Effect of maturity stage on chemcial composition in sacco degradation and in vitro fermentation of acorns (Quercus coccifera L.)

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Effect of maturity stage on chemical composition, *in sacco* degradation and *in vitro* fermentation of acorns (*Quercus coccifera L.*)

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SUMMARY - The nutritive value of acorns (*Quercus coccifera*) was studied in the forestry region of Bizerte (North of Tunisia). During the period between October and December 2002, samples from 4 trees were harvested every twenty days representing 4 successive maturity stages. For each stage, chemical composition, kinetic of *in sacco* DM degradation and *in vitro* gas production were determined. Acorns were low in ash and nitrogen, which were not influenced by maturity stage. Total cell wall (NDF) decreased significantly from stage 1 to stage 4, stages 2 and 3 were similar. Acid detergent fibre (ADF) was higher in stage 1 (21.9% DM) than in stages 2, 3 and 4, averaging 14.9% DM. The same trend was observed for acid detergent lignin (ADL). Stages 3 and 4 exhibited the highest total DM *in sacco* degradation and total gas production and were the most rapidly fermented *in vitro* (mean value of gas production rate: 0.063 ml/h). It was concluded that the period between stages 3 and 4 seemed to be the most suitable time to harvest acorns for goat feeding.

Keywords: Acorn, *Quercus coccifera*, chemical composition, *in sacco* degradation, *in vitro* fermentation, goat.

RESUME – *"Effet du stade de maturité sur la composition chimique, la dégradation* in sacco *et la fermentation* in vitro *des glands (*Quercus coccifera *L.)". Nous avons étudié la valeur nutritive des glands (*Quercus coccifera*) dans la région forestière de Bizerte (Nord de la Tunisie), durant la période entre octobre et décembre 2002. Des échantillons à partir de 4 arbres ont été prélevés tous les vingt jours, représentant 4 stades de maturité successifs. Pour chaque stade, nous avons déterminé la composition chimique, la cinétique de dégradation de la matière sèche (MS) ainsi que la production de gaz* in vitro*. La teneur en paroi végétale (NDF) a diminué significativement du stade 1 au stade 4, les stades 2 et 3 sont équivalents. La teneur en lignocellulose (ADF) était plus élevée au stade 1 (21,9% MS) qu'aux stades 2, 3 et 4 qui étaient équivalents (en moyenne 14,9% MS). La même tendance a été observée pour la teneur en lignine (ADL). Les valeurs de la dégradabilité totale de la MS ainsi que de la production totale de gaz étaient les plus élevées aux stades 3 et 4. Ces deux stades étaient les plus rapidement fermentés (vitesse de fermentation moyenne 0,063 ml/h). En conclusion, cette étude semble indiquer que la période entre les stades 3 et 4 est la plus favorable pour la récolte des glands destinés aux caprins.*

Mots-clés : *Gland,* Quercus coccifera*, composition chimique, dégradabilité* in sacco*, fermentation* in vitro*, caprins.*

Introduction

In Tunisia a large potential of local non-conventional feed resources is available during the year. In addition to crop residues and by-products, growing interest is shown in some forestry products such as shrub leaves and fruits for small ruminant feeding. Acorns from oak trees, mainly kermes oak (*Quercus coccifera*) and cork oak (*Quercus suber*) are abundant in Tunisian coastal-forestry regions (Ministry of Agriculture, 1995). The maturation of acorns from kermes oak is biennial. They reach their morphological maturity at the end of the autumn and the greatest percentage of trees with fruit is observed in November (Elena-Rossello *et al*., 1993). Acorns are generally harvested by farmers during the period from October to December and given to goats, marketed or traditionally conserved (Kayouli and Buldgen, 2001). Acorns seemed to be a highly energetic resource for small ruminants (Kayouli and Buldgen, 2001) and are often compared to barley (El Jassim *et al.,* 1998). This study aimed to determine the nutritional potential of acorns from kermes oak and the optimum stage of harvesting for feeding animals and conservation.

Materials and methods

Plant material

Acorns *(Quercus coccifera)* were studied in the forestry region of Bizerte (North of Tunisia, humid). Samples from 4 trees were harvested every twenty days representing 4 successive maturity stages, during the period between October and December 2002. Samples from each tree and for each maturity stage were considered independently. Dry matter (DM) was determined at 105°C in a forced-air oven and the samples were dried at 40°C during 48h and then ground to pass through 1 mm screen for chemical analysis and *in vitro* determinations, and through 2 mm screen for *in sacco* measurements.

Animals, diets and measurements

Four adult local goats with rumen cannula (average liveweight: 44.5 kg) were used for *in sacco* and *in vitro* determinations. They were housed in individual pens and received 70 g/kg/$LW^{0.75}$ of a ration composed of 70% of oat-vetch hay and 30% of commercial concentrate on a dry matter (DM) basis twice per day.

The nylon bag technique (Ørskov *et al*., 1980) was used to determine DM degradability in the rumen. The four stages of maturity were incubated sequentially, with 3 successive replications for each stage. About 3 g of acorns (ground at 2 mm) were introduced in nylon bags (50 µm pore size, 15 mg/cm^2) and incubated in the rumen for 3, 6, 12, 24, 36, 48, 72 and 96 h.

Acorn samples (300 mg, ground at 1 mm) were incubated in triplicate in 100 ml glass syringes according to the technique of Menke and Steingass (1988). Gas production was read for 1, 2, 4, 6, 12, 24, 36, 48, 72 and 96 h. For each maturity stage sample, three successive incubations were carried out.

In sacco data and gas productions were fitted using the model of Ørskov and McDonald (1979): $p=a+b(1-e^{-ct})$, where: (i) "p" is the DM degradation or gas production at time t; (ii) "a" is the rapidly DM degradable fraction or the immediate gas production; (iii) "b" is the slowly degraded fraction or gas production; (iv) "c" is the rate of degradation or of gas production; and (vi) "a+b" is the total DM degradation or gas production. Parameters were calculated using the Non Linear procedure (SAS, 1985). The General Linear Model procedure (GLM) with the option of Duncan multiple range was used for statistical analysis of data (SAS, 1985).

Laboratory analysis

Feeds and acorn samples (1 mm screen) were analysed for DM, ash and crude protein (CP) according to AOAC (1984). Neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were analysed as described by Goering and Van Soest (1970). Acorns were analysed for total tannins (Makkar and Goodchild, 1996).

Results and discussion

The effects of maturity stage on chemical composition of acorns are presented in Table 1. Acorns were low in nitrogen and ash, which were not influenced by maturity stage (averaged 3.9 and 2.2% DM respectively). Total cell wall (NDF) decreased significantly ($P<0.001$) from stage 1 (48.6% DM) to stage 4 (33.4% DM). Stages 2 and 3 were similar (averaged 41.4% DM). Acid detergent fibre (ADF) was higher ($P<0.001$) in stage 1 (21.9% DM) than in stages 2, 3 and 4, which were similar (averaged 14.9% DM). The same trend was observed for ADL (8.4 and 5.2 % DM respectively for stage 1 and the average of the other three stages). Globally, chemical composition values are in line with the results of the few studies carried out on acorns from Valonia and kermes oaks (El Jassim *et al.,* 1998) and cork oak (Kayouli and Buldgen, 2001).

Table 1. Effects of maturity stage on chemical composition of acorns (% DM)

	Maturity stage				
	S1	S2	S3	S4	SEM
DM**	49[c]	55.7[b]	56.7[b]	60.3[a]	0.7
Ash	2.2	2.3	2.2	2.2	0.05
CP	4	4	3.9	3.8	0.15
NDF***	48.6[a]	43.7[ab]	39[b]	33.4[c]	1.66
ADF***	21.9[a]	16.2[b]	14.4[b]	14.2[b]	0.75
ADL***	8.4[a]	5.6[b]	4.5[b]	5.5[b]	0.5

[a, b, c] For the same line values with the same letter do not differ significantly; **: P<0.01; ***: P<0.001.

The effects of maturity stage on parameters of DM-*in sacco* degradability are presented in Table 2. The DM rapidly-degraded fraction (a) was high in the four studied maturity stages. This parameter ranged from 31.1% in stage 1 to 37.7% in stage 4 (P<0.01). The highest (P<0.001) DM-slowly degradable fraction (b) was observed in stage 3 (49.1%). The maturity stage did not affect the rate of fraction b degradation (c). The average observed value was relatively low (0.031 h^{-1}). Potentially degradable DM fraction (a+b) was highest (P<0.001) in stages 3 and 4, being similar for both stages (averaged 82.9%). This result is similar to the values found by Kayouli and Buldgen (2001) for *in sacco* DM degradability of acorns from cork oak incubated during 96 h in goats receiving different diets based on shrubs (average value: 79.7%).

Table 2. Effects of maturity stage on parameters of DM *in sacco* degradability

	Maturity stage				SEM
	S1	S2	S3	S4	
a (%)**	31.1[cb]	30.9[c]	34.3[b]	37.7[a]	1.41
b (%)***	39.5[c]	46.5[ab]	49.4[a]	44.4[b]	1.5
c (h^{-1})	0.032	0.027	0.033	0.033	0.004
a+b (%)***	70.6[c]	77.4[b]	83.7[a]	82.1[a]	1.31

[a, b, c] For the same line values with the same letter do not differ significantly. **: P<0.01; ***: P<0.001.

The effects of maturity stage on parameters of *in vitro* gas production are presented in Table 3. The immediate gas production (a) was negative. This may indicate a lag time at the beginning of fermentation, probably related to experimental conditions. The lowest (P<0.01) total gas production (a+b) was observed in stage 1 (55.4 ml). Stage 2, 3 and 4 exhibited similar gas production (averaged 65.5 ml). The rate of fermentation was higher in stage 3 and 4 (averaged 0.063 h^{-1}) than in stages 1 and 2 (averaged 0.042 h^{-1}).

Throughout the experimental period (October to December) acorn cell walls content decreased significantly. This result suggests an increase in digestible cell contents, which may be explained by the accumulation of reserves in the cotyledons during the progress of maturity (Bowersox and Ward, 1968). The same result may explain the increase of DM degradability noted mainly in stage 3 and 4, since degradability and cell wall content are negatively correlated (Van Soest, 1982). In addition to washing losses, the high level of rapidly degraded fraction "a" may be explained by the reserves in the cotyledons composed mainly of degradable carbohydrates (Bowersox and Ward, 1968). The slow and relatively unchanged rate of degradation "c" may express the rate of fibre degradation mainly concentrated in the pericarp and the cup of acorns. The same explanation may be valid in gas production parameters, since the increase of the total gas production and the rate of fermentation registered mainly in stages 3 and 4 is in line with the decrease of ADF and ADL contents of acorns.

Table 3. Effects of maturity stage on *in vitro* gas production parameters

	Maturity stage				SEM
	S1	S2	S3	S4	
a (ml)*	-3.6[a]	-4.1[ab]	-4.28[b]	-4.5[b]	0.28
b (ml)**	59[b]	68.5[a]	71.1[a]	69.7[a]	2.94
c (h^{-1})***	0.04[b]	0.045[b]	0.062[a]	0.064[a]	0.003
a+b (ml)**	55.4[b]	64.5[a]	66.9[a]	65.2[a]	2.76

[a, b, c] For the same line values with the same letter do not differ significantly.
*: P<0.05; **: P<0.01; ***: P<0.001.

Acorns from oak species are often compared to barley. El Jassim *et al.* (1998) studied the usefulness of acorns from kermes oak as an alternative source for growing lambs in order to reduce the feeding cost. Acorns were introduced in concentrates at 25 and 50% levels of DM as substitution for barley. They found the lowest daily gain and a decreased digestibility coefficient of dietary constituent in lambs fed on the 50% acorn diets. Nitrogen retention was also lower in acorn diets. These authors claimed that these results may be related to the presence of antinutritive factors such as polyphenols in acorns. In our study, total tannin content is low (3.2 - 4.4% DM) and acorn intake by small ruminants could involve positive effects mainly in N metabolism. Indeed, condensed tannins at low levels may complex proteins at the pH of the rumen and protect protein from microbial enzymes (Reed *et al.,* 1990) and help in protecting soluble proteins from microbes (Wang *et al*., 1994). This may results in increased supply of dietary amino acids in the small intestine.

Conclusions

In conclusion, the current study has shown that the period between the end of November and the end of December seems to be the best time to harvest acorns. During this time, acorns from oak trees are abundant and should be conserved. The traditional means of conservation (drying in the shade or cooking in traditional oven) need to be studied and other conservation methods may be viewed. The conservation of acorns by silage and N supplementation of acorn-containing diets are currently investigated in our laboratory.

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