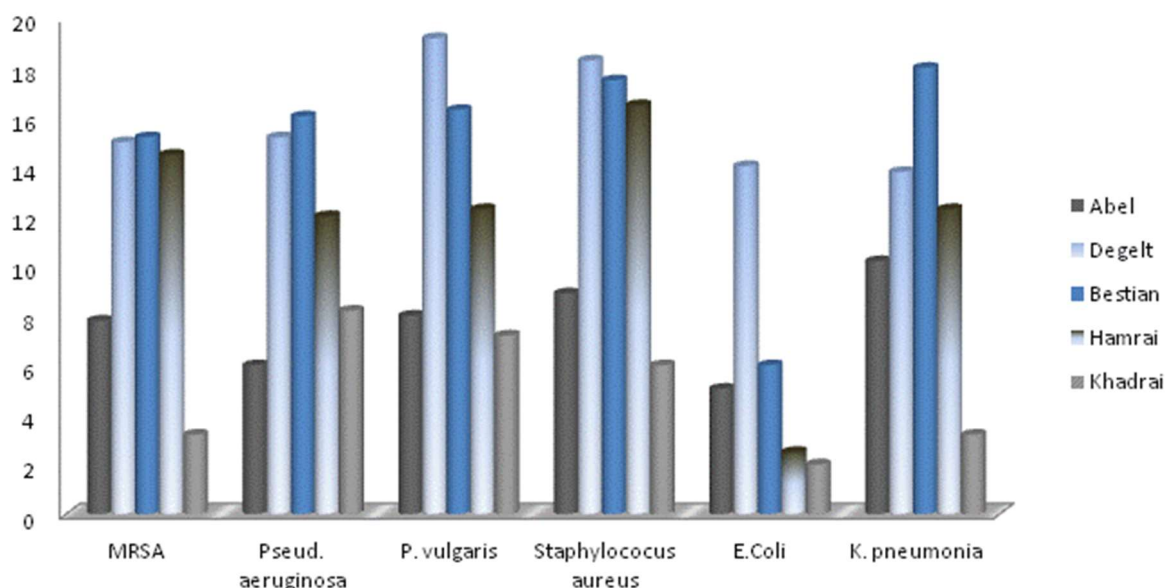


Figure2. Seeds methanolic extract inhibition result. MRSA (Methicillin resistant *Staphylococcus aureus*), Pseud. aeruginosa, P. vulgaris, *Staphylococcus aureus* . E. coli, K. pneumonia.)

IV. DISCUSSION:

Date seeds may have potential high valuable nutraceutical components, however, these compounds have diminutive use therapeutically, and they are discarded in animal feed. Limited research has been focused on therapeutic activity of date palm seeds and their components[45, 46]. In general, the results of the present study were similar to most phytochemicals of seed of *Phoenix dactylifera* reported in literature. As the results reveal that all seed samples possess flavonoids, glycosides, tannins, phytosterol, and fixed oil. In contrast, the current results showed absences of alkaloids and saponine while they were reported in Saudian and Iraquian studies[42, 47]. The omitted of alkaloids content from the five seeds extract supported by previous Algerian Maxican research[7, 46, 48]; however; presence of saponine was reported for Algerian and Mexican date seeds[46, 48].The existing differences and concentration of phytochemicals content may be varied environmentally or due to part used. Nevertheless, in literature many studies reported significant difference in phenolic , flavonoids and tannins contents [49, 50].

The highest antimicrobial activity was obtained by Deglet, Bestian and Hamrai methanolic against *Proteus vulgaris* on the following order (19.2±0.1 , 16.3±0.2 and 12.3± 02) this finding is reported for the first time by our study .Furthermore, potential activaties against *Staphylococcus aureus* (18.3±0.2, 17.5±0.3, 16.5±0.2) were demonstrated by methanolic extract of deglet, Bestian, and Hamrai respectively. These later results were supported by literature; as different date seeds cultivated in different countries revealed their bactericidal effects against *Staph.aureus*[50, 51]. Moreover, the same sample varieties showed high inhibitory activity against *Pseudomonas aeruginos* (16.04±0.1 Bestian), (15.02±0.1 Deglet), and (12.02±01 Hamrai), which matched investigation done by Tunisian study for date palm seeds [51]. Interestingly, where Iranian study reported *Klebsiella pneumonia* was resistant to different solvent extracts of different part of date palm[52], four samples out of five in our current study, exhibited potential activity against *k.pneumonia* and MRSA which confirmed previously in the literature[20]. Furthermore, Bestian demonstrated the highest inhibitory growth effect on *Klebsiella pneumonia* (18.0±0.1) followed by Deglet (13.8± 0.2) then Hamrai (12.3±0.1), lastly, Abel (10.2 ±0.1) with

neglected effect to Khadrai. Surprisingly, *E.coli* showed resistant to the main sample, except Deglet which demonstrated highest sensitivity (14 ± 0.2). This results shown similarity to another study in literature which reported antimicrobial activity of *Phoenix dactylifera* seeds against many gram-positive and gram-negative bacteria [20]. Overall, the Khadrai extract showed moderate to week effect to tested microorganism, which may be related to the diverse of phytochemicals and their concentration in the plant.

The dissimilarity of antimicrobial results of the five seed type is mostly related to concentration of phenolic flavonoid content, which reflect sensitivity of bacteria to extract[7]. The data presented by this study, supported by the previously published works, while provide first antimicrobial activity against *P.vulgaris* encourage the researcher to continue study and exploring the effectiveness of *Phoenix dactylifera* seeds against antibiotic-resistant infections.

V. CONCLUSION:

Over all, the results of present study shows the antibacterial properties of five Libyan samples of date seed extract against a wide spectrum of bacteria. Among the tested bacteria, *Proteus vulgaris*, *Staphylococcus aureus*, and *Klebsiella pneumonia*. The results drive investigator to carry out more research to identify the phytochemicals of date seeds which behind antibacterial properties. Particularly in Libya as *Phoenix dactylifera* is cultivated in three different geographical regions with different climate. Further research is suggested to isolate and enhance the availability of bioactive important compounds.

ACKNOWLEDGEMENT

Authors are grateful to staff of Phytochemistry lab and Microbiology lab in faculty of Pharmacy for supporting this research.

REFERENCES

- [1] Vayalil, P.K., *Date fruits (Phoenix dactylifera Linn): an emerging medicinal food*. Crit Rev Food Sci Nutr, 2012. 52(3): p. 249-71.
- [2] Selmani, C., D. Chabane, and N. Bouguedoura, *Ethnobotanical Survey of Phoenix Dactylifera L. Pollen Used for the Treatment of Infertility Problems in Algerian Oases*. Afr J Tradit Complement Altern Med, 2017. 14(3): p. 175-186.
- [3] El-Far, A.H., et al., *Date Palm (Phoenix dactylifera): Novel Findings and Future Directions for Food and Drug Discovery*. Curr Drug Discov Technol, 2018.
- [4] Boukouada, M., et al., *Chemical composition and antioxidant activity of seed oil of two Algerian date palm cultivars (Phoenix dactylifera)*. Nat Prod Commun, 2014. 9(12): p. 1777-80.
- [5] Baliga, M.S., et al., *A review of the chemistry and pharmacology of the date fruits (Phoenix dactylifera L.)*. Food research international, 2011. 44(7): p. 1812-1822.
- [6] Al. El-Far, H.S., M. Abdel-Daim, S.. Al Jaouni and S. Mousa, *Date Palm (Phoenix dactylifera): Protection and Remedy Food. Current trends in nutraceuticals*, 2016. 1(2): p. 2-9.
- [7] Bentradi, N., et al., *Studies on chemical composition and antimicrobial activities of bioactive molecules from date palm (Phoenix dactylifera L.) pollens and seeds*. African Journal of Traditional, Complementary and Alternative Medicines, 2017. 14(3): p. 242-256.
- [8] Hong, Y.J., et al., *The flavonoid glycosides and procyanidin composition of Deglet Noor dates (Phoenix dactylifera)*. J Agric Food Chem, 2006. 54(6): p. 2405-11.
- [9] Hammouda, H., et al., *Detailed polyphenol and tannin composition and its variability in Tunisian dates (Phoenix dactylifera L.) at different maturity stages*. J Agric Food Chem, 2013. 61(13): p. 3252-63.
- [10] Habib, H.M., et al., *Polyphenolic compounds in date fruit seed (Phoenix dactylifera): characterisation and quantification by using UPLC-DAD-ESI-MS*. J Sci Food Agric, 2014. 94(6): p. 1084-9.
- [11] Louhaichi, M., et al., *Initial assessment of medicinal plants across the Libyan Mediterranean coast*. Advances in Environmental Biology, 2011. 5(2): p. 359-370.
- [12] Kchaou, W., et al., *Phenolic profile, antibacterial and cytotoxic properties of second grade date extract from Tunisian cultivars (Phoenix dactylifera L.)*. Food Chem, 2016. 194: p. 1048-55.
- [13] A. Taftia, B.P., *Chemical composition of seed and seed oil from Iranian commercial date cultivars* Journal of Food and Bioprocess Engineering 2019. 2(1): p. 1-6.
- [14] Saafi, E.B.E.A., A.; Issaoui, M.; Hammami, M.; Achour, L., *Phenolic content and antioxidant activity of four date palm (Phoenix dactylifera L.) fruit varieties grown in Tunisia*. International Journal of food and science technology, 2009. 44: p. 2314-2319.
- [15] Abu-Reidah, I.M., et al., *Phenolic composition profiling of different edible parts and by-products of date palm (Phoenix dactylifera L.) by using HPLC-DAD-ESI/MS(n)*. Food Res Int, 2017. 100(Pt 3): p. 494-500.

- [16] Qadir, A., Shakeel, Faiyaz Ali, Athar Faiyazuddin, Md, *Phytotherapeutic potential and pharmaceutical impact of Phoenix dactylifera (date palm): current research and future prospects*. Journal of food science and technology, 2019. 57(4): p. 1191-1204.
- [17] R. Al-Alawi, J.A.-M., J. Al-Nadabi, B. Al-Shihi and Y. Baqi, *Date Palm Tree (Phoenix dactylifera L.): Natural Products and Therapeutic Options*. Frontiers in plant and science 2017. 8: p. 845.
- [18] Samad, M.A., et al., *Antibacterial Properties and Effects of Fruit Chilling and Extract Storage on Antioxidant Activity, Total Phenolic and Anthocyanin Content of Four Date Palm (Phoenix dactylifera) Cultivars*. Molecules. 21(4): p. 419.
- [19] Perveen, K., N.A. Bokhari, and D.A. Soliman, *Antibacterial activity of Phoenix dactylifera L. leaf and pit extracts against selected Gram negative and Gram positive pathogenic bacteria*. Journal of Medicinal Plants Research, 2012. 6(2): p. 296-300.
- [20] M. ALrajhi, M.A.-R., S. Eltom, E. Alhazmi, and M.M.a. ALreshidi, *Antibacterial activity of date palm cake extracts (Phoenix dactylifera)*. Cogent food A griculture, 2019. 5(1): p. 1625479.
- [21] Boulouar, N., A. Marouf, and A. Cheriti, *Antifungal activity and phytochemical screening of extracts from Phoenix dactylifera L. cultivars*. Nat Prod Res, 2011. 25(20): p. 1999-2002.
- [22] Jassim, S.A. and M.A. Naji, *In vitro Evaluation of the Antiviral Activity of an Extract of Date Palm (Phoenix dactylifera L.) Pits on a Pseudomonas Phage*. Evid Based Complement Alternat Med, 2008. 7(1): p. 57-62.
- [23] Chakroun, M., et al., *Evaluation of anti-diabetic and anti-tumoral activities of bioactive compounds from Phoenix dactylifera L's leaf: In vitro and in vivo approach*. Biomed Pharmacother, 2016. 84: p. 415-422.
- [24] Rock, W., et al., *Effects of date (Phoenix dactylifera L., Medjool or Hallawi Variety) consumption by healthy subjects on serum glucose and lipid levels and on serum oxidative status: a pilot study*. J Agric Food Chem, 2009. 57(17): p. 8010-7.
- [25] Mohamed, R.M., et al., *Chemical composition, antioxidant capacity, and mineral extractability of Sudanese date palm (Phoenix dactylifera L.) fruits*. Food Sci Nutr, 2014. 2(5): p. 478-89.
- [26] Al-Farsi, M., et al., *Comparison of antioxidant activity, anthocyanins, carotenoids, and phenolics of three native fresh and sun-dried date (Phoenix dactylifera L.) varieties grown in Oman*. J Agric Food Chem, 2005. 53(19): p. 7592-9.
- [27] Vayalil, P.K., *Antioxidant and antimutagenic properties of aqueous extract of date fruit (Phoenix dactylifera L. Arecaceae)*. J Agric Food Chem, 2002. 50(3): p. 610-7.
- [28] Mohamed Lemine, F.M., et al., *Antioxidant activity of various Mauritanian date palm (Phoenix dactylifera L.) fruits at two edible ripening stages*. Food Sci Nutr, 2014. 2(6): p. 700-5.
- [29] Ambigaipalan, P. and F. Shahidi, *Antioxidant potential of date (Phoenix dactylifera L.) seed protein hydrolysates and carnosine in food and biological systems*. J Agric Food Chem, 2015. 63(3): p. 864-71.
- [30] Eimad dine Tariq Bouhlali , M.B., Khalid Sellam , and C.A. Mohamed Benlyas , Younes Filali-Zegzouti, *Evaluation of antioxidant, antihemolytic and antibacterial potential of six Moroccan date fruit (Phoenix dactylifera L.) varieties*. Journal of King Saud University -Science, 2016. 28: p. 136-142
- [31] Ishurd, O., et al., *Antitumor activity of -d-glucan from Libyan dates*. Journal of medicinal food, 2004. 7(2): p. 252-255.
- [32] Ishurd, O. and J.F. Kennedy, *The anti-cancer activity of polysaccharide prepared from Libyan dates (Phoenix dactylifera L.)*. Carbohydrate Polymers, 2005. 59(4): p. 531-535.
- [33] Khan, F., et al., *Anti-cancer effects of Ajwa dates (Phoenix dactylifera L.) in diethylnitrosamine induced hepatocellular carcinoma in Wistar rats*. BMC Complement Altern Med. 17(1): p. 418.
- [34] Sheikh, B.Y., et al., *Comparative study of neuropharmacological, analgesic properties and phenolic profile of Ajwah, Safawy and Sukkari cultivars of date palm (Phoenix dactylifera)*. Orient Pharm Exp Med, 2016. 16(3): p. 175-183.
- [35] Hussein, Z.A., et al., *Possible anti-psoriasis effect of the methanol extract of Phoenix dactylifera L. seeds*. Journal of Herbmmed Pharmacology, 2020. 9(4).
- [36] Eddine, L.S., et al., *Scavenging Effect, Anti-Inflammatory and Diabetes Related Enzyme Inhibition Properties of Leaves Extract from Selected Varieties of Phoenix dactylifera L.* International Journal of Pharmacognosy and Phytochemical Research, 2014. 6(1): p. 66-73.
- [37] Elberry, A.A., et al., *Anti-inflammatory and antiproliferative activities of date palm pollen (Phoenix dactylifera) on experimentally-induced atypical prostatic hyperplasia in rats*. J Inflamm (Lond). 8(1): p. 40.
- [38] Al-Yahya, M., et al., *Ajwa dates (Phoenix dactylifera L.) extract ameliorates isoproterenol-induced*

- cardiomyopathy through downregulation of oxidative, inflammatory and apoptotic molecules in rodent model. *Phytomedicine*, 2015. 23(11): p. 1240-1248.
- [39] Karasawa, K. and H. Otani, *Anti-allergic properties of a matured fruit extract of the date palm tree (*Phoenix dactylifera* L.) in mite-sensitized mice*. *J Nutr Sci Vitaminol* (Tokyo), 2012. 58(4): p. 272-7.
- [40] Battaglia, M., et al., *Date palm status and perspective in Libya*, in *Date palm genetic resources and utilization*. 2015, Springer. p. 223-255.
- [41] Racchi, M., et al., *Genetic characterization of Libyan date palm resources by microsatellite markers*. *3 Biotech*, 2013. 4(1): p. 21-32.
- [42] R. Sundar, G.S., S. Shankar, S. Settu, L. Ravi, *Bioactivity of *Phoenix dactylifera* seed and its phytochemical analysis*. *International Journal of Green Pharmacy* 2017. 2(11): p. 292.
- [43] A. Pandey, S.T., *Concept of standardization, extraction and pre phytochemical screening strategies for herbal drug*. *Journal of Phramacognosy and Phytochemistry*, 2014. 2(5): p. 115- 119.
- [44] P.Murray, K.R., M.Pfaller, *Medical Microbiology*. 8th ed. 2015: Elsevier.
- [45] A. El-Far, H.S., M. Abdel-Daim, S. Al Jaouni and S. Mousa5, *Date Palm (*Phoenix dactylifera*): Protection and Remedy Food*. *Current Trend in Nutraceuticals*, 2016. 1(29).
- [46] F. Mihoub, F.G., S.Lakhdar-Toumi, *Bioactivity of Algerian palm dates *Phoenix dactylifera* L. Ukrainian Food Journal*, 2019. 8(2): p. 249-259.
- [47] S. Yaseen , H.A., A. Kadhimi, and E.Aboglidia, *Antibacterial activity of palm heart extracts collected from Iraqi *Phoenix dactylifera* L.* *Journal of Techniques*. 1(1): p. 52-59.
- [48] R.Torres, N.U., B.Salas,N.González, C. González, D. Chávez, I. Guerrero, L. Rubio, M.Vázquez, J.Montiel, A.Maza, P. Mahadevan and R. Krueger, *Nutritional assessment, phytochemical composition and antioxidant analysis of the pulp and seed of medjool date grown in Mexico*. *Peer J*, 2019. 10: p. 2-19.
- [49] John, J.A. and F. Shahidi, *Phenolic content, antioxidant and anti-inflammatory activities of seeds and leaves of date palm (*Phoenix dactylifera* L.)*. *Journal of Food Bioactives*, 2019. 5: p. 120-130.
- [50] Radfar, R., et al., *Assessment of phenolic contents and antioxidant and antibacterial activities of extracts from four varieties of Iranian date Palm (*Phoenix dactylifera* L.) seeds*. *Applied Food Biotechnology*, 2019. 6(3): p. 173-184.
- [51] Masmoudi-Allouche, F.z., et al., *Phytochemical profile, antioxidant, antibacterial, antidiabetic and anti-obesity activities of fruits and pits from date palm (*Phoenix dactylifera* L.) grown in south of Tunisia*. *Journal of Pharmacognosy and Phytochemistry*, 2016. 5(3): p. 15.
- [52] M. Shakiba, A.K., P. Parsia, *Atimicrobial activity of different parts of *Phoenix dactyfera**. *International Journal of Molecular and Clinical Microbiology*, 2011. 1: p. 107-111.