

The effect of date palm fruit (*Phoenix dactylifera* L.) on serum lipid and lipoprotein concentrations in rats fed cholesterol- supplemented diet

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Abstract.

BACKGROUND: The date palm fruit (*Phoenix dactylifera* L.) has been known for many health benefits, but its antihyperlipidaemic activity still remains unclear.

OBJECTIVE: To investigate effects of Birhi date palm fruit, “Khalal” and “Tamr”, on serum lipids, body weight and food intake in cholesterol-fed rats.

METHODS: Sixty male Sprague–Dawley rats were assigned into 5 cholesterol-free (control) or 5 cholesterol-supplemented (experimental) diets containing 0%, 5% and 10% of either Khalal or Tamr and given *ad libitum* to the rats for 6 weeks. Serum total cholesterol (TC), low- and very low-density lipoprotein cholesterol (LDL-C and VLDL-C), high-density lipoprotein cholesterol (HDL-C) and triglycerides (TG) were then quantified and other biological parameters were assessed.

RESULTS: Compared to control, cholesterol induced significant ($p < 0.05$) increase in serum LDL-C, TC/TG ratio and atherogenic index and decrease in TG and HDL-C/LDL-C ratio, whereas other lipid fractions, food intake and weight gain were unchanged. In all rats, none of the studied variables were appreciably affected by dates feeding, except for increased ($p < 0.05$) and linearly responded ($r^2 = 0.348, p < 0.01$) atherogenic index induced by Khalal. Lipid variables and their calculated ratios that were increased or decreased by cholesterol remained unaffected by dates feeding.

CONCLUSION: In normal or cholesterol-fed rats, Birhi Khalal or Tamr exert little or no effect on serum lipids and are ineffective to counteract the atherogenic effect of cholesterol.

Keywords: Birhi date palm fruit, khalal, tamr, lipid profile, cholesterol, rats

1. Introduction

Atherosclerosis is central to cardiovascular disease and its development is related to elevated concentrations of plasma cholesterol, particularly low-density lipoprotein cholesterol [1]. Cholesterol plaques are characteristic features of atherosclerotic lesions in humans and animals [2, 3]. Cardiovascular disease is an epidemic disorder and continues to be the most prevalent cause of death worldwide [4–6]. Given the several health risks and public health burdens of this disease [7], its prevention or management is becoming a major challenge. Therefore, identifying dietary factors that may favorably affect serum cholesterol is of great importance.

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Current management of cardiovascular disease involves various lifestyle changes, dietary and exercise regimes [7, 8] and the use of drugs or medical interventions [9, 10]. Nowadays, there is growing interest in the use of plant foods for the prevention and management of cardiovascular disease and other related disorders [11, 12]. A special emphasis is being devoted to the Mediterranean diet [13].

The date palm fruit (*Phoenix dactylifera L.*), a species belonging to the Arecaceae family, is one of the well known and oldest crops in the desert regions of the Middle East and North Africa and has formed the basis of survival of many ancient nomads [14]. The fruit is an influential food for local populations with great socio-economic and traditional importance [15]. Even today, the fruit constitutes an essential part of the local diet, mainly in the form of fresh dates at different stages of maturity including Kimri, Khalal, Rutab and Tamr [16]. The date palm fruit is known to have a good nutritional value, besides it contains several active phytochemicals such as polyphenols, sterols, tannins and carotenoids [17]. Further, the fruit is reported to possess a number of biological activities, such as antimicrobial, anti-inflammation, anticancer, anti-diarrhea, antiulcer and antioxidant [18].

Evidence for possible antihyperlipidaemic activity of the date palm fruit in humans is relatively limited and debatable, whereas that in animals is unavailable. Variable effects of different varieties of whole date palm fruit on serum lipids in healthy subjects have been reported [19, 20]. On the other hand, animal evidence that links different date palm preparations with serum lipids is consistent. Improvement in lipid profile has been repeatedly documented by studies investigating date palm seed flour, seed fiber, leaves extracts or pollen grains in various animal models [21–25]. Nevertheless, effects of Birhi date palm fruit at different maturity stages in humans and animals is yet to be elucidated. Therefore, this study aimed at investigating the effect of Birhi variety of date palm fruit at two maturity stages, “Khalal” and “Tamr”, on serum lipids and lipoproteins, body weight and food intake in rats fed cholesterol-supplemented or cholesterol-free diets.

2. Materials and methods

2.1. Preparation of date palm fruit

The date palm fruit (*Phoenix dactylifera L.*) at two stages of maturity, “Khalal” and “Tamr”, was obtained from the Al-Baraka Farms, Jordan. The fruit belongs to the Birhi variety. Khalal and Tamr used in this study were fresh and high quality, and conformed to the chemical and physical criteria set by the Jordanian standard specifications [26]. The Dates were washed with tap water and air-dried over cotton cloths, packed in sealed bags under vacuum and stored frozen at -20°C for further use [27]. Samples of Khalal and Tamr were pitted and allowed to pass through a food mincing machine (Kenwood[®], UK) with a sieve of 4 mm mesh till became smooth paste, then dried in a drying oven (Memmert, Karl lob, Germany) at 70°C and finely powdered with a mechanical mixer. The resultant powder was placed desiccated in sealed polythene bags and stored frozen at -20°C until chemical analysis [28]. Fresh quantities of Khalal and Tamr pastes were prepared daily and used in the formulation of experimental diets.

2.2. Animals

Adult male Sprague-Dawley rats ($n=60$) were obtained from the Experimental Animal Unit of the Department of Nutrition and Food Technology, The University of Jordan, Amman, Jordan. The animals were acclimatized for 2 weeks before the experiment, during which they were fed on chow diet with free access to tap water. They were individually housed in plastic cages with stainless steel wire-mesh bottom (North Kent Plastic Cages, Ltd, Dartford, England) under controlled temperature ($22 \pm 2^{\circ}\text{C}$) and hygienic conditions with 12-hour light, 12-hour dark cycle. All the experiments involving animals were approved by the Institutional Animal Ethics Committee and carried out according to the recommended guidelines for animal use.

2.3. Diets

Ten isocaloric and isonitrogenous diets were prepared, 5 of them were cholesterol-free and differed in their date palm fruit content (0%, 5% or 10% w/w) of either Khalal or Tamr (control diets), while in the other 5, 1% cholesterol

was added, at the expense of fat, to induce hyperlipidaemia (experimental diets). For the purpose of conserving the nutritional value of the date palm fruit, pastes of Khalal and Tamr were freshly prepared on daily basis, as described earlier, and were incorporated into the diets [29]. The pastes of Khalal or Tamr were first thoroughly mixed with cornstarch in a stainless steel blender (Kenwood®, England) and allowed to pass through a sieve of 1 mm mesh to obtain homogenous mixes, before the other ingredients were added. Diets were kept refrigerated at 4°C. The protein and carbohydrate contents of Khalal and Tamr were taken into consideration in the calculation of nutrient composition of the diets. The composition of experimental diets is described in Table 1. Dietary supply of protein, vitamins and minerals were in accordance with the dietary recommended allowances for rats from the American Institute of Nutrition [30]. Proximate nutrient composition of Khalal and Tamr is shown in Table 2.

2.4. Experimental protocol

At the beginning of the experiment, animals weighed 259.8 ± 1.8 g ($n=60$) and they were assigned into the 5 cholesterol-free (control) or the 5 cholesterol-supplemented (experimental) groups (6 rats/group). During the

Table 1
Composition of the experimental diets

Ingredient	Cholesterol-free diets (g·kg ⁻¹)					Cholesterol-supplemented diets (g·kg ⁻¹)				
	Control	Khalal		Tamr		Control	Khalal		Tamr	
Date	0	50	100	50	100	0	50	100	50	100
Cholesterol	0	0	0	0	0	10	10	10	10	10
Cornstarch	657	642.8	628.6	618.6	580.2	657	642.8	628.6	618.6	580.2
Egg albumin	180	178.6	177.2	178.6	177.2	180	178.6	177.2	178.6	177.2
Corn oil	90	90	90	90	90	80	80	80	80	80
Vitamin mix (AIN-93) ¹	10	10	10	10	10	10	10	10	10	10
Mineral mix (AIN-93) ¹	35	35	35	35	35	35	35	35	35	35
DL-Methionine	3	3	3	3	3	3	3	3	3	3
Choline bitartrate	25	25	25	25	25	25	25	25	25	25
Tert-Butylhydroquinone	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008
Carbohydrate (%)	65.7	65.7	65.7	65.7	65.7	65.7	65.7	65.7	65.7	65.7
Protein (%)	18.0	18.0	18.0	18.0	18.0	18.0	18.0	18.0	18.0	18.0
Fat (%)	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0
Total energy (kcal. 100 g ⁻¹)	424.8	424.8	424.8	424.8	424.8	424.8	424.8	424.8	424.8	424.8

¹ AIN: American Institute of Nutrition [30].

Table 2
Composition of Birhi Khalal and Tamr palm date fruit¹

Component	Khalal (g·100g ⁻¹)	Tamr (g·100g ⁻¹)
Moisture	62.4 ± 0.02	15.9 ± 0.02
Carbohydrate	28.4 ± 0.03	76.8 ± 0.02
Protein	2.7 ± 0.01	2.8 ± 0.02
Fat	0.4 ± 0.02	0.3 ± 0.01
Ash	2.7 ± 0.03	2.1 ± 0.02
Fiber	3.4 ± 0.05	2.1 ± 0.02
Total energy (kcal.100 g ⁻¹)	128	321

¹ Mean of three determinations ± SEM, on fresh matter basis.

experimental period, which lasted for 6 weeks, experimental diets and tap water were given *ad libitum*. Body weight and food intake were monitored weekly. Food efficiency ratio as body weight gain per 100 g food intake was also calculated. On the termination day and after an overnight fast, animals were anesthetized using chloroform. Blood was collected by performing cardiac puncture and serum was isolated and stored frozen at -20°C until chemical analysis.

2.5. Chemical analysis

Serum lipid and lipoprotein concentrations including total cholesterol, low- and very low-density lipoprotein cholesterol, high-density lipoprotein cholesterol and triglycerides were determined by using commercial kits and in accordance to the manufacturer's instructions (Hoffmann-La Roche Diagnostics, Germany). Analysis was performed at the Medical Laboratories of the Islamic hospital, Amman, Jordan, using a pre-calibrated automated clinical chemistry analyzer (Roche/Hitachi 912 chemistry analyzer). Ratios of total cholesterol/ triglycerides and high-density lipoprotein cholesterol/ low-density lipoprotein cholesterol were calculated. Atherogenic index was also calculated as described elsewhere [31]. Proximate nutrient composition of the Khalal and Tamr used in the feeding experiments was determined by the Weende method [32]. Proximate analyses included the determination of moisture, carbohydrate, protein, fat, fiber and ash.

2.6. Statistical analysis

Data analysis was performed using statistical analysis software (SAS version 9, USA). Statistical significance was assessed by two-way ANOVA followed by the Duncan's multiple range tests, and the significance was set at $p < 0.05$. Data were expressed as means \pm standard errors of the mean (SEM). Orthogonal polynomial comparisons were used to identify statistically significant trends. This test determines the nature of the response of the studied variables to increasing levels (0%, 5%, 10%) of Khalal or Tamr. Linear and quadratic trends were given as coefficient of determination (r^2) at $p < 0.05$.

3. Results

The proximate nutrient composition of the date palm fruit used this study is given in Table 2. On fresh basis, the % content of energy and energy-releasing nutrients in Khalal and Tamr was respectively: energy (128; 321 kcal/100 g), carbohydrate (28.4%; 76.8%), protein (2.7%; 2.8%) and fat (0.4%; 0.3%).

Initial body weights were essentially similar ($p \geq 0.05$) in all rats of the control and experimental groups (Table 3). Compared to control, cholesterol feeding did not significantly influence ($p \geq 0.05$) body weight, weight gain, and food intake and food efficiency ratio. In neither control nor experimental groups did Khalal or Tamr feeding affect these variables.

Lipid and lipoprotein concentrations and their calculated ratios of rats fed Khalal or Tamr date palm fruit are shown in Table 4. In contrast to control, cholesterol feeding resulted in significant ($p < 0.05$) increase in serum LDL-cholesterol concentration, total cholesterol/ triglycerides ratio and atherogenic index and decrease in triglyceride concentration and HDL-cholesterol/ LDL-cholesterol ratio, whereas total cholesterol concentration was unaffected. Noteworthy, serum concentrations of HDL-cholesterol and VLDL-cholesterol were modestly but insignificantly ($p < 0.08$) decreased by cholesterol feeding.

In both control and experimental groups, none of the lipid and other biological variables were notably affected by date palm fruit feeding, except for increased ($p < 0.05$) atherogenic index induced by Khalal feeding (Tables 3 and 4). Parallel to these results, Khalal also showed an ascending linear trend ($r^2 = 0.348$, $p < 0.01$) for atherogenic index in experimental groups (Table 5). With the exception of the latter, the lipid variables and their calculated ratios that were increased or decreased by cholesterol feeding remained essentially ($p \geq 0.05$) unchanged by dates feeding. Differences between effects of Khalal or Tamr on these variables were insignificant (Table 4). Further, in control groups, Khalal and Tamr showed quadratic trends for serum HDL-cholesterol concentration ($r^2 = 0.249$, $p < 0.05$; $r^2 = 0.245$, $p < 0.05$ respectively), with Khalal exhibited an ascending linear trend for food intake ($r^2 = 0.236$, $p < 0.05$) (Table 5).

Table 3
Body weight, weight gain, food intake and food efficiency ratio of rats fed Khalal or Tamr date palm fruit for 6 weeks^{1,2}

Variable	Cholesterol-free groups						Cholesterol-supplemented groups					
	Control		Khalal		Tamr		Control		Khalal		Tamr	
Date (g·kg ⁻¹)	0	50	100	50	100	50	100	0	50	100	50	100
Initial body weight (g)	259.7 ± 6.5 ^a	259.6 ± 6.4 ^a	258.4 ± 6.4 ^a	258.1 ± 6.0 ^a	259.7 ± 6.1 ^a	260.8 ± 6.3 ^a	261.5 ± 6.6 ^a	260.7 ± 6.1 ^a	260.6 ± 6.1 ^a	260.7 ± 6.1 ^a	260.6 ± 6.1 ^a	259.5 ± 6.2 ^a
Final body weight (g)	370.1 ± 7.1 ^b	375.2 ± 12.2 ^{ab}	380.6 ± 10.4 ^{ab}	368.3 ± 7.6 ^b	363.7 ± 7.2 ^b	387.1 ± 12.9 ^{ab}	396.3 ± 16.3 ^a	387.1 ± 12.9 ^{ab}	392.9 ± 7.8 ^a	392.0 ± 8.5 ^a	392.9 ± 7.8 ^a	391.5 ± 5.7 ^a
Weight gain (g·day ⁻¹)	2.63 ± 0.08 ^b	2.75 ± 0.17 ^{ab}	2.91 ± 0.18 ^{ab}	2.62 ± 0.15 ^b	2.48 ± 0.11 ^b	3.01 ± 0.24 ^{ab}	3.21 ± 0.24 ^a	3.01 ± 0.24 ^{ab}	3.15 ± 0.11 ^a	3.13 ± 0.19 ^a	3.15 ± 0.11 ^a	3.15 ± 0.04 ^a
Food intake (g·day ⁻¹)	14.33 ± 0.33 ^c	14.51 ± 0.40 ^{bc}	15.73 ± 0.58 ^a	14.52 ± 0.25 ^{bc}	14.11 ± 0.40 ^c	15.20 ± 0.55 ^{ab}	15.89 ± 0.71 ^a	15.20 ± 0.55 ^{ab}	15.83 ± 0.41 ^a	15.83 ± 0.41 ^a	15.53 ± 0.45 ^{ab}	15.47 ± 0.41 ^{ab}
FER ³	18.38 ± 0.63 ^{ab}	18.91 ± 0.83 ^{ab}	18.42 ± 0.60 ^{ab}	18.03 ± 0.84 ^{bc}	17.53 ± 0.43 ^b	19.65 ± 0.96 ^{bc}	20.13 ± 0.69 ^a	19.65 ± 0.96 ^{bc}	19.67 ± 0.78 ^{bc}	19.67 ± 0.78 ^{bc}	20.32 ± 0.69 ^a	20.41 ± 0.69 ^a

¹Values are means ± SEM. ²Means within a row with different superscripts are significantly different ($p < 0.05$). ³FER: Food efficiency ratio, body weight gain (g)/100 g food intake.

Table 4
Serum lipid and lipoprotein concentrations and their calculated ratios of rats fed Khalal or Tamr date palm fruit for 6 weeks^{1,2}

Variable	Cholesterol-free groups						Cholesterol-supplemented groups					
	Control	Khalal	Tamr	Control	Khalal	Tamr	Control	Khalal	Tamr	Control	Khalal	Tamr
Date (g. kg ⁻¹)	0	50	100	0	50	100	0	50	100	0	50	100
TC ³ (mg.dl ⁻¹)	110.6 ± 9.0 ^{ab}	132.5 ± 9.4 ^a	117.2 ± 7.7 ^{ab}	115.5 ± 12.0 ^{ab}	98.0 ± 3.9 ^b	114.5 ± 6.7 ^{ab}	115.5 ± 12.0 ^{ab}	134.4 ± 7.3 ^a	127.1 ± 8.2 ^a	115.4 ± 7.8 ^{ab}	115.4 ± 7.8 ^{ab}	123.3 ± 6.0 ^a
HDL-C ⁴ (mg.dl ⁻¹)	78.7 ± 6.3 ^{ab}	90.7 ± 3.5 ^a	78.4 ± 2.6 ^{ab}	70.2 ± 6.5 ^b	68.2 ± 2.1 ^b	80.9 ± 3.2 ^{ab}	70.2 ± 6.5 ^b	75.5 ± 4.1 ^b	71.3 ± 5.5 ^b	67.5 ± 4.6 ^b	67.5 ± 4.6 ^b	72.9 ± 3.6 ^b
LDL-C ⁵ (mg.dl ⁻¹)	16.2 ± 2.5 ^{bc}	22.1 ± 1.7 ^b	18.7 ± 1.8 ^{bc}	35.9 ± 4.4 ^a	14.0 ± 1.6 ^c	14.4 ± 1.9 ^{bc}	35.9 ± 4.4 ^a	40.4 ± 3.1 ^a	42.4 ± 1.8 ^a	35.9 ± 4.6 ^a	35.9 ± 4.6 ^a	37.7 ± 4.1 ^a
VLDL-C ⁶ (mg.dl ⁻¹)	15.7 ± 1.5 ^{ab}	19.8 ± 5.1 ^a	20.1 ± 5.4 ^a	9.4 ± 1.6 ^b	15.8 ± 1.4 ^{ab}	19.2 ± 2.3 ^a	9.4 ± 1.6 ^b	18.4 ± 3.9 ^{ab}	13.4 ± 2.4 ^{ab}	12.0 ± 2.4 ^{ab}	12.0 ± 2.4 ^{ab}	12.7 ± 4.0 ^{ab}
Triglycerides (mg.dl ⁻¹)	136.3 ± 9.3 ^{ac}	164.4 ± 19.2 ^a	164.6 ± 22.3 ^a	109.3 ± 11.0 ^b	132.3 ± 10.7 ^{ab}	157.0 ± 16.5 ^a	109.3 ± 11.0 ^b	120.0 ± 6.7 ^{bc}	118.7 ± 5.9 ^{bc}	119.2 ± 7.6 ^{bc}	119.2 ± 7.6 ^{bc}	116.3 ± 9.7 ^{bc}
HDL-C/LDL-C	5.14 ± 0.39 ^{ab}	4.21 ± 0.28 ^b	4.37 ± 0.36 ^b	1.99 ± 0.10 ^c	5.08 ± 0.38 ^{ab}	5.96 ± 0.59 ^a	1.99 ± 0.10 ^c	1.90 ± 0.13 ^c	1.69 ± 0.12 ^c	2.03 ± 0.28 ^c	2.03 ± 0.28 ^c	2.02 ± 0.20 ^c
TC/TG	0.82 ± 0.06 ^b	0.84 ± 0.07 ^b	0.75 ± 0.06 ^b	1.09 ± 0.14 ^a	0.76 ± 0.06 ^b	0.75 ± 0.04 ^b	1.09 ± 0.14 ^a	1.15 ± 0.12 ^a	1.08 ± 0.06 ^a	0.98 ± 0.06 ^{ab}	0.98 ± 0.06 ^{ab}	1.09 ± 0.10 ^a
Atherogenic index ⁷	0.41 ± 0.03 ^c	0.45 ± 0.05 ^c	0.49 ± 0.07 ^c	0.64 ± 0.04 ^b	0.44 ± 0.02 ^c	0.41 ± 0.03 ^c	0.64 ± 0.04 ^b	0.78 ± 0.02 ^a	0.79 ± 0.04 ^a	0.72 ± 0.05 ^{ab}	0.72 ± 0.05 ^{ab}	0.69 ± 0.02 ^{ab}

¹Values are means ± SEM; ²Means within a row with different superscripts are significantly different ($p < 0.05$); ³TC: Total cholesterol; ⁴HDL-C: High-density lipoprotein cholesterol; ⁵LDL-C: Low-density lipoprotein cholesterol; ⁶VLDL-C: Very low-density lipoprotein cholesterol; ⁷Atherogenic index = (TC - HDL-C) / HDL-C [31].

Table 5
Orthogonal polynomial trend analysis of studied variables of rats fed Khalal or Tamr date palm fruit for 6 weeks

Variable	Cholesterol-free groups				Cholesterol-supplemented groups			
	Khalal		Tamr		Khalal		Tamr	
Trend ¹	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic
Weight gain	0.102	0.001	0.052	0.015	0.009	0.018	0.024	0.010
Food intake	0.236 ^{*(A)}	0.043	0.013	0.034	0.039	0.016	0.011	0.007
FER ²	0.000	0.023	0.053	0.001	0.000	0.014	0.029	0.006
Total cholesterol	0.015	0.166	0.009	0.164	0.044	0.076	0.024	0.009
HDL-C ³	0.000	0.249 [*]	0.007	0.245 [*]	0.002	0.033	0.009	0.028
LDL-C ⁴	0.039	0.180	0.024	0.017	0.114	0.006	0.006	0.002
VLDL-C ⁵	0.032	0.008	0.107	0.031	0.049	0.106	0.042	0.005
Triglycerides	0.076	0.024	0.078	0.050	0.041	0.022	0.017	0.019
HDL-C/LDL-C	0.128	0.086	0.093	0.040	0.182	0.010	0.001	0.001
TC/TG	0.037	0.031	0.051	0.004	0.001	0.016	0.000	0.049
Atherogenic index ⁶	0.085	0.001	0.002	0.047	0.348 ^{** (A)}	0.087	0.054	0.062

¹ Values are coefficients of determination (r^2); $^*p < 0.05$; $^{**}p < 0.01$; ^(A) Ascending; ² FER: Food efficiency ratio, body weight gain (g)/100 g food intake; ³ HDL-C: High-density lipoprotein cholesterol; ⁴ LDL-C: Low-density lipoprotein cholesterol; ⁵ VLDL-C: Very low-density lipoprotein cholesterol; ⁶ Atherogenic index = (TC - HDL-C)/ HDL-C [31].

4. Discussion

The energy and macronutrient content of the date palm fruit used in this study was comparable to the documented range values [16, 33]. Khalal was found to contain higher water and lesser energy and carbohydrate than Tamr, the result which is consistent with the changes that may occur during maturation. Khalal, the mature full coloured, represents an early stage of maturity, whereas Tamr, the hard raisin-like, represents the late stage of maturity [16]. It is known that moisture content decreases as the fruits become mature, increasing contents of energy and carbohydrate [33]. However, relative variability in the composition of the date palm fruit has been reported [16, 33]. This variability of can be attributed to a number of factors, such as differences in genotype, maturity stage, product quality, postharvest handling and storage conditions and methods of analysis [16, 27].

Cholesterol has been widely used in animals to induce atherogenic states [34, 35]. Consistently, after 6 weeks of cholesterol feeding, an atherogenic state was established in this study. This is evident as LDL-cholesterol elevation was paralleled by marked rise in total cholesterol/ triglyceride ratio and atherogenic index and a fall in triglyceride concentration and HDL-cholesterol/ LDL-cholesterol ratio, though total cholesterol concentration was not affected. Cholesterol had some reducing effect on HDL-cholesterol and VLDL-cholesterol concentrations, but this effect did not reach statistical significance. This suggests impaired lipid metabolism in cholesterol-fed rats. The present findings regarding the influence of cholesterol feeding on LDL-cholesterol, HDL-cholesterol and triglycerides are in close agreement with those of several reports [31, 36]. However, total cholesterol concentration has been shown to increase [35, 36] or remain unchanged [37] as a result of cholesterol feeding in animals. In fact, this may reflect differences in experimental protocols and procedures, particularly animal model, diet composition and duration of feeding. There is also a general agreement regarding the lack of influence of cholesterol on food intake, weight gain and food efficiency ratio [31, 37], a matter that is consistent with the results of the current study.

To the best of our knowledge, this study is perhaps the first demonstration that links whole date palm fruit consumption with lipid parameters in rats. We examined the effect of feeding of Birhi date palm fruit on serum lipid and lipoprotein concentrations in the cholesterol-fed rat. This model is known to have abnormal lipid profile and functional atherosclerosis [2, 35]. Fresh date palm fruit was used, preserving thereby the possible bioactive constituents [29]. The fruit was incorporated into isocaloric and isonitrogenous diets at three levels, 0%, 5% and 10%, and fed *ad libitum* for 6 weeks. Male rats were used and changes in body weight and food intake were also considered. Surprisingly, none of the concentrations of serum lipid and lipoprotein fractions were significantly influenced. Khalal

was an exception which increased atherogenic index. This effect was also indicated by the ascending linear trend response. It should be noted that food intake, weight gain and food efficiency ratio were normal during this study, eliminating the possibility of inadequate dietary intake and nutrient imbalance that might affect the results. The exact reason or mechanism responsible for these findings is not clear. However, the following discussion will focus on the available literature.

To date, the literature on the influence of date palm fruit on serum lipid status in humans is scarce. We found only two human studies with conflicting results that have investigated this aspect in healthy individuals. It has been reported that the consumption of 100 g/day by 10 healthy volunteers, for a period of 4 weeks of either Medjool or Hallawi variety of date palm fruit decreases serum triglyceride concentration, without affecting concentrations of total cholesterol, LDL-cholesterol, VLDL-cholesterol and HDL-cholesterol [19]. In the other study, in contrast to Ghars, Tamesrit variety of date palm fruit has been shown to reduce serum LDL-cholesterol concentration in 52 normal individuals ingesting 70 g/day of either variety for 21 days, with no influence on serum concentrations of total cholesterol, HDL-cholesterol and triglycerides and ratio of HDL-cholesterol/ LDL-cholesterol [20]. Apparently, some of these reported results are consistent with our findings. However, a remarkable controversy exists. These previous studies involved healthy subjects with normal lipid profile and reported no nutritional or dietary information. Evidently, we were unable to show an effect of date palm fruit on LDL-cholesterol and triglyceride concentrations in normal and hyperlipidaemic rats. In fact, the amount, duration, variety and maturity stage of date palm fruit besides other lifestyle factors, such as energy intake and basal diet composition, as well as initial serum lipid concentrations, are among many potential confounders that may contribute to this inconsistency.

In this context, there are several animal studies investigating different preparations of date palm, but not the fruit. Lipid profile improvement has been repeatedly reported. Feeding rats with diets containing defatted date palm seed flour at 1.5%, 2.5% and 5.2% has been shown to decrease plasma concentrations of total cholesterol, LDL-cholesterol and triglycerides [23]. Reduced plasma total cholesterol, LDL-cholesterol and triglyceride concentrations and increased HDL-cholesterol concentration have been documented in rats fed diets supplemented with 2.5% date palm seed fibers [21]. When date palm pollen grains were incorporated into the diet of rats at 0%, 2% and 4% [22] or administered orally to rabbits at 30, 60 and 90 mg/kg/day [25], a significant improvement of the lipid profile has been reported. Similar improvement has been shown in streptozotocin-induced diabetic rats given date palm leaves extracts [24]. Among the many problems in these studies is that they did not specify the date palm variety used. Further, different parts of the date palm plant excluding the fruit, with various processing and extraction procedures have been used. These limit the comparison of the current results with those of the other studies.

5. Conclusion

Taken together, Birhi date palm fruit of Khalal or Tamr appear to exert a minimal effect on lipid profile and are ineffective to counteract the atherogenic effect of cholesterol in rats. However, because of the scarcity of studies on humans and animals, the significance of the present hypothesis demands further investigations.

Competing interests

The authors declare that they have no competing interests.

Authors' contributions

The authors contributed sufficiently to the study design, experimentation, analyses, statistics and writing and editing the manuscript.

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