REGULAR ARTICLE

In vitro assessment of nutritive value of date palm by-products as feed for ruminants

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ABSTRACT

Date-palm leaves, pedicels, date-pits and waste dates are date-palm by-products (DPBP), routinely used as a feed resource by oasis farmers. This work aims to assess *in vitro* the chemical composition and nutritional value of these DPBP, including waste dates from three varieties (Bouarus, Harchaya and Kentichi). Vetch-oat hay was used as a reference feed material. Palm leaves showed the highest NDF, ADF, lignin and crude protein contents (609, 435, 84, 64 g kg^{-1} DM, respectively). Cumulative gas production at 144 h of incubation was greatest for Kentichi dates (330 mL g^{-1}DM) and lowest for date-pits (69 mL g^{-1}DM). Regardless of the variety, waste dates showed the highest DM effective ruminal degradability (ED = 0.48-0.49; estimated for a passage rate of 0.03 h^{-1}) and organic matter digestibility (OMD), whereas date-pits seemed to be a poorly degradable material, with an ED coefficient of 0.09 g g^{-1} DM. The lowest total VFA production from ruminal fermentation was with date-pits and the highest with waste dates of the Bouarus variety. These results suggest that waste dates, albeit their low protein content, should be considered highly digestible with energy concentrations as high as that of vetch-oat hay. Palm leaves and pedicels can be considered as highly fibrous emergency roughages for low-producing animals. When DPBP are used as feedstuffs, protein supplements will be necessary to balance the ruminant diets.

Keywords: By products; Chemical composition; *In vitro* fermentation; Nutritive value; Rumen

INTRODUCTION

Animal feeding represents one of the highest production costs in livestock farming, especially in arid regions (e.g., Algeria). Production of food for humans and feed for animals is constrained by the shortage of water that limits the utilization of arable land (Osman et al., 2005). The production of forages decreases severely during the dry season, extended in these areas for 6-7 months (from May to October). Ruminants farmed in these regions may be handicapped to cover their nutritional requirements with only natural vegetation, if this is not supplemented with an additional feed, with the result of a reduced performance. Under these conditions, the use of date palm by-products (DPBP) in animal diets could counteract in part the shortage of animal feed resources and subsequently increase milk and meat production.

The date palm is a hardy monocot tree acclimated to arid and desert environments, with a hot and dry climate. This tree, which constitutes the backbone of the oasis agriculture, is an important source of food for the Saharan population and feed for their livestock and offers a wide range of agricultural products (fiber, building material) and benefits (shade) traditionally used for domestic purposes. Waste pulp, discarded fruits and seeds (pits or stones) are the three major byproducts derived from the date fruit processing. Fruits of inferior quality or low-grade that are not intended for human consumption (discarded) may be used as feed, and in the current study will be referred to as waste dates. Palm leaves and pedicels are obtained during the fruit harvesting. DPBP have been used historically in animal nutrition by local farmers in a traditional way (Chehma and Longuo, 2004) and can be a significant source of feed (El Hag, 1995). Dates are rich in carbohydrates

(70–80%), mostly sugars, and thus show a high energy content. Although highly fibrous, date-pits have been considered an alternative feedstuff that may be included up to 50-75% in diets for ruminants (Hossain et al., 2014). Along with their nutrient and energy content, date fruits and seeds are rich in some plant secondary compounds with recognized pharmacological properties and positive effects on the consumer (Al-Orf et al., 2012).

Waste dates are conventionally used as supplementary feed for livestock in southern Algeria (Boudechiche et al., 2008). Waste dates are a rich in carbohydrates, dietary fiber and minerals, with lower contents in lipids and protein (Sidhu, 2006). Easily digestible sugars (fructose and glucose) are the main carbohydrates in dates (Al-Farsi et al., 2005; Myhara et al., 1999). Because of their composition, waste dates are highly palatable and digestible feedstuffs (Al-Dobaid et al., 2009), and may be a valuable source of energy to be used as suitable supplements to low quality roughages. There are some reports on the chemical composition and nutritive value of dates, but the available data on the potential value of DPBP as feeds are rather limited (Dayang et al., 2014). Not only waste dates, but also pedicels and dry palm leaves are used as feedstuffs for ruminants. In particular, there is scarce information on the rate and extent of fermentation of these by-products in the rumen.

The *in vitro* methods for feed evaluation have numerous advantages over *in vivo* methods (López, 2005). They are less expensive, less time consuming and allow incubation to be maintained more precisely than *in vivo*. In addition, *in vitro* techniques utilize small amounts of test feeds making them applicable to screening feeds that are not available in sufficient quantity for *in vivo* experiments.

The objective of this study was to evaluate, *in vitro*, the ruminal fermentation and nutritive value of dry palm leaves, pedicels, date-pits and waste dates from three different varieties. Vetch-oat hay was used as a reference material.

MATERIALS AND METHODS

Forage and roughage material
Palm leaves, pedicels, date-pits (taken from Deglet-Nour tree) and waste dates from three different varieties (Bouarus, Harchaya, Kentichi) were harvested by hand in Biskra, in the Saharan Atlas region of Algeria (34° 51.01 ' N, 04° 43.40 ' E). Mature waste dates of uniform size, free of physical damage and injury from insects and fungal infection, were selected and used for all experiments. Between twenty and thirty specimens of each palm leaves, pedicels and date-pits were collected to obtain a representative aliquot biomass. Vetch-oat hay, used as a

control reference, was collected from the ITELV (Technical Institute of Breeding, Ain Mlila, Algeria). Samples and the reference forage from the different specimens were pooled, oven-dried at 50 °C (Makkar, 2003), and subsequently ground to pass a 1 mm screen.

Chemical analysis
Dry matter (DM, method ID 934.01), ash (method ID 942.05) and crude protein (CP, method ID 954.01) contents were determined following the methods of AOAC (2006). Neutral and acid detergent fibre (NDF and ADF, respectively) and sulphuric acid detergent lignin (ADL) were determined with an ANKOM 220 fibre analyser as described by Mertens (2002), AOAC (2006; Official method 973.18) and Robertson and Van Soest (1981), respectively. All fibre fractions were expressed including residual ash. As ether extract was not analyzed in the present experiment, the non-fiber carbohydrate (NFC) content of feeds was calculated by subtraction of CP, NDF and ash from total DM (Sniffen et al., 1992). The fat content of DPBP is usually very low (<1%) except for date pits in which fat may represent up to 12% (Sidhu, 2006).

Volatile fatty acids (VFA) in the medium after 24 hours of incubation were determined by gas chromatography in an Agilent 6890 apparatus (Agilent Technologies España S. L., Madrid, Spain) fitted with a capillary column (HP-FFAP Polyethylene Glycol TPA-Treated, 30 m x 530 μm i.d.x 1 μm film thickness).

Animals and rumen fluid extraction for *in vitro* studies
Rumen fluid was extracted from adult Rasa Aragonesa ewes (average weight 71.5 ± 0.87 kg) fitted with a permanent ruminal cannula (50 mm internal diameter). Animals were daily fed 600 g lucerne hay (167 g CP, 502 g NDF, 355 g ADF and 71 g ADL kg^{-1} DM) and 300 g barley straw (31 g CP, 733 g NDF, 522 g ADF and 108 g ADL kg^{-1} DM), and had free access to water and mineral/vitamin block. Samples of rumen contents (ca. 300 mL from each sheep) were withdrawn prior to morning feeding, transferred into thermos flasks and taken immediately to the laboratory, where rumen fluid was strained through various layers of cheesecloth and kept at 39 °C under a constant flow of CO_2.

In vitro gas production kinetics
Gas production profiles were obtained using the technique described by Theodorou et al. (1994) slightly modified. Ground samples (500 mg) were incubated in 50 mL of diluted rumen fluid (10 mL mixed rumen fluid + 40 mL medium prepared under a CO_2 constant flow) in 120 mL serum bottles. The composition of the incubation medium was that described by Theodorou et al. (1994). Incubations were performed using three different inocula (rumen fluid from three sheep used separately) with two bottles per

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rumen fluid inoculum (for a total of six observations –three replicates– per sample). Two runs were carried out and six bottles (three per run) containing only diluted rumen fluid were incubated as blanks and used to compensate for gas production in the absence of substrate. Once filled up, all bottles were closed with rubber stoppers, crimped with aluminium seals, shaken and placed in an incubator at 39ºC. Volume of gas produced was recorded at 3, 6, 9, 12, 16, 21, 26, 31, 36, 48, 72, 96, 120 and 144h after inoculation with an HD2124.2 manometer provided with a TP804 pressure gauge (DELTA OHM, Caselle di Selvazzano, Italy). Pressure readings were converted into volume by using a pre-established linear regression between pressure recorded in the same type of bottles and known inoculated gas volumes (Marinas et al., 2003). At the end of the incubation (after 144 h), the contents of each serum bottle were filtered using sintered glass crucibles (coarse porosity no. 1, pore size 100–160 μm) under vacuum. Then the residue was washed out with distilled water and oven-dried at 100ºC for 48 h to estimate the potential DM disappearance (D144, g g^{-1} DM).

Calculations

In order to estimate the fermentation kinetic parameters, gas production data were fitted using the exponential model proposed by France et al. (2000):

$$G = A \left[1 - e^{-c(t-L)} \right] \text{ for t} \geq \text{L},$$

where G (mL/g) denotes the cumulative gas production at time t; A (mL g^{-1}) is the asymptotic gas production; c (h^{-1}) is the fractional rate of substrate fermentation and L (h) is the lag time. Volume of gas (mL g^{-1} DM) produced after 24 h of incubation (G24) was used as an index of digestibility and energy feed value, as suggested by Menke and Steingass (1988). According to France et al. (2000), the extent of degradation (effective degradability) in the rumen (ED, g g^{-1} DM) for a given rate of passage (k, h^{-1}) was estimated as:

$$ED = \frac{c \times D144}{c + k} e^{-kL},$$

where $D144$ is the dry matter disappearance after 144 hours of incubation. To calculate ED, a rate of passage of 0.03 h^{-1} (characteristic of a sheep fed a forage diet at maintenance level) was used.

The energy value and digestibility of feedstuffs were calculated from the amount of gas produced at 24 h of incubation with supplementary analyses of crude protein and ash, as follows (Menke and Steingass, 1988).

$$\text{ME (MJ/kg DM)} = 2.2 + 0.136 \times \text{G24} + 0.057 \times \text{CP} + 0.029 \times \text{CP}^2$$

$$\text{OMD (\%)} = 14.88 + 0.889 \times \text{G24} + 0.45 \times \text{CP} + 0.0651 \times \text{ASH}$$

Where
ME is the metabolizable energy
G24 is 24 h net gas production (mL/200 mg DM)
CP is crude protein (% of DM)
OMD is organic matter digestibility
ASH is ash (% of DM).

Statistical analysis

One way analysis of variance (Steel and Torrie, 1997) was performed on gas production fermentation kinetics, metabolizable energy, organic matter digestibility and volatile fatty acids data, with DPBP as the only source of variation (fixed effect) and source of inoculum (random effect) as a blocking factor. Tukey's test was used for the multiple comparison of means (p<0.05). Analysis of variance and correlation between different variables were performed using SAS software package (SAS Institute, 2008).

RESULTS

Chemical composition

The chemical composition of the DPBP is shown in Table 1. All DPBP had low CP contents (<7%) which in waste dates ranged from 18 g kg^{-1} DM in Kentichi to 40 g kg^{-1} DM in Harchaya. Protein content was greater in date-pits (53 g kg^{-1} DM) and palm leaves (64 g kg^{-1} DM) than in waste dates.

The ADF:NDF ratio was higher in palm leaves (0.71) and pedicels (0.64) than in vetch-oat hay (0.57). This result indicates the high cellulose and/or lignin content of the two DPBP. Based on their chemical composition, palm leaves, pedicels and date-pits could be classified as highly fibrous feeds with relatively medium protein content.

Table 1: Chemical composition (g kg^{-1} dry matter) of various date palm by-products and of the vetch-oat hay

Date palm by-products	DM (g kg^{-1} feed)	OM	CP	NDF	ADF	ADL	NFC
Leaves	963	848	64.4	609	435	83.8	137
Pedicels	942	938	35.6	557	358	79.0	288
Date-pits	977	989	52.5	636	410	56.9	277
Bouarus waste dates	954	949	27.5	79	45	22.9	796
Harchaya waste dates	941	960	40.4	87	49	26.5	773
Kentichi waste dates	958	964	17.7	93	47	23.6	811
Vetch-oat hay	929	937	53.8	579	331	46.1	233

DM: Dry matter; OM: Organic matter; CP: Crude protein; NDF: Neutral detergent fibre expressed with residual ash; ADF: Acid detergent fibre expressed with residual ash; ADL: Acid detergent lignin expressed with residual ash, NFC; Non-fiber carbohydrate

In vitro gas production kinetics

Data of *in vitro* fermentation kinetics are shown in Table 2. The lowest values of gas production, D144 and ED were observed for palm leaves, date-pits and pedicels (being particularly low for date-pits), whereas waste dates had significantly higher values. Similar trends were observed for the *in vitro* fermentation kinetics estimated from the gas production curves.

Fig. 1 shows the cumulative gas production profiles of the seven different samples that were incubated in buffered rumen fluid. For waste dates, pedicels and vetch-oat hay fermentation started readily without lag time, whereas for
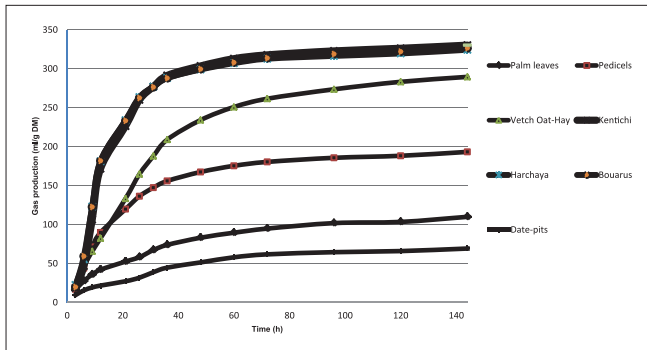

Fig 1. Cumulative gas production profiles for date palm by-products during *in vitro* incubation

date pits and palm leaves there was some lag before any volume of fermentation gas could be measured.

The highest asymptotic gas production was observed in Kentichi, Harchaya and Bouarus dates (330, 327, 324 mL g^{-1} DM, respectively), whereas date-pits recorded the lowest value (69 mL g^{-1} DM).

Energy content and organic matter digestibility

The estimated metabolizable energy (ME), and organic matter digestibility (OMD) are presented in Table 2. The ME contents were particularly higher in all waste dates, while palm leaves and date-pits had significantly lower values of ME (5.43 and 4.12 MJ kg^{-1} DM; respectively). The OMD also differed significantly among DPBP (p<0.001), being greater with waste dates and lower with date-pits. The OMD of pedicels was significantly higher than that of palm leaves, but lower than that of waste dates.

Fermentation end-products

There were differences (p<0.001) among DPBP in total and individual VFA concentration and acetate to propionate ratio after 24 h of incubation (Table 3). The lowest total VFA concentration was with date-pits (21.1 mmol L^{-1}) and the highest with Bouarus waste dates (42.7 mmol L^{-1}), whereas Kentichi waste dates showed an intermediate value (32.5 mmol L^{-1}). The concentration of acetate (major fatty

Table 2: *In vitro* fermentation kinetics (estimated from gas production curves) and predicted metabolizable energy (ME) content and organic matter digestibility (OMD) of date palm by-products and of the vetch-oat hay

Date palm by-products	A (mL g^{-1} DM)	*c* (h^{-1})	G24 (mL g^{-1} DM)	D144 (g g^{-1} DM)	ED (g g^{-1} DM)	ME (MJ kg^{-1}DM)	OMD (%)
Leaves	105d	0.035c	59.8d	0.311d	0.162d	5.43e	29.6d
Pedicels	188c	0.049b	130c	0.387c	0.233c	6.55d	41.4c
Date-pits	69.9e	0.028d	33.9e	0.187e	0.087e	4.12f	22.7e
Bouarus waste dates	324a	0.057a	242a	0.776a	0.494a	9.67b	62.8a
Harchaya waste dates	322a	0.058a	242a	0.762a	0.487a	9.96a	63.1a
Kentichi waste dates	331a	0.052b	236a	0.786a	0.484a	9.55b	62.7a
Vetch-oat hay	293b	0.032cd	155b	0.596b	0.296b	7.79c	46.7b
S.E.M.	9.96	0.0038	8.48	0.0225	0.01294	0.181	1.20

A: Asymptotic gas production, *c*: Fractional rate of fermentation; G24: Gas production at 24 h of incubation; D144: DM disappearance after 144 h of incubation; ED: Extent of degradation for a fractional passage rate of 0.03 h^{-1}; S.E.M.: Standard error of the mean, [a-g]Means in a column with different superscripts are significantly different (P<0.05)

Table 3: Total (mmol L^{-1}) production of volatile fatty acids, molar proportions (%), and acetate to propionate ratio (A:P) after 24 h of *in vitro* incubation of date palm by-products

Date palm by-products	Total	Acetate	Propionate	Isobutyrate	Butyrate	Isovalerate	Valerate	A:P
Leaves	21.9e	63.7a	23.3c	0.79ab	9.23a	0.77ab	2.25a	2.78a
Pedicels	30.0c	58.4a	28.5b	0.64bc	10.36ab	0.59bc	1.54bc	2.05b
Date-pits	21.1e	66.1a	22.0c	0.84a	7.83bc	0.84a	2.44a	3.07a
Bouarus	42.7a	58.9a	32.4ab	0.38d	7.02c	0.34d	1.03d	1.83bc
Harchaya	26.0d	58.6a	30.6b	0.71abc	7.57bc	0.83a	1.74b	1.95b
Kentichi	32.5c	48.0b	36.5a	0.67abc	12.99a	0.46cd	1.47bc	1.33c
Vetch-oat Hay	36.2b	65.0a	34.0ab	0.58c	7.68bc	0.49cd	1.34cd	1.93b
S.E.M.	2.02	5.88	3.50	0.119	1.861	0.154	0.242	0.347

[a-g]Means in a column with different superscripts are significantly different (<0.05), S.E.M.: Standard error of the mean. Bouarus, Harchaya and Kentichi, refer to waste dates of these varieties

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acid) differentiated two groups: Bouarus (25.1 mmol L^{-1}), while the fermentation of palm leaves resulted in a lower acetate concentration (13.9 mmol L^{-1}). The same tendency is shown for the concentrations of propionate that are higher in Bouarus and Kentichi dates and lower in palm leaves and date-pits. Pedicels showed intermediate values of acetate and propionate concentrations (17.6 and 8.6 mmol L^{-1}; respectively).

Waste dates produced the highest molar proportions of butyrate and isobutyrate, whereas the acetate/propionate ratios were significantly different between treatments (p<0.001). Acetate to propionate ratios ranged from 1.33 in Kentichi waste dates to 3.1 in date-pits.

Relationships between chemical composition, and gas production and fermentation products

The CP, NDF, ADF and ADL contents of the feedstuff were negatively correlated (p<0.05) with 9, 21 and 36 h gas production, and with G24 (Table 4). In addition, the fiber fractions were negatively correlated (p<0.01) with D144. On the contrary, the NFC content was positively correlated with gas production at 21, 31 and 144 h, and also with D144 (p<0.05) and G24 (p<0.01).

Extent of degradation (ED) was strongly and positively correlated with *A* (r =0.95, p=0.001) and *c* (r =0.95,

p=0.014), whereas D144 was also positively correlated with *A* (r =0.98, p<0.001) and *c* (r =0.77, p=0.042).

Cell wall fractions (NDF and ADL) were negatively correlated with asymptotic gas production (*A*) and fractional rate of gas production (*c*).

The NDF (p<0.01), ADF (p<0.01) and ADL (p<0.05) were negatively correlated with ME and OMD (Table 4), whereas the NFC fraction was positively correlated with ME (p<0.05), and with OMD (p<0.01).

Positive correlations (Table 5) occurred between CP and ADF contents of the DPBP and the acetate to propionate ratio (p<0.05), and also between 24 h gas production and acetate concentration (p<0.05). Propionate was correlated with ME and OMD (p<0.05). Positive correlations (p<0.05) were observed between propionate concentration and D144 (r=0.82) or ED (r=0.78) of DPBP.

DISCUSSION

The DPBP studied herein are commonly used as feedstuffs for small ruminants farmed in Mediterranean rangelands (Kababya et al., 1998). Dates are used as fresh fruits for human consumption, and date by-products are used to

Table 4: Correlation coefficients between chemical composition parameters (g kg⁻¹ DM), volume of *in vitro* gas production, gas production kinetics parameters, and predicted metabolizable energy (ME; MJ kg⁻¹ DM) and organic matter digestibility (OMD), obtained for date palm by-products

| | Gas production (mL g⁻¹ DM) | | | | Gas production constants | | | | | Energy value | |
	9 h	21 h	36 h	144 h	A	c (h⁻¹)	ED	D144	G24	ME	OMD (%)
CP	−0.75*	−0.78*	−0.76*	0.69	−0.68	−0.78*	−0.75*	−0.69	−0.77*	−0.70	-0.76*
NDF	−0.93**	−0.94**	−0.89**	−0.82*	−0.82*	−0.88**	−0.95**	−0.89**	−0.77	−0.91**	-0.92**
ADF	−0.95**	−0.96**	−0.93**	−0.87*	−0.87*	−0.86*	−0.97***	−0.93**	−0.95**	−0.94**	-0.93**
ADL	−0.78*	−0.83*	−0.84*	−0.81*	−0.82*	−0.57	−0.85*	−0.86**	−0.83**	−0.82*	-0.83*
NFC	0.89**	0.91**	0.86*	0.78*	0.78*	0.85*	0.92**	0.86**	0.89**	0.86*	0.88**

A: Asymptotic gas production (mL g⁻¹ DM); ED: Extent of degradation for a fractional passage rate of 0.03 h⁻¹; D144: DM disappearance after 144 h of incubation (g g⁻¹ DM); G24: Gas production at 24 h of incubation (mL g⁻¹ DM); CP: Crude protein; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ADL: Acid detergent lignin; c: Fractional rate of gas production (h⁻¹); NFC: Non-fiber carbohydrates. *P<0.05, **P<0.01, ***P<0.001

Table 5: Correlation coefficients between volatile fatty acids (mmol L⁻¹) production, chemical composition (g kg⁻¹ DM), *in vitro* gas production kinetics parameters, and predicted metabolizable energy (ME; MJ kg⁻¹DM) and organic matter digestibility (OMD), obtained for date palm by-products

| | DM | Chemical composition | | | | Gas production kinetics parameters | | | | | Predicted ME and OMD | |
		CP	NDF	ADF	ADL	A	c (h⁻¹)	ED	D144	G24	ME	OMD (%)
Total	−0.49	−0.58	−0.49	−0.55	−0.56	0.75*	−0.46	0.65	0.69	0.48	0.67	0.68
Acetate	−0.47	−0.31	−0.29	−0.35	−0.39	0.55	0.29	0.45	0.49	0.81*	0.47	0.47
Propionate	−0.49	−0.71	−0.64	−0.69	−0.68	0.86*	0.56	0.78*	0.82*	0.22	0.79*	0.81*
Isobutyrate	−0.13	−0.45	−0.13	−0.18	−0.19	0.28	0.04	0.18	0.22	0.58	0.19	0.22
Butyrate	−0.20	−0.82*	−0.48	−0.50	−0.37	0.59	0.49	0.56	0.57	−0.10	0.54	0.59
Isovalerate	−0.14	0.30	0.06	0.07	0.14	−0.17	0.07	−0.18	−0.17	−0.75	−0.09	−0.11
Valerate	0.82*	0.37	0.50	0.55	0.44	−0.78*	−0.63	−0.69	−0.72	−0.89**	−0.76*	−0.74
A:P	0.57	0.80*	0.73	0.78*	0.66	−0.92**	−0.71	−0.87*	−0.89**	0.69	−0.88**	−0.90*

DM: Dry matter; CP: Crude protein; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ADL: Acid detergent lignin; A: Asymptotic gas production (mL g⁻¹ DM); c: Fractional rate of gas production (h⁻¹); ED: Extent of degradation for a fractional passage rate of 0.03 h⁻¹; D144: DM disappearance after 144 h of incubation (g g⁻¹ DM); G24: Gas production at 24 h of incubation (mL g⁻¹ DM); A: P. Acetate to propionate ratio. *P<0.05, **P<0.01, ***P<0.001.

produce juice, syrup, jam, jelly or fermented beverage (Chandrasekaran and Bahkali, 2013). Discarded fruits (waste dates) from sorting, grading, storage or processing are often used as animal feed (El-Hadrami, 2012). These by-products are available when fodder yield from pastures is decreased, and may represent a interesting supplement to the herbaceous vegetation (Delillis and Fontanella, 1992). DPBP are more important when the availability of grass declines.

The value of DPBP as feeds is limited by their low CP content (Wilson, 1977). The protein content in pits from seven Saudi date varieties ranged from 4.8 to 7.5 % (Al-Showiman, 1990). Mature date fruits usually contain less than 3% protein (Al-Orf et al., 2012). The CP contents of DPBP found in the present work for waste dates were in agreement with those reported by Longuo et al. (1989), Chehma and Longuo (2004) and Genin et al. (2004) who reported a low nitrogen content (4.5%) for waste dates of deglet noor variety. The CP content of DPBP was thus below the level required (70-80 g CP/kg DM) for optimum rumen function (Van Soest, 1994; Boufennara et al., 2012). Norton et al. (1994) concluded that such a low dietary N supply is not sufficient to ensure an optimal metabolic activity and growth of ruminal microbiota.

The cell wall (NDF, ADF and lignin) contents of waste dates are in agreement with el-Shazly et al. (1963) and Al-Ani et al. (1991) but not with Genin et al. (2004) and Al-Farsi et al. (2007). Chemical composition of palm leaves was similar to that reported by Pascual et al. (2000), but the NDF was lower than that mentioned by Genin et al. (2004). Such inconsistency in the chemical composition of DPBP may be due to the type and origin of the parent date fruit and the extraction technique used in date palm processing. Other factors (soil characteristics, date palm tree variety or age) may cause variations in the chemical composition of date fruits. The contents of NDF, ADF and CP in date-pits were similar to the results obtained by Genin et al. (2004). Date pits are the only DPBP relatively rich in fat (5-12%), and are very rich in dietary fibre (Hossain et al., 2014). The high level of fibre content in palm leaves, pedicels and date-pits could be related in part to the climatic and local conditions prevailing in the area. Cell wall content in plants and fruits is normally increased under hotter and drier conditions (Pascual et al., 2000). Concerning the vetch-oat hay used as reference, its chemical composition was similar to that reported by Hadjigeorgiou et al. (2003) for a range of grass hays.

The use of the *in vitro* gas production methodology to estimate digestion of feeds is based on the well established relationship between the feed digestibility and *in vitro* gas production, in combination with the feed chemical composition (Menke and Steingass, 1988; López, 2005). The *in vitro* gas production system aids to better assess nutrient utilization, and its accuracy in describing digestibility in animals has been extensively validated (López, 2005). Since the utilization of roughages is largely dependent upon microbial fermentation within the rumen, description of roughages in terms of their degradation characteristics would provide a useful basis for their evaluation.

In the current experiment, the highest cumulative gas production after 144 hours of incubation was observed for Kentichi waste dates (330 mL g^{-1} DM) and the lowest was obtained with date-pits (69 mL g^{-1} DM). The high cell wall and lignin contents in date-pits could have accounted for the limited substrate degradation and fermentation, and consequently for the low gas production observed.

The profile of fermentation of the substrates is a characteristic of the fibrous substrates which determine both the rates of degradation and passage in the rumen. Therefore, in order to maximize the nutrient supply from palm leaves, long residence times in the rumen may be required which may allow the rumen microbes to attach and degrade the fiber that may otherwise be resistant to microbial enzymes (Salawu et al., 1999).

Gas production from waste dates was clearly higher than that from conventional forages, such as mountain herbage (Andrighetto et al., 1992), pasture hay (Gulsen et al., 2004), or alfalfa hay (Lantcheva et al., 1999). These results suggest that these waste dates can be used as feeds for ruminants. Waste dates are rich in soluble sugars, vitamins and minerals, and consequently have been used as fermentation substrate for the production of antibiotics (Sidhu, 2006) or other biotechnological processes based on microbial fermentation (Chandrasekaran and Bahkali, 2013). Our results suggest that these waste dates would be also a highly fermentable feedstuff in the rumen that could represent a substantial supply of energy to the animal. Adequately combined with a source of degradable N, it can favor the synthesis of microbial protein in the rumen. Vetch-oat hay is very fermentable due to the association of a legume (*Vicia sativa*) and a grass (*Avena sativa*), which provides degradable nitrogen and fermentable organic matter for an adequate microbial synthesis (Getachew et al., 2004).

The extent of degradation (ED) of DPBP ranged between 0.494 g g^{-1} DM for Bouarus waste dates and 0.087 g g^{-1} DM for date-pits. The high values obtained with waste dates could be explained by the low levels of cell wall fractions NDF, ADF and ADL.

Dry matter disappearance after 144 h of incubation (D144) differentiates two groups. Leaves, pedicels and date-pits

had low values (<39%), whereas waste dates showed a higher potential rumen degradation (>70%), confirming the high energetic value of these by-products. These results are consistent with lower fibre contents of waste dates in comparison with date-pits, palm leaves and pedicels. Generally, as cell wall (fibre) increases, digestibility and energy content decrease (Van Soest, 1994). Our results were in line with those reported by Mahgoub et al. (2005), Genin et al. (2004) and Dayani et al. (2012), who observed higher digestibility of date pulp with lower cell-wall content.

The low *in vitro* DM digestibility of date palm leaves was in agreement with other published studies (Longuo et al., 1989; Genin et al., 2004; Arhab et al., 2006). Date pits showed significant lower values of D144 than those found by Gihad et al. (1989) in sheep. Highly fibrous DPBP could be treated with chemicals (NaOH) or fibrolytic enzymes to enhance their digestibility (Sidhu, 2006).

The CP content of DPBP was negatively correlated to gas production at 9 h and 36 h, in agreement with data reported by Getachew et al., (2004) for feedstuffs collected from commercial dairy farms. On the other hand, Larbi et al., (1998) noted that the CP content of some shrubs was positively correlated with potential gas production.

Cell wall components (NDF, ADF and ADL) were negatively correlated with gas production at all incubation times, maybe as a result of a reduced microbial activity on highly lignified fibrous substrates. The negative correlation between cell wall content (NDF and ADF) and volume of gas production at all incubation times is consistent with other studies (Larbi et al., 1998) and the negative effect of NDF and ADF on digestibility is in close agreement with Lantcheva et al. (1999). This demonstrates that the effect of NDF and ADF on fermentation becomes less important as the level of NDF and ADF declines. Date by-products may also contain phenolic acids and condensed tannins (Sidhu, 2006). These secondary compounds could limit ruminal degradation of some of the DPBP, but most likely not that of waste dates, which are highly fermentable.

Menke and Steingass (1988) derived equations to predict OM digestibility and ME content of feed from *in vitro* gas production and chemical composition. The ME values of DPBP predicted using these equations were in agreement with those reported by Genin et al. (2004) who mentioned higher *in vitro* digestibility of waste dates with respect to other DPBP.

When feedstuffs are incubated *in vitro*, gas is produced mainly from the fermentation of carbohydrates (Blummel and Orskov, 1993), with a small contribution from the fermentation of protein or fat (Wolin, 1960). Al-Farsi and

Lee (2008) and Sharifi et al. (2015) reported that date fruit consists of 70–88% carbohydrates, most of which is in the form of sugars, mainly glucose, sucrose and fructose. Consequently, date fruits are considered a concentrate source of energy. The variation in the *in vitro* gas production of DPBP was directly related to differences in other fermentation end-products (total VFA production).

A high acetate to propionate ratio is an indication of a more acetogenic fermentation, associated to the activity of fibrolytic bacteria degrading substrates rich in structural carbohydrates (Getachew et al., 2004; Blummel et al., 1999). This was evident in the current experiment from the higher acetate to propionate ratio in date-pits and palm leaves. Similar ratios have also been obtained from other substrates rich in NDF (Getachew et al., 2004). Acetate to propionate ratios of 3.0-4.1 were reported *in vivo* (DePeters et al., 1997) and of 2.0-4.1 *in vitro* (Blummel et al., 1999) for different feedstuffs. With waste dates (less NDF content) the acetate to propionate ratio was decreased, reflecting an increased higher propionate production from the fermentation of non-fibre carbohydrates. According to McDonald et al. (2011), the fermentation of food rich in starch tends to produce more of propionate, whereas the fermentation of fibrous substrate yields more acetate. Acetic and butyric fermentations result in a greater production of gas, whereas the fermentation to propionate only releases gas when the acid is buffered. Thus feeds that produce high amounts of propionate yield relative less fermentation gas per unit of fermented substrate (Beuvink and Spoelstra, 1992).

CONCLUSIONS

The results obtained in the present work allows clustering the studied DPBP in two groups: one including the highly fibrous poor-quality ones (palm leaves and date-pits) and another one including the highly digestible waste dates. However, protein supplementation will be required when waste dates and the other date by-products are included in the diet for ruminants. Palm leaves and date-pits, with high fibre and limited CP content can be considered as roughage bulky feedstuffs for range ruminants when grazing herbaceous material becomes insufficient. Nevertheless, *in vivo* feeding trials would be needed to validate this conclusion.

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Authors contribution

S. B. made a major contribution to the paper and conducted most of the experiments, analyzed the results and was responsible for the manuscript writing. L. B. have contributed in chemical analysis. Z. A. assisted in collecting rumen fluid from sheep and helping in recording volume of gas produced at several incubation times. A. de V. and M. F. were responsible for planning, designing the whole experiments and revising the manuscript. S. L. made a major contribution in consultant assistance, data analysis, presentation and manuscript preparation.

Abbreviations used

A, asymptotic gas production; A:P, acetate to propionate ratio; ADF, acid detergent fibre expressed with residual ash; ADL, acid detergent lignin; *c*, fractional rate of gas production; CP, crude protein; DM, dry matter; DPBP, date palm by-products; D144, dry matter disappearance after 144 h of incubation; ED, effective degradability; G24, cumulative gas production at 24 h; ME, Metabolizable energy; NFC, non-fiber carbohydrate; NDF, neutral detergent fibre expressed with residual ash; OM, organic matter; OMD, organic matter digestibility; VFA, volatile fatty acids.

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