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COMPOSITIONAL ANALYSIS AND PHYSICOCHEMICAL AND MECHANICAL TESTING OF TANNED RABBIT SKINS

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Abstract: Chemical composition and physicochemical and mechanical parameters of New Zealand White rabbit tanned skin were evaluated. Skin samples from 70-d-old males, *in natura* and semi-finished, were collected for evaluation. The *in natura* treatment comprise skins without any processing, while semi-finished treatment comprise skins after soaking, fleshing, liming, de-liming, purging, degreasing, pickling, tanning, neutralising, re-tanning and dyeing, followed by oiling, drying, stretching and softening. After tanning, samples from the dorsal and flank regions were removed for tensile and physicochemical testing in the longitudinal and transverse directions. A split plot design was used with plot treatments (leather regions: R1=dorsal and R2=flank) and subplots directions (S1=longitudinal and S2=transversal), using 10 examples per treatment. At the end of processing, the leather analysis revealed low moisture (31.76%), protein (46.48%) and fat content (24.95%), and a high ash content (8.58%). Leather presented a pH of 4.9 and contained 2.0% chromium oxide, 25.5% extractable substances in dichloromethane, and these characteristics were coupled with a higher tensile strength (10.84 N/mm^2) in the dorsal region. However, samples in the same region proved to have higher elasticity (64.57%) in the longitudinal direction, although there was no difference in the progressive tearing analysis (21.07-23.50 N/mm). Overall, our analyses suggest that, in this case, the tanned leather product does not have sufficient resistance for application in clothing production.

Key Words: leather resistance, skin resistance, rabbits.

INTRODUCTION

Cuniculture provides producers with several interesting products, including meat as the main product and by-products such as skin and animal hair, as well as tails and front paws for the manufacture of key chains. Rabbit skin is considered an excellent by-product and may be used in the manufacture of garments, bedspreads, arts and crafts and other items. However, for production purposes, specimens must have good carcass characteristics and a marketable skin quality. The skin of the New Zealand White rabbit strain is highly desirable as it provides resistance, good elasticity, and other advantageous attributes that differentiate it from the skins of other species (Franco *et al.,* 2012).

The skin has a great capacity to absorb and retain water, a feat which is associated with its thermoregulatory function in the animal body. The skin is essentially composed of water, minerals, natural fat, protein material, pigments and carbohydrates (Gutterres, 2003). Although tanning maintains the fibrous quality of the skin, fibres are separated to remove interfibrillar materials and chemical residues. Following this stage, the skins are treated with tanning reagents that transform them into leather, or rather, a raw product characterised by softness, elasticity, flexibility, tensile strength and physical/mechanical properties that permit its application in various production sectors (Souza, 2008).

The use of different tanning reagents and the retrieval of skin from different parts of the animal with regard to its cephalic-caudal axis and in different directions (longitudinal and transversal) may cause modifications in the degree of leather resistance. This hypothesis requires quantitative testing to determine the degree of resistance in such skin samples. To this end, the current study evaluates the chemical composition and the physicochemical and mechanical characteristics of the skin of New Zealand White rabbits submitted to tanning.

MATERIALS AND METHODS

Rabbits were housed in the cuniculture area of the State University of Maringá (FEI – Iguatemi PR Brazil) and after slaughter, 40 skins of 70-d-old New Zealand White (NZB) male rabbits were collected. Skins were removed at slaughter, packed and stored at –18°C until processing.

Skin Tanning Process

Skin tanning was carried out in the laboratory of the State University of Maringá (FEI-Iguatemi PR Brazil) which has previously been used for fish and small/medium-sized species skin processing. Skins were thawed at room temperature and tanned with chrome salts and re-tanned with vegetable (Weibull) and synthetic (F Syntac) tannin, according to the processing stages described by Hoinacki (1989) and Souza (2004). Steps employed in the process comprised soaking, fleshing, liming (3% sodium sulphide, 3% lime, 100% water, 2 times), de-liming, purging, degreasing, pickling, tanning (6% chromium salts), neutralising, re-tanning (2% and 2% Weibull and Syntac F) and dyeing, followed by oiling (8% oils), drying, stretching and softening. The percentages used were based on the weight of the fleshed hides.

Leather Resistance Rests

After tanning, skin samples from the dorsal and flank regions were removed for tensile strength and elasticity tests (ABNT NBR ISO 3376, 2014) and for progressive tearing analyses (ABNT-NBR 3377-2, 2014). Samples were taken from leather (ABNT-NBR 11032, 2013) using a rocker and afterwards kept in an air-conditioned environment of around 23°C and 50% air humidity for 24 hours (ABNT-10455, 2014). The thickness of each sample (ABNT-NBR 2589, 2014) was assessed for tensile strength, elasticity, and gradual tearing. An EMIC dynamometer with a clearance speed of 100±20 mm/min was employed for endurance tests. The load cell in the dynamometer was set at 200 kgf.

Physicochemical Testing of Leather

Leather samples were collected and ground to form "leather powder", which was then used to measure pH (ABNT-NBR 11057, 2006), levels of extractable substances in dichloromethane (ABNT-NBR 11030, 2013), and levels of chromium oxide (Cr_2O_3) (ABNT-NBR ISO 5398-1, 2014).

Chemical Composition Tests for Hides and Skins

The chemical composition of skin and semi-finished leathers was determined. To determine the composition of skins, four samples were removed and packed in polyethylene bags, labelled and frozen at –18°C after slaughter. After thawing, these samples were ground in a cutting mill. To test the chemical composition of semi-finished leathers, four tissue samples collected after fixation steps and oiling, drying and softening were used.

Analyses for moisture, ether extract (fat), and ash contents (minerals) were performed as described by AOAC (2012) while crude protein contents were analysed using the Kjeldahl method (Silva and Queiroz, 2002). The above analyses were performed at the Animal Nutrition Laboratory (LANA) of the State University of Maringá, Maringa PR Brazil.

Statistical Analysis

A split plot design was employed, with plots as treatments (leather regions: R1=Dorsal and R2=flank) and subplots as the leather direction (S1=longitudinal and S2=transversal) using ten examples per group: R1S1; R1S2; R2S1 and R2S2. The results of the physical and mechanical tests were subjected to analysis of variance and averages using Tukey's test at 5% probability (SAEG, 2000).

RESULTS AND DISCUSSION

Figure 1 shows the results of the chemical composition analysis. There were statistical differences between *in natura* skins and semi-finished leathers after the greasing, drying and softening stages.

When compared to *in natura* rabbit skins, the semi-finished leather exhibited several different characteristics. Moisture content in the *in natura* rabbit skins was 64.36% while this was reduced to 31.76% in the semi-finished leather.

The protein content of *in natura* skin was 29.62%, in contrast to protein content in semi-finished leather of 38.48%. Skin processing involves removal of the

Figure 1: Chemical composition of *in natura* skin and semi-finished leather after processing. ■ *In natura*; □ Semi-finished.

hypodermis and, together with the adipose tissue, small amounts of collagen fibres. During processing, globular proteins are also removed through the actions of proteolytic enzymes added at the purge stage. Figure 1 shows that, despite the loss of soluble proteins during tanning, crude protein levels were concentrated due to the reduction in moisture content.

In natura skin presented 5.96% fat (ether extract) content. After chemical treatment and the addition of degreasers and emulsifiers of natural greases, collagen fibres were washed and excess fat removed to facilitate the reaction of tanning agents with the collagen fibres. According to Ben (2005), lipids must be removed early in the process as they hamper the penetration of chemicals and react with collagen fibres. This can interfere with the tanning process and hence the strength and quality of the final product. The same author reported that levels of over 4% lipid in the skin are harmful to tanning operations. If the degreasing stage is not effective, fat spots appear on the leather surface after dyeing and drying (Franco, 2007). However, when processing ends, after the fibres have reacted with tanning agents, an oil emulsion must be added for the lubrication of collagen fibres.

In the current study, skin fat levels initially observed at 5.96% rose to 24.95% after drying due to the addition of oils in the greasing stage. Drying of the material caused a reduction in moisture levels and consequently the concentration of oils, providing a high ether extract content in semi-finished leather. The amount of grease present in leather was due to the action of oils on fibrillar structures during the greasing process. This stage is directly related to the intrinsic qualities of leather, such as touch and softness, generating the desired leather compatibility.

When compared to *in natura* skin (2.08%), the mineral (ash) contents of semi-finished rabbit leather (8.58%) increased due to the incorporation of certain products (e.g. sodium chloride, chromium salts, sodium bicarbonate and tannins) at various stages of processing. The concentration of minerals in semi-finished leather also occurred during the drying of leather due to moisture reduction.

Table 1 shows the average thickness, strength, stretching and interaction data of carcass region and direction.

Table 1: Average rates of tensile and elongation of physical and mechanical tests of rabbit leather.

Factors	Thickness (mm) (mean±SD)	Tensile (N/mm²) (mean±SD)	Elongation (%)
R1=Dorsal	0.87±0.08[a]	10.84±1.07[a]	56.71
R2=Flank	0.69±0.03[b]	7.98±0.92[b]	48.86
S1=longitudinal	0.82±0.14[a]	9.11±0.88[a]	56.29
S2=transversal	0.75±0.19[a]	9.71±1.21[a]	49.29
P-value			
Region (R)	<0.05	<0.01	<0.05
Direction (S)	NS	NS	<0.05
R×S	NS	NS	<0.05
Coefficient of variation (%)	22.51	24.52	17.06

SD: standard deviation. Means not sharing superscript in the same column were significantly different at *P*<0.05; NS: non-significant.
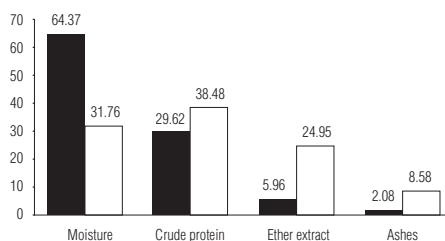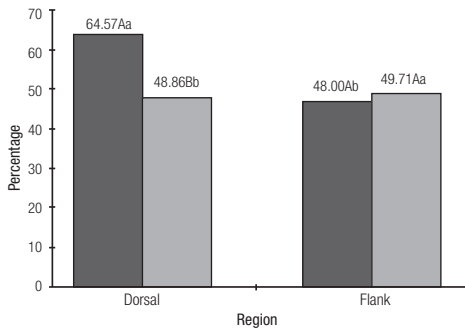
Figure 2: Deployment of Region (dorsal or flank) *vs.* Direction (longitudinal: ■ or transversal: ▨) of rabbit leather in elongation tests. Region stretching test within the direction of leather (direction within leather regions). The small letters indicate comparison between directions by Tukey test at 5% probability. The capital letters indicate comparison between regions by Tukey test at 5% probability.

We observed a correlation between region and leather direction (Figure 2); when chromium salts were added to the leather we observed higher elasticity in the dorsal region in a longitudinal direction (64.57%). The flank region in a longitudinal direction showed lower elasticity rates (48.00%).

The average thickness of dorsal region leather (0.89 mm) was significantly higher than that of the flank region leather (0.66 mm) although we observed no significant average difference for thickness with regard to the direction of leather. Furthermore, there were no significant differences in progressive tearing characteristics for the 2 regions of leather (dorsal=23.50 N/mm and flanks=21.07 N/mm) and the 2 directions (longitudinal=22.80 N/mm and transversal=21.77 N/mm). The maximum force applied in tensile and elongation tests and gradual tearing tests in leather was significant for each region but did not differ with leather direction (Tables 1 and 2).

The maximum forces applied in tests for the dorsal regions (20.36 N [Table 1] and 96.21 N [Table 2]) were significantly higher than on the flank regions (14.57 N [Table 1], 50.00 N [Table 2]). The intrinsic quality values of rabbit leather provided by physical and mechanical tests were then analysed after removal of samples. The dorsal region had higher tensile rates than the flank region but did not differ significantly when directions were compared. We expected that the flank region would present lower tensile strength and tearing and less elasticity due to the low concentration of collagen fibres with weaker interlacing when compared to other regions. The dorsal region of leather is richer in collagen fibre, creating texture and leather resistance (Carl and Freire 2000).

Oliveira *et al.* (2007) analysed the resistance of NZB rabbit leather according to sex at the age of 70 d. The leather of 70-d-old males had a mean thickness of 1.15 mm. When the leather resistance test was performed, Oliveira *et al.* (2007) reported an average tensile rate of 6.54 and 7.88 N/mm^2 respectively for longitudinal and transversal direction with no statistically significant difference between directions. These rates were lower than those obtained in the current study of 9.11 N/mm^2 for longitudinal and 9.71 N/mm^2 for transversal direction.

Progressive tearing rates by Oliveira *et al.* (2007) were 16.66 and 17.80 N/mm for the longitudinal and transversal directions, respectively. These rates were lower than those obtained in the current study. The observed differences for

Table 2: Mean rates of physical-mechanical progressive tearing tests of rabbit skin.

Factors	Thickness (mm)	Progressive Tearing (N/mm)	Maximum Force (N)
Region (R)			
R1=Dorsal	0.89±0.15[a]	23.50±3.22[a]	96.21±4.71[a]
R2=Flank	0.66±0.18[b]	21.07±2.94[a]	50.00±5.19[b]
Sample Direction (S)			
S1=longitudinal	0.77±0.03[a]	22.80±1.33[a]	71.86±4.04[a]
S2=transversal	0.76±0.04[a]	21.77±2.42[a]	74.36 ± 6.63[a]
P-value			
Region (R)	<0.01	NS	<0.01
Direction (S)	NS	NS	NS
R×S	NS	NS	NS
Coefficient of variation (%)	19.20	15.49	32.36

Means not sharing superscript in the same column were significantly different at *P*<0.05; NS: not significant.

progressive tearing, tensile strength, and elongation may be partly due to the tanning technique applied (4% sodium sulphide and 4% lime in the liming process) as sodium sulphide in high concentrations can damage collagen fibres and affect leather resistance. In the current study, the percentage was lower, with only 3% of sodium sulphide.

According to BASF (2005), the reference rates for chromium tanned leather for clothes, regardless of the type of re-tanning, should be a maximum of 60% for elongation, a minimum of 25 N/mm^2 for tensile strength or tension, and a minimum of 35 N/mm for progressive tearing resistance. The provisional requirements for garment leather quality for suede, nappa leather, nubuck and finished nappa leather established by the Commission for Leather Specification are 15 and 20 N/mm for resistance to progressive tearing. The tensile strength rate should be 12 N/mm^2 for the above-mentioned products. However, Hoinacki (1989) reported lower rates for chromium tanned cowhide with a tensile strength of at least 9.80 N/mm^2 and progressive tearing of 14.70 N/mm for clothing manufacture. According to Gerhard (1998), the requirements for quality finished leather clothing must be at least 15 N/mm for progressive tearing. The rates in the current study were higher than those by Gerhard (1998).

Studies which differentiate leather resistance characteristics from different animal body regions target leather for specific products depending on the region from where it was removed. When leather is analysed according to the body regions, the application for clothing or for other products depends on elevated resistance (Prado *et al.,* 2013). However, analysis of leather direction reveals directions with high elasticity or strength. For viability analysis, the mean rates for hides are estimated between longitudinal and transverse directions, with the average rate for traction being 9.41 N/mm^2 and elongation at 51.29% and 22.28% N/mm^2.

When the results in our current study were compared with those recommended in the literature, tensile characteristics were lower than reference standards set by BASF (2005) but very close to those found by Hoinacki (1989). The characteristic elongation rate found in the current study complies with that referenced by BASF (2005). The progressive tearing average rates obtained were above the reference rates for suede, nubuck and nappa leather recommended by the same author. Thus, the results of the leather resistance tests obtained in the current study were not sufficient to recommend these types of leather for use in clothing.

Leather chromium oxide content was 2.0%, below the minimum 2.5% recommended by BASF (2005). This percentage is due to the proportion of tanning agents attached to collagen fibres. Ether extracted substances (fat) reached 25.5% and indicated that the oil content in leather was above the recommended levels of 16% to 18% (BASF (2005)). This result may be due to an excessive amount of oils added in the greasing stage (Figure 3) or poor oil fixation at the end of the leather processing.

Comparing these physicochemical results with resistance tests showed that chromium content was less than that required while pH (4.9) was above the desired level (pH 3.5) for fixation of oils, dyes and tannin, and consequently this will influence resistance of the leather. Thus, the analysis suggests that leathers tanned by the present technique are not suitable for garment manufacturing, although they may be used to manufacture items such as wallets, purses, belts and other products, using a cloth backing or padding to reinforce the prepared product.

CONCLUSIONS

At the end of processing (semi-finished leather), leather has low moisture content due to drying, with a consequent increase in protein, fat and ash, influenced by the different stages in the tanning process and the addition of chemicals. The technique used for tanning led to high contents of fat/oil in the semi-finished leather, low chromium content, and a final pH above that recommended for the fixation of oils and dyes, which combine to influence the leather resistance.

Rabbit leather from the dorsal region showed greater tensile strength and leather longitudinal direction in the dorsal region revealed greater elasticity. There was no difference in leather resistance due to progressive tearing in region and direction. However, the tanning technique did not provide enough leather strength for clothing manufacture, although it may be used for the manufacture of other associated articles.

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