

Fatty Acid Profile Differences Among the Muscle Tissue of Three Rodents (*Hydrochoerus hydrochaeris*, *Cuniculus paca* and *Cavia porcellus*) and one Lagomorph (*Oryctolagus cuniculus*)

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Abstract The consumer demand for functional food with nutraceutical properties has increased in recent years, and the food's fatty acid profile is one of the factors that define its potential value as a functional food. The aim of the present study was to compare fatty acids composition of semitendinosus muscle in two native rodents, the capybara (*Hydrochoerus hydrochaeris*) and lowland paca (*Cuniculus paca*) and two domesticated species, one rodent, the guinea pig (*Cavia porcellus*) and one lagomorph, the common rabbit (*Oryctolagus cuniculus*). The animals were fed with forage-based diet. The fatty acid composition in lyophilized tissue was determined by gas chromatography. The capybara, lowland paca and guinea pig had the highest levels of omega-3 (n-3) fatty acids, predominantly α -linolenic acid ($p < 0.05$). The guinea pig and common rabbit showed a higher content of linoleic acid (18:2 n-6) ($p < 0.05$). The n-6: n-3 fatty acids ratio was higher to the common rabbit ($p < 0.05$) with 8.9, in contrast, capybara and lowland paca had the lowest ratio, with lower than zero values indicating a predominance of n-3 fatty acids. These results were presented without any diet manipulation. The guinea pig meat contained an adequate balance of n-6 to n-3 fatty acids (< 4) whereas in rabbit meat there was an excess of n-6 fatty acids. The capybara and lowland paca can be considered as sources of n-3 fatty acids, represented by α -linolenic acid (18:3 n-3). These results confirm that native species not subjected to genetic selection and intensive feeding, showed higher content of n-3 fatty acids.

Keywords: Omega-3 fatty acids, small herbivores, fatty acid profile, α -linolenic acid

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1. Introduction

In recent years, numerous studies have been focused on increase the n-3 fatty acid content in meat, eggs or milk through modifications of animal diets [1,2]. This increased focus is a result of the increased demand for functional foods that can prevent or control diseases. The functional food market value can attain twice the commercial value of ordinary food [3], and foods enriched with n-3 fatty acids can be considered functional foods because n-3 fatty acids are essential for health and can prevent and control diseases such as arthritis, inflammation, autoimmune disorders, cardiovascular disease and cancer [4,5]. Currently, it is known that an interaction occurs between nutrition and genetic material, mainly between n-6: n-3 fatty acids ratio, specific genetic variants and disease phenotypes [6,7]. Many of the chronic conditions of cardiovascular disease, diabetes, cancer, obesity,

autoimmune diseases, rheumatoid arthritis, asthma and depression are associated with increased production of thromboxane A₂ (TXA₂), leukotriene B₄ (LTB₄), suppress interleukin 1 β (IL-1 β), tumor necrosis factor- α (TNF α) and interleukin-6 (IL-6), and C-reactive protein (CRP). All these factors are increased by increases in omega-6 fatty acid intake and decreased by increases in omega-3 fatty acid intake [8]. Furthermore, the balance of omega-6 and omega-3 fatty acids is very important for homeostasis and normal development.

However, few studies have addressed to native foods fatty acid composition, which were staple foods for our ancestors, compared to the food that are produced under commercial conditions and had been subject to selection and breeding. The muscular fibre type composition and fatty acid profile has been showed that changes for intensively produced species respect to natives species [9].

The capybara, lowland paca, guinea pig and rabbit are predominantly herbivorous species that are ceceal

fermenters and practice caecotrophy. The capybara is considered the largest rodent in the world (50 kg of body weight) and is the most abundant in the flooded savannas of South America. It is distributed from Panama to Uruguay and north of Argentina, and it mainly inhabits the edge of the Atlantic coast of South America [10]. However, the capybara as a species has not yet undergone programs of genetic improvement and intensive production. The lowland paca or spotted paca has a similar past to the capybara and guinea pig as a food source for the natives who inhabited the Andean region of South America. The lowland paca is one of the largest rodents in the world (12 kg of body weight). It lives in Andean forests, inhabits flooded savannas, and its diet is based on vegetables and aquatic animals. The lowland paca is a species that can adapt to living in intensively cultivated areas [11]. The guinea pig is a species native to the Andes, breeding commercially to human intake; there are approximately 35,000,000 of animals in countries such as Peru, Bolivia, Ecuador and Colombia and is appreciated for the high quality of its meat and linked to the local gastronomic traditions. The guinea pig reaches a body weight of 900 g in seven weeks of age, and it is fed a diet based on forage, with protein and energy supplementation [12]. The breeding of rabbit is globally standardized, and similar to the guinea pig, they have been subjected to genetic selection and breeding. Traditionally, the rabbit has been recognized as a suitable meat source for human feeding because of its high protein and low fat content compared to poultry, beef or pork [13].

Thus far, the studies performed on the capybara and lowland paca have focused on the biology, geographic distribution and breeding [14]. However, little is known of their meat composition, in particular the fatty acid profile. The knowledge of meat fatty acid profile and their comparing between wild and domesticated species could help to define changes in the profile and metabolism of fatty acids as a result of domestication. Also, it is important to characterize these meats from the human food quality perspective. This study characterized the fatty acid profile in four small herbivorous species, rabbit and guinea pig bred commercially, capybara and lowland paca were produced under not conventionally breeding.

2. Materials and Methods

Twelve samples of each species were collected from commercial farms (guinea pig and rabbits) or wild animals breeding (capybara and lowland paca). The animals were fed with forage-based diet. Three hundred grams of semitendinosus muscle to analysis were taken. The tissue was lyophilized and conserved at -10°C until analysis. Lipids were extracted by a chloroform/methanol 2 to 1 solution [15] using a mechanical homogenizer. The homogenized product was filtered, and 5 ml of distilled water was added to 20 ml of this filtrate. The mix was centrifuged at 3000 rpm for 20 minutes, the supernatant was discarded, 1 ml of the lower organic phase was placed in a previously weighed test tube, and the solvent was evaporated under a gentle stream of nitrogen. The dry lipids were dissolved in a chloroform/methanol 1 to 1 solution (1 ml for each 100 mg of lipids). A 50 µl aliquot of the lipid solution was esterified by the methyl

esterification Meth-Prep II reagent (Alltech Associates Inc., Deerfield, IL, USA) to produce fatty acid methyl esters.

The fatty acid methyl esters were analyzed by a Shimadzu GC-2014 (Kyoto, Japan) gas chromatograph equipped with a flame ionization detector (260°C). Separation was performed on a 30 m x 0.32 mm Supelco® Omegawax 320 column with 0.25 µm film thickness. Separation was achieved by a temperature ramp program (initial temperature of 80°C, then increased by 10°C/min until attaining 190°C, held for 20 min at 190°C, then increased by 2°C/min until attaining 220°C and held for 10 min at 220°C). Helium was the transporter gas, and the injection was performed in “split” mode (ratio 1 to 50). The fatty acid methyl esters were identified by comparing their retention time to a standard mix of fatty acids (Supelco 37 component FAME Mix, Supelco, Inc., Bellefonte, PA, USA). The fatty acid composition was expressed as a percentage of each fatty acid to the total of the identified fatty acids.

The statistical analysis was performed using an analysis of variance at one way to evaluate the results and compare the fatty acid content between the four species under study. The differences were tested Tukey’s test [16].

3. Results

The muscle fatty acid profile of the four species under study and lists the content of saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs), polyunsaturated fatty acids (PUFAs), n-3 and n-6 fatty acid concentrations and ratio of n-6 to n-3 fatty acids are presented in the Table 1.

The guinea pig presented the greatest content of PUFAs ($P<0.05$) at more than 40% compared to the paca, capybara and rabbit. The capybara and lowland paca showed higher levels of n-3 fatty acids ($P<0.05$) with 20.0 and 22.5%, respectively; whereas guinea pig and rabbit had 9.0 and 3.5%, respectively. The α -linolenic fatty acid (C18:3 n-3) was prevalent among the n-3 fatty acids. However, a large content of eicosatetraenoic acid (C20:3 n-3) was found in the lowland paca with a 9.0%, also docosapentaenoic acid (C20:5 n-3) at 1.0%. The guinea pig and rabbit showed the highest content of n-6 fatty acids ($P<0.05$), with more than 30% of linoleic acid (C18:2n-6), where astocapybara and lowland paca, the contents were 12 and 13%, respectively. The rabbit showed the highest n-6 to n-3 ratio ($P<0.05$) with a value of 8.9, when is compared with another studied species. Remarkably, the capybara and lowland paca showed n-6: n-3 ratio lower than 1, which implies that n-3 fatty acids predominated in these species.

The capybara and rabbit showed the highest levels of total MUFAs respect to lowland paca. The predominant fatty acid of this group in all species under study was the oleic acid (C18:1n-9), and there were no differences among the species. The second-most prevalent MUFA was palmitoleic acid, which was higher in the rabbit compared to the guinea pig ($P<0.05$). Heptadecenoic acid (C17:1) was only detected in the capybara and lowland paca, at levels $\leq 1\%$.

Capybara showed higher saturated fatty acids than guinea pig ($P<0.05$). Palmitic acid (C16:0) was the

predominant saturated fatty acid to all of the species. Palmitic acid levels ranged from 25.6 to 27.8% and there were no significant differences among species. Stearic acid (C18:0), another saturated fatty acid, was the second-

most prevalent fatty acid, and it was higher in the lowland paca meat ($P < 0.05$) compared to rabbit with 8.4 and 6.0%, respectively.

Table 1. Fatty acid composition in the capybara (*Hydrochoerus hydrochaeris*), lowland paca (*Cuniculus paca*), guinea pig (*Cavia porcellus*) and common rabbit (*Oryctolagus cuniculus*) muscle¹

Fatty acid	Percent of fatty acid in relation to total fatty acids			
	Capybara	Lowland paca	Guinea pig	Common rabbit
C14:0 (myristic)	3.0 ± 0.1	2.8 ± 0.2	--	4.0 ± 0.1
C16:0 (palmitic)	27.8 ± 0.2	26.6 ± 1.1	25.3 ± 0.5	25.5 ± 0.1
C16:1 (palmitoleic)	2.0 ± 0.1 ^{ab}	2.2 ± 0.2 ^{ab}	1.3 ± 0.1 ^b	4.4 ± 0.1 ^a
C17:0 (heptadecanoic)	1.2 ± 0.1	1.4 ± 0.1	--	0.52 ± 0.1
C17:1 (heptadecanoic)	1.0 ± 0.1	0.9 ± 0.1	--	--
C18:0 (stearic)	7.1 ± 0.1 ^{ab}	8.4 ± 1.0 ^a	7.4 ± 0.2 ^{ab}	6.0 ± 0.1 ^b
C18:1 n-9 (oleic)	24.5 ± 0.6	21.6 ± 0.9	24.7 ± 0.5	25.0 ± 0.4
C18:2 n-6 (linoleic)	10.7 ± 0.3 ^b	13.3 ± 1.3 ^b	31.3 ± 0.1 ^a	31.1 ± 0.3 ^a
C18:3 n-6 (gamma-linolenic)	1.3 ± 0.1	--	--	--
C18:3 n-3 (alpha-linolenic)	20.0 ± 0.3 ^a	12.5 ± 0.9 ^b	7.7 ± 0.2 ^c	1.5 ± 0.1 ^d
C20:3 n-3 (eicosatetraenoic)	--	9.0 ± 0.1 ^a	1.3 ± 0.2 ^b	2.0 ± 0.2 ^b
C22:5 n-3 (docosapentaenoic - DPA)	--	1.0 ± 0.1	--	--
Total saturated	39.4 ± 0.7 ^a	36.3 ± 2.1 ^{ab}	32.6 ± 0.4 ^b	36.0 ± 0.2 ^{ab}
Total monounsaturated	27.9 ± 0.4 ^a	24.6 ± 0.9 ^b	26.1 ± 0.6 ^{ab}	29.3 ± 1.2 ^a
Total polyunsaturated	32.7 ± 0.3 ^b	36.1 ± 0.2 ^{ab}	40.3 ± 0.1 ^a	34.5 ± 1.1 ^b
Total n-3	20.0 ± 0.3 ^a	22.5 ± 0.3 ^a	9.0 ± 0.2 ^b	3.5 ± 0.1 ^c
Total n-6	12.0 ± 0.3 ^b	13.3 ± 0.9 ^b	31.3 ± 0.1 ^a	31.1 ± 1.5 ^a
Ratio n-6:n-3	0.6 ± 0.1 ^c	0.5 ± 0.1 ^c	3.5 ± 0.2 ^b	8.9 ± 0.2 ^a

¹Means ± standard error, n=12

^{abc}horizontal superscripts show statistically significant means ($P < 0.05$)

4. Discussion

The present study is the first to compare the fatty acid profiles of three rodent and one lagomorph species, with two domesticated herbivores species (guinea pig and common rabbit) and two wild species (capybara and lowland paca) included to assess their animal breeding potential.

Although dietary fatty acid composition is a determinant factor of the animal tissue fatty acid content [17], such profiles in herbivorous species also depend on factors other than diet. These factors include fatty acid microbial metabolism and hydrogenation in the digestive tract [18], enzymatic activity in the digestive tract, oxidation status [19] and genetic aspects in the specific case of rabbits [20], novo synthesis and beta-oxidation in the mitochondria lead to accumulation of fatty acids in tissues and depend of genes expression such as acetyl-CoA carboxylase (ACC) 1, ACC2, and fatty acid synthase (FAS). A decrease in the n-3 fatty acid content may occur in animal tissue because of animal domestication and intensive breeding. In general, n-3 fatty acid levels found in the capybara, lowland paca and guinea pig were equal to or greater than the levels found in studies where the animal diet was manipulated with n-3 fatty acids sources. This enrichment was performed to increase the n-3 fatty acid levels in the consequent animal products, such as the meat or eggs, in different species. For example, Crespo and Esteve-García [21] obtained contents of 24.4% to α -linolenic acid in broiler chickens fed with a diet supplemented with 10% of linseed oil, whereas Galobart et al. [22] obtained contents of 10.3% to α -linolenic acid in the egg yolk of hens fed a 5% of linseed oil in diet. The herbivorous rodent species studied were under natural feeding conditions based on forage, the levels of n-3 fatty acids were higher in these species than other herbivorous species, including lamb, beef and buffalo meat [23].

The percentage of n-3 fatty acids in the meat of capybara was less than reported by Delfino and Sarno [24]. They found levels of 25% to α -linolenic acid, 0.3% eicosapentaenoic acid (C20:5n-3, EPA) and 2.5% to docosahexaenoic acid (C22:6n-3, DHA). In the present study, a 20% of α -linolenic acid level was found and neither EPA nor DHA was detected. These differences might be attributed to different environments and diets since the data from Delfino and Sarno [24] were from Argentina's capybaras, whereas the present study was conducted with Colombia's capybaras.

The main dietary fatty acid for monogastric animals, particularly for rabbit, is linoleic acid (C18:2 n-6) [25]. The levels of linoleic acid were high in the guinea pig (31.3%) and rabbit (22.1%), with similar value to the reported in previous studies [13,26]. In contrast, the lowest levels of linoleic acid were found in the wild species of lowland paca and capybara. These results confirm that the foods commonly consumed from commercial production system are usually deficient in n-3 and rich in n-6 fatty acids, caused by intensification of agriculture [19,27] which promotes the disequilibrium of the n-6 to n-3 ratio in the modern human diet.

The meat of capybara, lowland paca and guinea pig showed a suitable n-6 to n-3 fatty acids ratio (less than 4) for human nutrition [5]. However, the values for rabbit meat did not fall within these levels. In previous studies with rabbit meat, a decreased n-6 to n-3 fatty acids ratio to less than 4 values of has been reached by diet modifications and the inclusion of linseed oil [24,28] or Chia seed (*Salvia hispanica* L.) [2]. However, the same authors confirmed a reduced oxidative stability of the fat samples through a thiobarbituric acid reactive substance (TBARS) assay when the diet was enriched with n-3 fatty acids. Other studies with different domesticated species have also reported higher levels to n-6 to n-3 fatty acids ratio, with 11.3, 10.7 34.9 ratio in rabbit meat, chicken breast and beef longissimus, respectively [26,29]. Maintain the equilibrium between n-6 and n-3 fatty acids

is essential. Eicosanoids of great physiological significance are generated from these two fatty acid groups and show antagonistic functions. Therefore, the n-6 and n-3 fatty acids must be maintained within a balance [19]. Thus, the ratio of n-6 to n-3 requires values within the physiological range of 4:1 to 2:1 [7]. An excess of omega-6 fatty acids and a deficiency of omega-3 fatty acids are frequently found to the most of foods. This tendency has been attributed to the intensification of agriculture [5,7] and has been confirmed by the present study. Native species such as capybara and lowland paca have not been subjected to genetic selection pressure and maintain suitable levels of n-3 and n-6 fatty acids.

Another important factor related to fatty acid profiles is the PUFAs and SFAs ratio because this determines the fluidity and physical properties of the cell membranes. Low values have been associated with diabetes, obesity, hypertension, cancer and cardiovascular disease [30]. The meat of the guinea pig and lowland paca showed a PUFAs and SFA ratio of 1.2, whereas the capybara and rabbit meat had a ratio of 0.8. All species of the present study showed a better (higher) ratio between polyunsaturated and saturated fatty acids compared to other domesticated species. Chicken meat had a value of 0.3 [1], beef had a value of 0.23 [23] and pork had a value of 0.54 [31].

The SFA content was relatively low to the species studied when they were compared to other herbivorous animals, such as bovine or moose [23]. DalleZote [26] reported a 38.3% of SFA content in rabbit leg and 38.9% to beef. Enser et al. [32] reported a 36.9% of SFA content in pork meat. Low levels of stearic acid with high levels of oleic acid (18:1n-9) and the presence of monounsaturated fatty acids C16:1 and C17:1 indicate a proper enzymatic activity of stearyl-CoA desaturases [31], mainly of delta-9-desaturase enzyme in the liver or the possible bacterial isomerization of 18:1n-9 [33] in the species under this study. The muscular fiber type composition and fatty acid profile has been showed that changes for intensively produced species respect to natives species [9].

5. Conclusions

The results of the present study suggest that the meat of the capybara, and lowland paca could be considered as differentiated food because their high content of omega-3 fatty acids and low (<1) n-6: n-3 fatty acids ratio. These meats also have a higher PUFA to SFA ratio when they are compared with chicken, beef and pork. There are contrasting differences in the fatty acid composition in the species under study compared to another herbivorous species, such as bovine or lamb. It is possible that different fatty acid metabolism exists among these species that is associated with the ruminant digestive system. Thus, would be interesting to investigate the differences in fatty acid metabolism and deposition between herbivorous monogastric and polygastric species, as well as the genes that regulate the metabolism of fatty acids through different animal species.

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