Radical scavenging activity and health and risk fatty acid indices of soft goats' milk cheeses

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Abstract. Dairy products are important sources of valuable nutrients. However, it is feared that some of their components, particularly the lipid fraction, may contribute to health problems. In the present research, 21 commercial soft goats' milk cheeses of the three varieties: Sainte-Maure, Feta, and Panela were assessed regarding their radical scavenging activity (RSA), polyphenols content, and cholesterol/saturated-fat (CSI), atherogenic (AI), thrombogenic (TI), and health-promoting (HPI) indexes. Qualitative RSA was higher in Sainte-Maure and Panela than in Feta. However, the best quantitative RSA was recorded for the Feta brand "Bon Rennes" (42%). Total polyphenols content expressed as gallic acid equivalents (GAE) was statistically lower in Panela than in Sainte-Maure and Feta cheese (2.2 vs. 2.97 and 3.07 mg GAE/100 g, respectively). Cholesterol content was higher in Feta and Sainte-Maure than in Panela. The «Mikonos Light» brand Sainte-Maure cheese had notably low values of CSI (1.2), while Feta cheeses averaged 2.72. Based on these results, consumption of Mikonos Light is recommended. «Chateau Blanc» brand is also recommended for its high HPI and low TI. In contrast, «Laclette» is the least recommended brand because of its high cholesterol, and saturated fatty acid (SFA) contents and low HPI. Modification of saturated fatty acids ratio by some means, such as animal feeding, would improve the HPI of the local soft goats' milk cheeses.

Key words: Antioxidant activity, Fuctional foods, Health-promoting index, Polyphenols.

Capacidad de atrapamiento de radicales libres, perfil de ácidos grasos e índices de riesgo y promoción a la salud en quesos suaves de leche de cabra

Resumen. Resumen. Los productos lácteos son fuente de valiosos nutrimentos. Sin embargo, se cree que algunos constituyentes, particularmente su composición lipidica puede tener efectos negativos en la salud. Debido a ello, se evaluaron 21 quesos comerciales de las tres variedades: Sainte-Maure, Panela y Feta en relación a su capacidad de atrapamiento de radicales libres (CARL), polifenoles totales, perfil de ácidos grasos, e índices de colesterol/grasa saturada (ICGS), aterogénico (IA), trombogénico (IT) y promoción a la salud (IPS). Los quesos Sainte-Maure y Panela superaron a Feta en CARL cualitativa, sin embargfo, «Bon Rennes» (Feta), registró la CARL cuantitativa mayor (42%). El contenido de polifenoles totales expresado como equivalentes de ácido gálico (EAG) fue significativamente mayor en las variedades Sainte-Maure y Feta (2.97 y 3.07 mg EAG/100 g), en comparación con el panela (2.2 mg EAG/100 g). El colesterol fue mayor en las variedades Feta y Sainte-Maure que en Panela. Sainte-Maure «Mikonos Light» presentó el ICGS más bajo (1.2), mientras que los Feta presentaron en promedio 2.72. En base a estos resultados, se recomienda el consumo de Mikonos Light debido a su bajo valor de ICGS y colesterol y de «Chateu Blanc» por un alto IPS y un bajo IT. Contrariamente, «Laclette» es el menos recomendable por sus altos valores de IA, IT, colesterol y grasa saturada, y bajo IPS. «Cabrero» presentó un ICGS bajo, sin embargo debido a su alto valor de grasa saturada y IA y bajo valor de

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IPS no es recomendable. Modificaciones en la relación de ácidos saturados:poliinsaturados a través de diversas estrategias de alimentación, podría incrementar el IPS en los quesos de leche de cabra.

Palabras clave: Actividad antioxidante, Alimentos funcionales, Índice de promoción a la salud, Polifenoles

Introduction

The incidence of chronic human illnesses associated with older people is expected to increase in the next decades as they constitute a growing share of the population. On a worldwide scale, the incidence of diseases related to food intake and dietary imbalances, such as obesity, diabetes and coronary heart disease is forecast to increase in young people also. Nowadays, in developing countries these maladies cause 79% of deaths from non-communicable diseases (WHO, 2003). It has been suggested that dietary changes could moderate the impact of such diseases (Bhat and Bhat, 2011). Recommendations include a reduction of saturated fat intake. Further, several indices including cholesterol and fatty acid profile have been developed to characterize foods as promoters or threats to health (Connor et al., 1986; Ulbricht and Southgate, 1991; Chen et al., 2004). Likewise, diets aimed at promoting health that include foods with functional compounds have been widely recommended (Bhat and Bhat, 2011). In this respect, the dairy industry has reacted by increasing contents of functional components during the manufacturing processes and modifying milk properties to retain larger amounts of such compounds (Han et al., 2010). However, the importance of dairy products as functional foods is still debatable. Available information of antioxidant activities and functional properties of dairy products marketed in Mexico is extremely scarce; thus the assessment of their potential benefits to human health is necessary to foster their consumption and to help prevent potential illnesses related to diet (Bhat and Bhat, 2011; Gassi et al., 2012). The objectives of this study were to evaluate goats' milk cheese marketed in Mexico in terms of qualitative and quantitative radical scavenging activity, total polyphenols, profile acids; and cholestrol/saturated-fat, fatty of atherogenicity, thrombo-genicity and healthpromoting indices.

Materials and Methods

Cheese sampling and extraction and qualitative (QLRSA) and quantitative (QRSA) radical scavenging activity determination

Goats' milk cheeses were purchased from retail markets in Mexico City. Each purchase was made on three different days and included all available goats' milk cheese brands and varieties. The batch purchases occurred on the first five days of March, May and July of 2010. Twenty-one goats' milk cheeses were identified and classified into three varieties: Sainte-Maure (15), Feta (5) and Panela (1). The cheeses were stored at -4°C until analyzed. All batches were sampled three times. For each cheese, three consecutive methanolic extractions were performed to quantify the total polyphenols. QLRSA assay was performed using the cheese extracts according to the methodology of Sharma et al. (1998) with some modifications described by Cuchillo et al. (2010a). The QRSA of cheese extracts was determined according to Hatano et al. (1988). The QRSA of each sample was calculated as follows: radical scavenging (%) = ((At₀ - At_{end})/ At₀) x 100, where At₀ is the initial absorbance at time zero and $\operatorname{At}_{\operatorname{end}}$ is the final absorbance after 60 min. Total polyphenol in cheese alcoholic extracts was deter-mined by the Folin-Ciocalteu method described by Taga *et al.* (1984).

Determination of methyl-esters fatty acids (FAME), conjugated linoleic acid (CLA) and health and risk indices (HPI, CSI, AI and TI)

Lipids were dissolved in hexane and sodium hydroxide methanol solutions for saponification. Trans-esterification of cheese fat to methyl-esters was carried out according to method 969.33, AOAC (2003). Cholesterol was determined by the procedure described by Fenton and Sim (1991). Fatty acid concentrations were quantified according to Cuchillo et al. (2010b). Cholesterol/saturated fat index (CSI) was computed using the formula of Connor et al. (1986): CSI = (1.01 x g saturated fat) + (0.05 x mg)cholesterol). Atherogenicity (AI) and trombo-genicity (TI) indices, were calculated by the formula of Ulbricht and Southgate (1991) in which MUFA and PUFA designate mono and polyunsaturated fatty acids, AI = C12:0 + (4 x C14:0) + C16:0 / n-6 PUFA + n-3 PUFA + MUFA. TI = C14:0 + C16:0 + C18:0 / (0.5 MUFA) + (0.5 n-6 PUFA) + (3 n-3 PUFA) + (n-3 PUFA / n-6 PUFA). Health promoting index (HPI) was

calculated according to Chen *et al.* (2004): HPI= n-6 PUFA + n-3 PUFA + MUFA / C12:0 + (4x C14:0) + C16:0. Results of three measurements per sample were averaged before further statistical assessment. The results were analyzed with a completely random variance analysis. Purchasing days were treated as repeated measures. Comparison of means with a significant difference (P>0.05) was by Tukeys' test. All data were analyzed using the general linear model (GLM) SAS (2003).

Results

Qualitative radical scavenging activity (QLRSA) was higher for Sainte-Maure and Panela than Feta varieties (Table 1). However, the best quantitative radical scavenging activities (QRSA) were recorded for Bonn Rennes and Lanzarote, both Feta variety cheeses (42.2 and 37.8%, respectively). The highest mean concentration of total polyphenols was found in Queso Caprina (4.23 mg $100g^{-1}$ of gallic acid equivalents). No Pearson correlation was found between total polyphenols and QRSA (r = 0.026). Cholesterol/saturated fat index (CSI) tended to be higher in Feta (2.7) than Sainte-Maure (1.9) and Panela (1.7) varieties (Table 2). Mikonos light showed the

lowest CSI with 1.2. In contrast, Lanzarote [(Feta variety) 2.9], Castelvell (2.9) and Rancho Vistalegre (2.8) were the cheeses with the highest CSI. Atherogenicity index (AI) was highest for Cabrero (3.1), Laclette (3.0), and Notre Dame (2.9); the first being a Panela and the second and third, Sainte-Maure varieties (Table 2). CLA ranged from 0.3 to 0.9% with no clear tendency for variety or brand of cheese. The lowest thrombogenicity index (TI) was for Chateau Blanc (2.2) a Sainte-Maure cheese. The best health-promoting index (HPI) was that of Artesanal Gourmand (0.50) whereas the worst corresponded to Cabrero (0.32) and Laclette (0.33).

Kinds and brands of cheese	Total Polyphenols*	QLRSA by TLC and	QRSA by DPPH		
	51	DPPH radical ¹	radical ¹		
Artesanal Gourmand ^Â	3.10 ^{def}	++	24.2 ^{ef}		
Bon Rennés ^Â	2.72 ^{hi}	++	13.7 ^{ij}		
Castelvell ^Â	2.46 ^{ij}	++	28.6 ^d		
Chateau Blanc ^Â	3.34 ^{cd}	++	14.6 ^{hi}		
DianeÂ	2.78ghi	+	26.0 ^{de}		
El Queso de Cabra ^Â	2.71 ^{hi}	+	34.6°		
La Parroquía de Xico ^â	3.75 ^b	+++	4.61 ^k		
LacletteÂ	3.21 ^{cde}	+	11.0 ^j		
Lanzarote ^Â	3.36 ^{cd}	++	17.2 ^{hg}		
Le Blanc ^Â	3.14 ^{cdef}	+	24.1 ^{ef}		
Mikonos Light ^â	3.30cd	+	19.2 ^g		
MontchevreÂ	1.40 ^k	++	5.51 ^k		
Notre Dame ^Â	2.87 ^{hfg}	++	23.1 ^f		
Queso Caprina ^Â	4.23ª	+	33.8°		
Rancho Vistalegre ^Â	2.19j	++	32.1°		
Bon Rennés [‡]	3.10 ^{def}	+	42.2ª		
Castelvell [‡]	3.31cd	+	13.4 ^{ij}		
Laclette Gourmet [‡]	3.20 ^{cd}	+	2.91		
Lanzarote [‡]	3.44 ^{bc}	+	37.8 ^b		
Rancho Vistalegre‡	2.30 ^{efgh}	+	18.8g		
Cabrero†	2.22j	++	5.7 ^k		

 Table 1.
 Total polyphenols, qualitative (QLRSA) and quantitative (QRSA, %) antioxidant activity of commercial Mexican soft goats' milk cheeses

a,b,c,d,e,f,g,h,i,j,kMeans with different letters indicate differences among cheeses (P<0.05).

^ACheese variety Sainte-Maure. [‡]Cheese variety Feta. [†]Cheese variety Panela. ^{*}mg 100g⁻¹ of GAE.

+ = Weak intensity. ++ = Intermediate intensity. +++ = Strong intensity.

TLC = Thin layer chromatography. 1DPPH+ =1,1-diphenyl-2-picrylhydrazyl.

Table 2. Cholesterol (mg/100 g), fatty acids (%) and health and risk fatty acid indices of commercial Mexican soft goats' milk

cheeses. Kinds and brands of cheese	Chol	SFA	MUFA	PUFA	CLA	CSI	AI	TI	HPI
ArtesanalGourmand $^{\hat{A}}$	131.4 ^{abcd}	63.4 ⁱ	31.8ª	4.8ghi	0.7 ^{bcd}	2.0 ^{ef}	2.0 ^j	2.5 ^{fgh}	0.50ª
Bon Rennés ^Â	103.5 ^{cd}	67.8 ^{cdef}	28.4 ^{cde}	3.9j	0.5^{efg}	1.5h	2.5 ^{efg}	3.0 ^{bc}	0.39 ^{de}
CastelvellÂ	160.4^{abcd}	67.3efg	21.0 ^{defg}	5. 7 ^{bc}	0.6 ^{cdef}	1.9fg	2.5fgh	2.6fg	0.40de
Chateau Blanc ^Â	137.0 ^{abcd}	66.5 ^{fgh}	27.9cdef	5.5bcd	0. 3 g	2.0ef	2.3ghi	2.2 ⁱ	0.44 ^b
DianeÂ	151.2 ^{abcd}	64.9hi	28.8 ^{bcd}	6.3ª	0.7bcde	2.0ef	2.2 ^{ij}	2.4ghi	0.46 ^b
El Queso de Cabra ^Â	191.9abc	65.1 ^{hi}	30.2 ^{ab}	4.7 ^{hi}	0.8abc	2.6cd	2.3 ^{hi}	2.6 ^{def}	0.44 ^b
Parroquía de Xico ^Â	133.4 ^{abcd}	68.2 ^{cdef}	27.3 ^{cdefg}	4.5 ⁱ	0.6^{cdef}	1.9 ^{fg}	2.6 ^{def}	2.7 ^{def}	0.38 ^{de}
LacletteÂ	216.5ª	70.4 ^{ab}	24.3 ^{ij}	5.3bcdef	0.6 ^{cdef}	2.1 ^{def}	3.0 ^{ab}	3.1ªb	0.33g
LanzaroteÂ	111.9 ^{bcd}	68.1 ^{cdef}	26.7 ^{efgh}	5.3cdefg	0.6 ^{cdef}	1.6h	2.6 ^{deg}	2.7 ^{def}	0.39 ^{de}
Le Blanc ^Â	144.2 ^{abcd}	66.4^{fgh}	28.0cdef	5.7bc	0.9 ^{ab}	2.1 ^{def}	2.3ghi	2.3hi	0.44 ^b
MikonosLightÂ	85.2 ^d	67.5 ^{efg}	26.7efgh	5.7 ^b	0.6^{def}	1.2i	2.5 ^{fgh}	2.6 ^{ef}	0.40c
MontchevreÂ	107.7cd	67.3efg	26.2fgh	6.5ª	0.7bcde	1.5h	2.5 ^{fgh}	2.5 ^{fgh}	0.40de
Notre Dame ^Â	156.4^{abcd}	69.5 ^{bcd}	25.1 ^{hi}	5.4 ^{bcde}	0.5^{defg}	2.2 ^{de}	2.9 ^{abc}	2.7 ^{def}	0.34 ^{fg}
Queso Caprina ^Â	141.4^{abcd}	65.8 ^{gh}	28.9 ^{bc}	5.4 ^{bcde}	0.9ª	2.1^{def}	2.6 ^{def}	2.7 ^{def}	0.39 ^{de}
Rancho Vistalegre ^Â	160.2 ^{abc}	67.4efg	27.5 ^{cdefg}	5.1 ^{defgh}	0.5 ^{efg}	2.3cd	2.6 ^{def}	2.7 ^{def}	0.38de
Bon Rennés‡	211.9ª	68.7 ^{bcde}	26.5fgh	4.8ghi	0.7 ^{bcde}	2.5¢	2.8cde	3.3ª	0.36ef
Castelvell [‡]	208.6 ^{ab}	69.6 ^{bc}	25.1 ^{hi}	5.3 ^{bcdef}	0.5 ^{efg}	2.9ª	2.8 ^{bcd}	2.6 ^{def}	0.36 ^{ef}
LacletteGourmet [‡]	168.7 ^{abcd}	66.7fgh	27.7cdef	5.6 ^{bc}	0.6 ^{cdef}	2.5¢	2.6 ^{def}	2.8cde	0.38de
Lanzarote [‡]	200.4 ^{abc}	67.6 ^{def}	27.0 ^{defg}	5.3bcdef	0.6cdef	2.9ª	2.7 ^{def}	2.8cde	0.38 ^{de}
Rancho Vistalegre [‡]	194.8 ^{abc}	69.0 ^{bcde}	25.8 ^{ghi}	5.1 ^{efgh}	$0.4^{\rm fg}$	2.8 ^{ab}	2.7 ^{def}	2.8 ^{cd}	0.38 ^{de}
Cabrero†	128.5 ^{bcd}	72ª	23.2 j	4.9fghi	0.7bcde	1.7gh	3.1ª	3.0bc	0.32g

a,b,c,d,e,f,g,h,iMeans with different letters indicate differences among cheeses (P<0.05).

 $^{\hat{A}}$ Cheese variety Sainte-Maure.

*Cheese variety Feta.

[†]Cheese variety Panela.

Chol = cholesterol.

SFA = saturated fatty acids.

PUFA = polyunsaturated fatty acids.

MUFA= monounsatured fatty acids.

CLA = conjugated linoleic acid isomers (cis-9, trans-11; trans-9, cis-11; trans-10, cis-12; mg g⁻¹ of fat).

CSI = cholesterol-saturated fat index (1.01 x g saturated fat) + (0.05 x mg cholesterol).

AI= atherogenic index (C12:0+(4xC14:0)+C16:0)/(n-6PUFA+n-3PUFA+MUFA).

TI = trombogenic index (C14:0+C16:0+C18:0)/((0.5MUFA)+(0.5n-6PUFA)+(3n-3PUFA)+(n-3PUFA/n-6PUFA).

HPI=health promoting index (n-6PUFA+n-3PUFA+MUFA/C12:0+(4*C14:0)+C16:0).

Discussion

The evaluation of qualitative radical scavenging activity (QLRSA) did not correspond directly to the quantitative radical scavenging activity (QRSA). This discrepancy is possible because scavenging properties depend on the nature and amount of bioactive compounds, as well as their singular abilities to control free radicals; i.e., the interactions among functional ingredients and their synergic, additive or antagonist characteristics may affect scavenging rates (Sharma et al., 1998). There was not a significant correlation between total polyphenols and QLRSA (r = 0.026), probably because other milk antioxidants, such as glutathione peroxidase, glutathione transferase, etc. interfered in the assay masking the scavenger effect of polyphenols. The type of cheese did not influence the total polyphenol content. A possible explanation of the large variation in polyphenol concentration both among type and within type of cheese, could be due to different consumption rates of such compounds from feedstuffs (Cuchillo et al., 2013). However, with the variable data of the present study and no clear tendency for type or brand of cheese, it is not possible to speculate about the feeding system utilized. Further investigation should trace cheese products and take into account production systems.

In relation to cholesterol composition, previous studies have reported values from 155 to 198 mg/100 g (Galina et al., 2007) and from 172 to 195 mg/100 g (Cuchillo et al. (2010b) in goats' cheese resulting from similar feeding conditions. In both experiments one group of animals grazed on shrub land, while another group was barn fed. Free ranging resulted in lowered cholesterol levels in cheese in both studies; however, this effect was significant only in the first case (P<0.05). The intake of goat cheeses does not represent a health risk, if it does not exceed the WHO (2003) guidelines of daily cholesterol intake of 300 mg (about 150 g of goat cheese). The SFA results in the present study agree with those of Pajor et al. (2009), who reported values up to 73.4% of SFA. However, the present SFA values are higher than those previously reported for cheeses made from the milk of goats supplemented or not with vegetable oils and oilseeds rich in PUFA (66% vs. 60% of SFA, respectively), (Gassi et al., 2012). Increased concentrations of MUFA and PUFA

could explain the reduction of SFA, since as shown by Gassi et al. (2012), the inclusion of PUFA-rich feedstuffs in the diet, modifies rumen ecology and milk composition to incorporate more unsaturated fatty acids, which decrease the SFA value. Most of the cheeses here analyzed, had PUFA and MUFA concentrations similar to those previously reported by Pajor et al. (2009) and Gassi et al. (2012) with values ranging from 24 to 29.6%, and from 4.3 to 7.2% for MUFA and PUFA, respectively. Grazing systems can result in increased PUFA and CLA concentrations due to the contributions of grass forages. It seems likely that cheeses with high MUFA and PUFA come from either pastoral-based feeding or other systems that include PUFA-rich feedstuffs.

It is not surprising that the CSI was found higher in Feta cheeses (2.7) because of their high mean cholesterol content (169.5 mg/100 g). Connor et al. (1986) reported for a 2% fat cheese (low-fat cottage) a CSI of 1.0, which is close to the range of our values from 1.2 to 2.9, whereas for a 25 to 30% fat cheese a CSI of 6.0 was reported. The AI value found in most of the cheeses (up to 3.0) were higher than those reported by Ulbricht and Southgate (1991) for milk, butter and cheese (2.0). Only one Sainte-Maure variety cheese (Artesanal Gourmand) was found to have that same value (2.0) whereas the rest were higher. The use of PUFA supplementation has also reduced the AI of goats' milk with decreased C12:0, C14:0 and C16:0 fatty acids content (Gassi et al., 2012). Therefore, it would be desirable to increase the dietary intake of fatty acids content such as CLA (C18:2) isomers. Interestingly, Feta cheese did not record the highest AI even with their high CSI. The most likely explanation of the increased AI of Panela cheese is the lower PUFA content (4.9% of fat) compared to Feta cheeses (5.2% of fat) and high SFA percentage (72%). TI values (up to 3.3) were higher than those found by Ulbricht and Southgate (1991) in milk and cheese (2.0). The low proportions of PUFA and MUFA prevented a lower TI, thus increased proportions of those unsaturated fatty acids in cheese fat would be necessary to improve As for HPI, the best result this criterion. corresponded to a Sainte-Maure cheese [Artisanal Gourmand (0.50)].

Conclusion

The largest cholesterol and CSI were greatest in Feta cheeses. Differences in all parameters were

detected among varieties and within varieties among brands of cheese. Based on these results we recommend most highly the consumption of Mikonos Light due to its low cholesterol and CSI value. Chateau Blanc is also recommended for its low TI and high HPI. In contrast, Laclette is less recommended because of its high cholesterol, SFA, AI and TI values and its low HPI. Cabrero cheese is not recommended either due to its high SFA, AI and low HI, In spite of a low CSI.

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