Thermal degradation study of vegetable tannins and vegetable tanned leathers

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Keywords: vegetable tannins, vegetable tanned leather, thermogravimetry, pyrolysis, mass spectrometry

Abstract

In this study, hydrolyzable tannins (commercial chestnut, valonea and tara extracts), condensed tannins (commercial quebracho and mimosa extracts) as well as calf leathers produced using these vegetable tanning agents were characterized by thermal decomposition methods using slow and high heating rates. Calf gelatin obtained by heating calf pelt in water at 70 ˚C was chosen as a reference material. Thermogravimetry/mass spectrometry (TG/MS) and pyrolysis-gas chromatography/mass spectrometry (Py-GC/MS) experiments were performed on the tannin and leather samples. The evolution profiles of the decomposition products as well as the thermal stability of tannins and leathers were studied by TG/MS. A net difference was observed in the thermal behavior of hydrolyzable and condensed tannins. The condensed tannins produced the highest char yield, while the hydrolyzable tara extract released the most volatile products. The tannins of higher reactivity produced more stable leathers as it results from their higher decomposition temperature. The composition of both vegetable tanned leathers and vegetable tanning agents was characterized by the pyrolysis product distribution measured by Py-GC/MS method. Resorcinol and its methylated derivative (orcinol) were found to be characteristic decomposition products for both condensed tannins, i.e. mimosa and quebracho. It was identified among the pyrolysis products of the mimosa and quebracho tanned leathers, as well. Characteristic decomposition product, a bisphenol derivative was identified among the pyrolyzates of hydrolyzable tannins and the leathers tanned with the hydrolyzable tannin agents.

Keywords: Vegetable tannins, Vegetable tanned leather, Thermogravimetry, Pyrolysis, Mass spectrometry

Highlights

Condensed tannins yielded higher amount of char than hydrolyzable tannins.
Tannins of higher reactivity produced thermally more stable leather.
Resorcinol was identified as a marker of the condensed tannins in leather pyrolyzate.
Pyrolytic profiles can be used to differentiate between hydrolysable and condensed tannins.

1. Introduction

The transformation of the animal skin into what is termed by leather, a stable, non-putrescible material, resistant to bio-deterioration, heat and humidity, smooth and flexible, is a complex technological process concerned with the chemical modification of collagen, prior to tanning and the tanning reactions in particular. The skin of any animal is largely composed of collagen, so it is the properties of this fibrous protein and the chemistry of tanning, namely the chemical interaction between collagen and tannin, that impact on the properties of the leather. Current tanning technology is dominated by chromium (III), which was introduced about 150 years ago and gradually replaced the traditional tannages based on plant polyphenols – so called vegetable tannins.

Therefore, vegetable tanned leather is the most common type of heritage leather found in museums and collections. Although today ca. 90% of the world's leather production is chrome tanned, the remainder is tanned with vegetable tannins and synthetic organic aldehydes.

Annually, leather industry produces remarkable amounts of chrome-containing waste, which requires a special disposal due to the carcinogetic potential of Cr(VI), being amongst world's top 10 toxic polluters. In EU, landfill and incineration of chrome leather wastes are still the most used options, and only in small part they are used in the production of biodiesel [1]. This practice has caused increased industrial Cr(VI) emissions that contaminate drinking water sources and widespread human exposure. Although Cr(VI) has been considered the species of main concern for human health effects, both forms must be considered because they can be interconverted by a variety of oxidants and reductants. Lately, it was shown that exposure to trivalent or hexavalent chromium can lead to measurable DNA–protein crosslinks formation, indicating exposure of the cells to reactive forms of chromium [2]. In drinking water collection, treatment, and distribution systems, virtually all transformations from Cr(III) to Cr(VI) and vice versa are mediated by constituents that are either naturally present in or purposefully added to water. Rapid changes are possible in the presence of dissolved oxygen, chlorine, chloramine, H_2O_2, MnO_2 solids, ferrous iron, stannous chloride, sulfites, etc. [3]. Moreover, high pH value, temperature, UV light, unsuitable storage conditions and the effect of using lubricants with double bonds in the molecule during production, possibly turn up the oxidation of the trivalent chromium into the hexavalent form in tanned leather [4]. Therefore there is a big push towards environmental sustainability and occupational safety and health in the EU leather industry. In this context, the traditional vegetable tannages has resuscitated in the last decade.

Tannins are complex and heterogeneous polyphenolic secondary metabolites, biosynthesized by higher plants, with molecular weights ranging from 500 to over 20000 Da [5]. Vegetable tannins can be divided into three main groups: hydrolyzable, condensed and complex tannins. The structure of the hydrolyzable tannins is built from the basic molecule, 1,2,3-trihydroxybenzene (pyrogallol) [6]. In the molecular structure of the hydrolyzable

tanning agents, sugar molecules are bonded to pyrogallols. The hydrolyzable tannins are hydrolyzed by acids (or enzymes) into a sugar molecule or a related polyhydric alcohol and a phenolic carboxylic acid. Depending on the nature of this phenolic carboxylic acid, the hydrolyzable tannins are usually subdivided into gallotannins and ellagitannins [5-7]. Tannins from *Castanea sativa* bark (chestnut extract) [8,9] and those from *Quercus valonea* acorn cups (valonea extract) belong to ellagitannins [10], while tannins from *Caesalpinia spinosa* fruits pods (tara extract) are gallolylated quinic acids and belong to the gallotannins [11].

Condensed tannins contain a group of polyhydroxyflavan-3-ol oligomers and polymers linked by C–C bonds between flavanol subunits [12,13]. Mimosa extract, derived from the bark of the black wattle tree (*Acacia mearnsii*) [14], and quebracho extract, originated from quebracho trees (*Schinopsis balansae* and *Schinopsis lorentzii*) [15], are prorobinetinidin and profisetinidin polymers, respectively.

In addition to conferring biodeterioration resistance and providing specific physical and mechanical properties necessary for the desired applications (i.e. flexibility, dimensional stability, water resistance, color fastness, etc.), tanning results in an increased hydrothermal stability of leather compared to skin. Although it was obvious that the tannin type influences the leather hydrothermal stability, a full understanding of the tanning mechanism has not been achieved so far. The most recent and comprehensive tanning theory is Covington's theory based on the link-lock concept that requires an initial reaction to link the collagen into the surrounding matrix of water and a second reaction component to lock the linked structure together, creating a macromolecular structure around the triple helices [16]. For vegetable tannins, the primary (link) reaction is mainly through multiple hydrogen bonding, while the subsequent reaction is to crosslink the tannin molecules together (lock), thus creating matrices of crosslinked polyphenolic species bound to collagen effectively working as a single moiety.

Recently, it was reported that the resistance of tanned leather to accelerated ageing by dehydrothermal treatment and visible light irradiation is higher than that of untanned collagen (parchment) and depends on both the tannin type, i.e. hydrolysable or condensed, and the animal (collagen) species, i.e. calf or sheep [7,17].

It has been shown that the tannin type has an influence on the balance between the two main deterioration processes of collagen, i.e., the thermal destabilization through damaged intermediate states formation and the stabilization through cross-link formation [7,17]. Moreover, thermal destabilization occurred more rapidly for sheep leathers exposed to aging (namely, shrinkage temperatures decreased with a steeper slope) by compared to calf leathers regardless of the type of tannin [17]. On the other hand, it was established in our previous study by TG/MS and Py-GC/MS that the natural aging process of the tanned leathers can be explained by the degradation of the tanning agents beside the denaturation of collagen [18]. All these findings are consistent to the link-lock theory, according to which the shrinkage of collagen in leather depends on the ease with which the species, i.e. tannins, bound to collagen can be displaced as the collagen shrinks.

The complex structure of some of the modern tanning agents [19,20] and commercially available leathers [21] has rarely been characterized by thermoanalytical and pyrolytic techniques. Therefore, the goal of this study was to apply thermogravimetry/mass spectrometry (TG/MS) and pyrolysis-gas chromatography/mass spectrometry (Py-GC/MS) to

determine the thermal stability of vegetable tanning agents and vegetable tanned leathers and analyze the composition of their pyrolysis product mixtures, respectively.
The results of this paper can contribute to a better understanding of vegetable tannins and vegetable tanning chemistry. They show that Py-GC/MS is a valuable tool for identifying the type of tannins in modern and historical leathers. For historical items this micro-destructive technique may be used to comprehend leather technology as well as degradation susceptibility.

2. Experimental

2.1. Samples

The mimosa and tara tanning agents were purchased from Seta S.A. (Brazil), while extracts of quebracho and chestnut wood were sourced from SilvaTeam S.p.a. (Italy). Valonea extract was received from the Leather Engineering Department, Ege University (Turkey). The leather samples were prepared at the Leather and Footwear Research Institute (ICPI) of the National Research and Development Institute for Textiles and Leather (INCDTP), Bucharest using a patented technology designed for bookbinding leather for archival, library and museal use [22].

Vegetable tanned leather was obtained from calf skin tanned using the above mentioned five vegetable tannins. The calf hide was soaked and washed in tapwater with a detergent concentration 0.2 %. After the draining, fleshing and two steps of liming procedures were applied. In order to eliminate the lime from the animal skin ammonium sulfate was used as a deliming compound. Pickling was followed by a few times of agitation and rinsing steps. Calf skin was tanned with vegetable tanning agents in a tank of water containing the tannins. The mass of tannins was equivalent with 10% of untanned hide's mass. The tanned leather was placed on a wooden frame to rest for 48 hours.

2.2. Preparation of gelatin

Gelatin was prepared based on the traditional recipe for parchment glue mentioned by Cennino Cennini in his *Il Libro dell'arte* written in the early fifteenth century: fine calf parchment (untanned hide) shavings were soaked in cold water overnight and then gently boiled at around 70 – 80 ºC for long time in order to obtain a concentrated glue [23].

2.3. Thermogravimetry/mass spectrometry (TG/MS)

The thermogravimetry/mass spectrometry system consists of a modified thermobalance (Perkin Elmer TGS-2) and a quadrupole mass spectrometer (Hiden HAL). Approximately 3 mg tanning agent, tanned leather and gelatin samples were measured under argon atmosphere at a flow rate of 140 ml/min. The samples were heated at a rate of 20 °C/min from 25 to 900 °C in a platinum sample pan. The evolved volatile decomposition products were introduced through a heated capillary into the ion source of the mass spectrometer, which was operated at 70 eV electron energy.

2.4. Pyrolysis-gas chromatography/mass spectrometry (Py-GC/MS)

Approximately 0.6 mg gelatin, tanned leather or 1.6 mg vegetable tanning agents were pyrolyzed at 600 °C for 20 s in helium atmosphere using a Pyroprobe 2000 pyrolyzer interfaced to an Agilent 6890A/5973 GC/MS. The interface and the GC injector were heated to 280 °C. The injector operated in pulsed split mode and the split ratio was chosen 1:20. The pyrolysis products were separated on a DB-1701 capillary column (30 m × 0.25 mm, 0.25 μm film thickness). The GC oven was programmed to hold at 40 °C for 4 min then increase the temperature at a rate of 6 °C/min to 280 °C (hold for 7 min). The mass range of m/z 14−500 was scanned by the mass spectrometer in electron impact mode at 70 eV electron energy. The identification of the molecules is based on NIST 2011 mass spectral library. Three replicates were carried out on each sample.

2.5. Micro hot table (MHT)

Temperature of shrinkage (T_s) of vegetable tanned leather determined by the standard method SR EN ISO 3380-2003 compared to untanned hide.

3. Results and discussion

3.1. TG/MS results

Fig. 1 illustrates the thermogravimetric (TG) and derivative thermogravimetric (DTG) curves of all studied vegetable tanning agents. As Fig. 1a shows the carbonaceous residue (char) of the two condensed tannins (quebracho and mimosa) are higher than that of the hydrolyzable tannins (chestnut, valonea and tara).

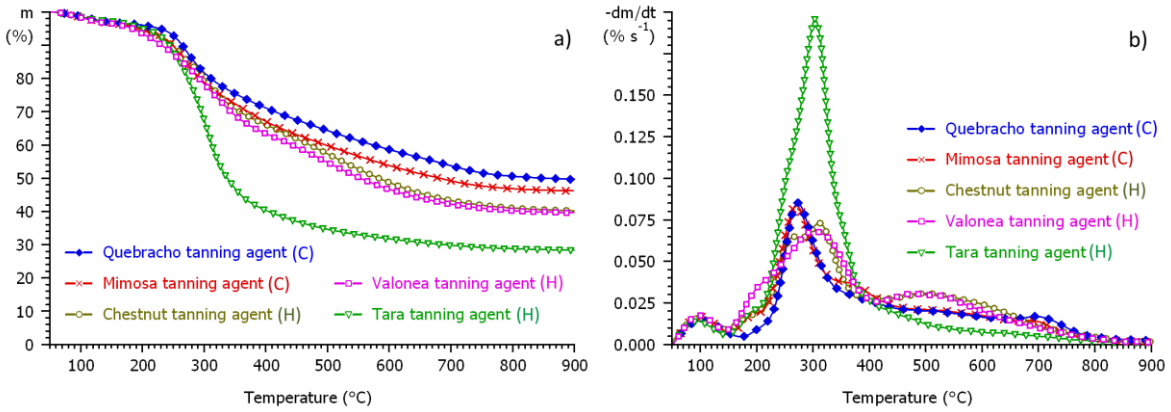

Figure 1 – TG (a) and DTG (b) curves of vegetable tanning agents. The types of tannins are indicated: C: condensed, H: hydrolyzable.

The condensed tannins have condensed structure built from catechin moieties, which are flavonoids consisting of two dihydroxybenzene rings. During the thermal decomposition of this highly aromatic structure, the cross-linking reactions are dominant, which lead to the intensive char formation. Less carbonaceous residue forms during the thermal decomposition of the hydrolyzable tannins due to the more intense fragmentation of the anhydrosugar moieties. The thermal decomposition of the tara tanning agent significantly differs from the

other vegetable tannins; its decomposition rate is much higher in the temperature range of 250–370 °C, which explains the lowest char content.

The thermal decomposition process of the vegetable tanning agents starts with the evaporation of the adsorbed water from 50 to 170 °C as the DTG curves indicate in Fig. 1b. At the same temperatures, a peak can also be seen on the mass spectrometric curve of the *m/z* 18 ion, as will be discussed below. The second DTG peak can be attributed to the thermal decomposition of the organic components in the temperature range of 170–400 °C. Above this temperature, charring reactions take place with the release of small molecules. The DTG curves of the two condensed tannins (mimosa and quebracho) are very similar, indicating their similar chemical structure. The DTG curves of two hydrolyzable tannins (chestnut and valonea) which are both ellagitannins, are very similar, too. Tara, the third hydrolyzable tannin, is a gallotannin, which decomposes quite differently from the other tannins.

Fig. 2 presents the TG and DTG curves of gelatin and vegetable tanned leather samples. Gelatin has the lowest char content, while more carbonaceous residue is formed from the tanned leathers. This behavior was to be expected because gelatin does not contain tannin, so the amount of char produced during its thermal decomposition is the result of collagen decomposition only.

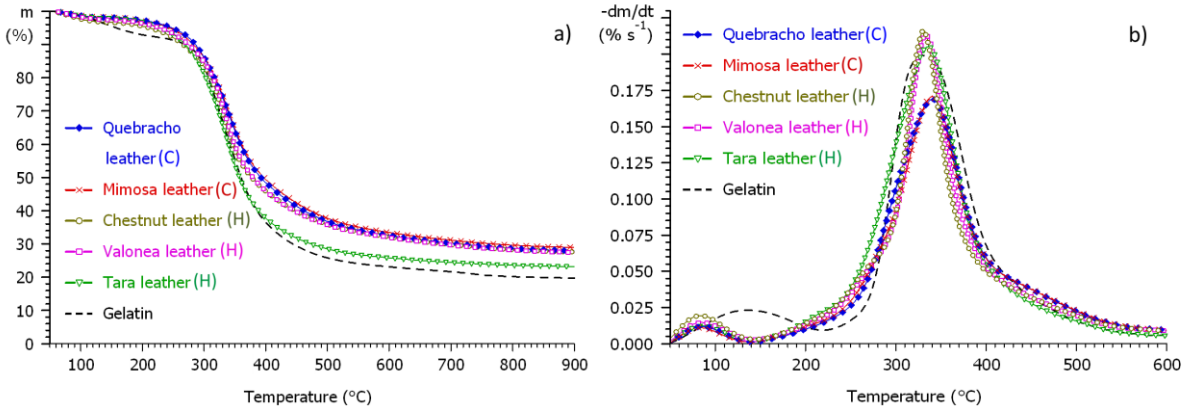

Figure 2 – TG (a) and DTG (b) curves of gelatin and leathers tanned by vegetable tanning agents. The types of tannins are indicated: C: condensed, H: hydrolyzable.

The evaporation of adsorbed water ends to 140 °C during heating up the tanned leathers, while the temperature range of water evolution is extended to 220 °C in the case of gelatin (Fig. 2b). The reason of the elongated water release can be attributed to the presence of the water-mediated hydrogen bonds in the gelatin structure between the charged groups of the amino acid side chains such as lysine, arginine, glutamic and aspartic acids, hydroxyl groups of the side-chains in serine, threonine and hydroxylysine, and the carbonyl and –NH– groups of side chains, while the main polypeptidic chains are not involved in the intramolecular bonding [24,25]. Since, more polar groups are exposed to the solvent in the unfolded collagen structure than in the fibrillar collagen structure; gelatin can bind significantly more water molecules than the original collagen structure. The differences in the evaporation of the adsorbed water of tanned leathers compared to gelatin well correlates to the different degree of ordering of bound water in gelatin and vegetable tanned leather and the different dynamics of water in these two types of collagen-based materials revealed by unilateral Nuclear Magnetic Resonance (NMR) [26].

As the DTG curves show in Fig. 2b, the thermal decomposition of gelatin starts above 220 °C, while the tanned leathers decompose above 170 °C, similarly to the tanning agents. The oxygen-containing functional groups of tannins were probably cleaved, e.g., two hydroxyl groups were condensed by the release of water. The maximum thermal decomposition rate (DTG_{max}) of the leathers tanned with condensed tannins (quebracho and mimosa) is significantly lower than that of leather tanned with hydrolysable tannins and gelatin.
The thermal stability of the samples can be characterized by the temperature of the DTG_{max} (T_{peak}). Fig. 3 shows the T_{peak} data of the studied vegetable tanning agents, vegetable tanned leathers and gelatin.

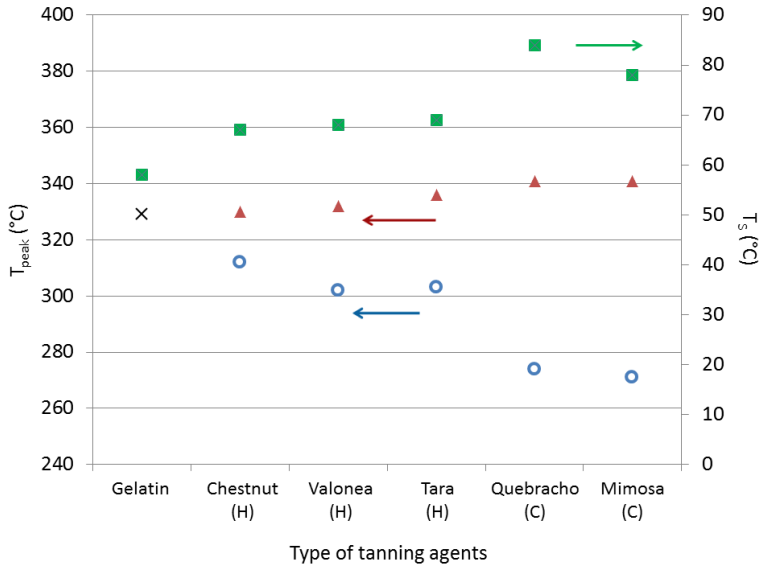

Figure 3 – Temperature of the maximum decomposition rate (T_{peak}) of tanning agents (○), tanned leathers (▲), and gelatin (×) and temperature of the shrinkage (T_S) of gelatin and vegetable tanned leathers (■). The types of tannins are indicated: C: condensed, H: hydrolyzable.

Although the difference between T_{peak} values of the tanned leathers is not high, it is characteristic and reproducible. As expected, T_{peak} trend is parallel with the shrinkage temperature (T_s) trend (Fig. 3). The standard deviation of the T_{peak} data is less than 0.3 °C and that of the T_s data is between 0.9-1.7 °C; leading to a good reproducibility of the measurements. The shrinkage temperature of collagen-based materials is the most commonly quoted measurement of hydrothermal stability. The principle of the method is to suspend the sample (either in the form of a strip or only few fibers) then to heat the water at a rate of 2 ºC min^{-1}, according to official standard method [27] or Micro Hot Table (MHT) method [28], respectively. The shrinkage temperature is noted when the sample visibly shrinks. Less likely, the thermal stability of the vegetable tanning agents and vegetable tanned leathers show opposite trends: the lower the thermal stability of the tanning agent, the higher the thermal stability of the tanned leathers. The higher reactivity of the vegetable tanning agents leads to the higher thermal stability of vegetable tanned leather. The condensed tannins show higher thermal reactivity than the hydrolyzable tannins. They establish stronger interactions with collagen producing more stable leather from a thermochemical point of view. From a chemical point of view, the reason for this behavior could be the stronger chemical bonds by

which the tannin with higher reactivity interacts with skin collagen, thus resulting in more stable leather. This is consistent with what is known about the interaction between the flavonoid tannins and collagen [17]: covalent binding *via* quinoid reactions (link step) and crosslinking (lock step) by an aldehydic reactant (oxazolidine) yield higher hydrothermal stability, i.e. higher T_s, than those yielded by the interaction of pyrogallol tannins with collagen. The difference between the thermal stability of leather tanned with the two different types of tannins comes from the difference in reaction type, i.e. the contribution of covalency to the hydrogen bonds makes a measurable difference to the shrinkage temperature.

Fig. 4 presents the DTG curves and the evolution profiles of the main gaseous products of gelatin (a), leather tanned with valonea tannin (b), and valonea tanning agent (c). The *m/z* 18 ion curve represents the evolution of water, which occurs in two main steps.

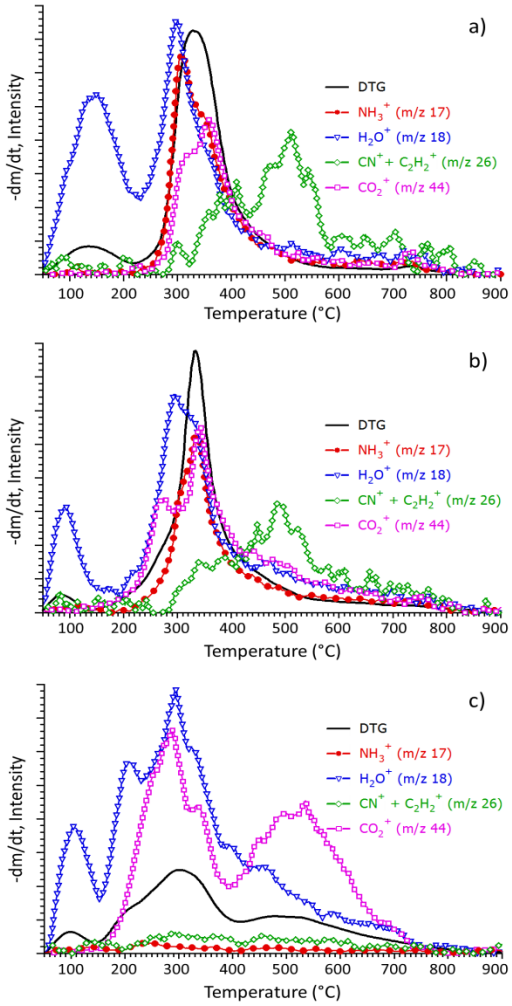

Figure 4 – DTG and mass spectrometric curves of gelatin (a), leather tanned with valonea tanning agent (b), and valonea tanning agent (c). Mass spectrometric curves illustrate the evolution profiles of ammonia (*m/z* 17), water (*m/z* 18), cyanide and acetylene groups (*m/z* 26), and carbon dioxide (*m/z* 44).

The first peak of the *m/z* 18 curve corresponds to the evaporation of adsorbed (less structured or less tightly bond) water. As discussed before, gelatin has a higher amount of adsorbed water comparing to the tanned leather, due to the water-mediated hydrogen bond network.

The more intensive and broader evolution profile of *m/z* 18 up to 220°C in Fig. 4a also supports this explanation. On the other hand, water is released during the whole temperature range of the thermal decomposition process indicating it is an important decomposition product. Numerous hydroxyl groups are present in both the collagen and tanning agent molecules, which apparently release water during decomposition. The evolution profiles of the mass spectrometric curves at *m/z* 17 represent the formation of ammonia in the TG/MS measurements. The main source of ammonia is the amino acid residues of gelatin and leather. The vegetable tanning agents are nitrogen-free substances, therefore no ammonia evolves from this sample. It should be noted that water also has a fragment ion at *m/z* 17; however, its intensity was subtracted from the total intensity of *m/z* 17. By evaluating the formation of several fragment ions, it was established that the ion intensity curve of *m/z* 26 can be divided into two main parts. The first step between 300 and 440 °C can be attributed to the evolution of the cyanide groups (CN^+) originating from the collagen content of leather and gelatin. The second peak of the *m/z* 26 fragment ion in the temperature range of 440–600 °C was identified as $C_2H_2^+$ moieties formed by the charring process. Carbon dioxide (*m/z* 44) above 200 °C originates from the carboxylic groups of collagen and tannin. The intensive CO_2 evolution from the tanning agent observed from 400 to 750 °C can be attributed to the secondary reactions of various oxygen-containing groups [18,29].

3.2. Pyrolysis results

Pyrolysis-gas chromatography/mass spectrometry (Py-GC/MS) experiments have been performed in order to identify the main decomposition products of vegetable tannins and vegetable tanned leathers. On the basis of the TG curves of tannins and leathers (Figs. 1 and 2) the pyrolysis temperature was chosen 400 °C because the main decomposition step of all samples terminates by this temperature. Above this temperature especially the charring processes take place.

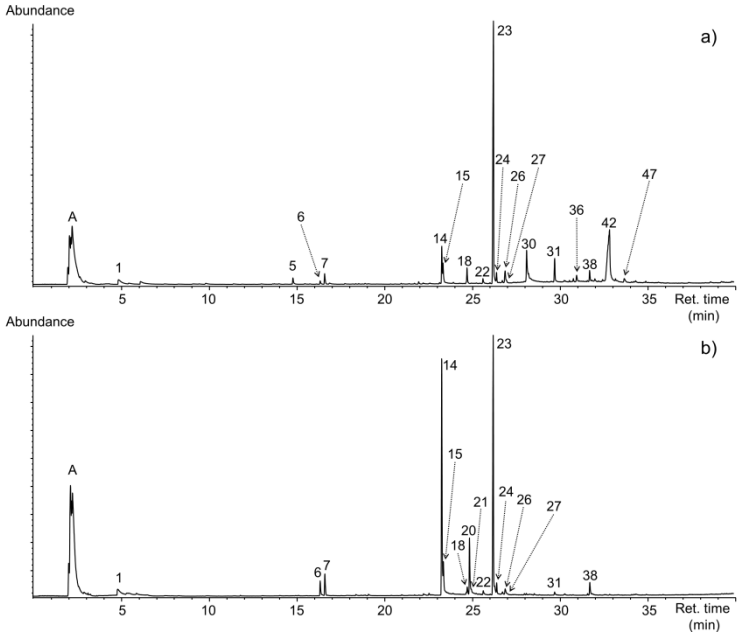

Figure 5 – Pyrograms of the condensed vegetable tannins: mimosa (a) and quebracho (b). Numbered peak identities are given in Table 1.

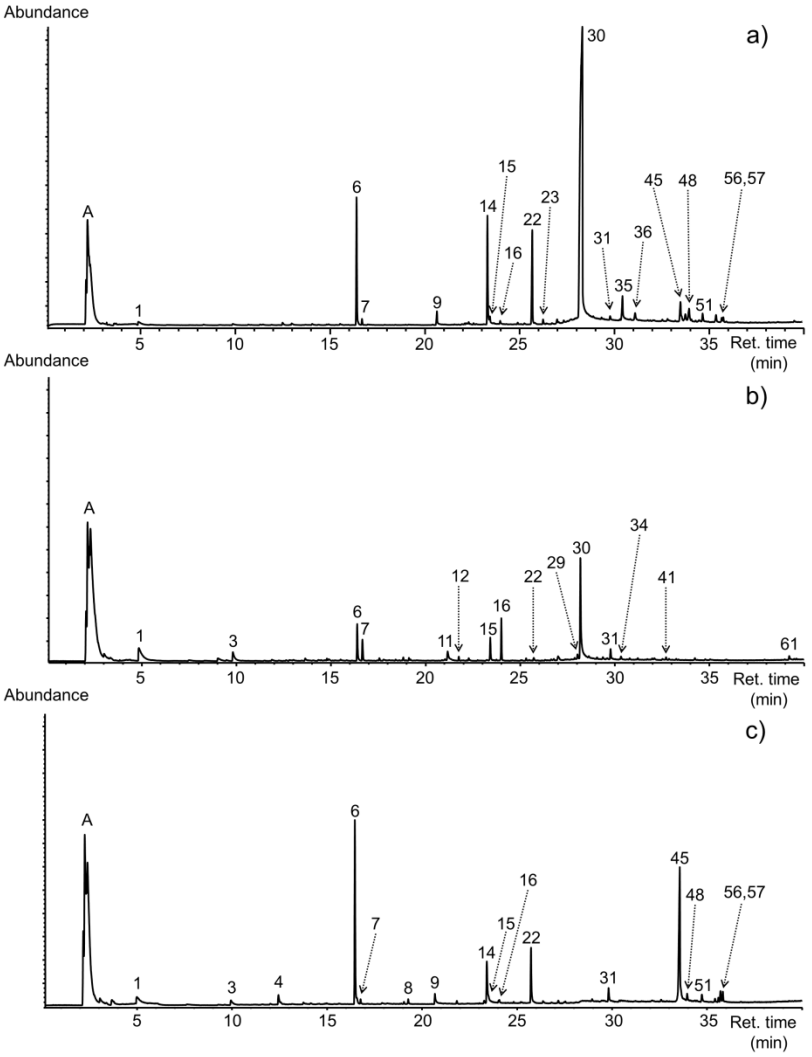

Figure 6 – Pyrograms of the hydrolyzable vegetable tannins: tara (a), chestnut (b), and valonea (c). Numbered peak identities are given in Table 1.

Fig. 5 shows the pyrograms of the volatile products originating from mimosa (Fig. 5a) and quebracho (Fig. 5b) condensed tannins. Fig. 6 presents the pyrograms of the products of hydrolyzable vegetable tannins: tara (Fig. 6a), chestnut (Fig. 6b) and valonea (Fig. 6c). The identification, the most abundant MS ions and the probable sources of the pyrolysis products are listed in Table 1. The first group of unresolved peaks (peak #A) at low retention time corresponds to smaller molecular mass products of the samples. This group consists of mainly carbon dioxide and water formed by the fragmentation of tannin components and moisture evaporation. The evolution of sulfur dioxide was also detected indicating the decomposition of ammonium sulfate content of the tannin samples.

The main higher molecular weight products of both condensed and hydrolyzable tannins were mono- di- and tri-hydroxybenzene compounds. Resorcinol (peak #23) was the main aromatic decomposition product of the condensed tannins; it derives from the dihydroxyphenyl segments of the catechin units. Resorcinol appeared in the pyrograms of tara hydrolyzable tannin in much smaller amount (Fig. 6a) while not detected in the pyrogram of chestnut and valonea tannin (Fig. 6b. and 6c). Phenol (peak #6) released from all the studied condensed

and hydrolyzable tannins in relatively high amounts, indicating the partial dehydration of dihydroxy- and trihydroxyphenyl groups (Figs. 5 and 6). Guaiacol and syringol (peaks #7 and #15) are the main decomposition products of the lignin part of all tanning agents. Lignin monomers originates from quebracho (trans-isoeugenol, dihydro-coniferyl alcohol and syringyl-acetone (peaks #21, #34 and #41)) and chestnut (guaiacyl-acetone (peak #29)). Pyrogallol (peak #30) was the most important decomposition product of tara and chestnut, which reflects its main structural unit of gallotannin, the polymer of gallic acid (Fig. 6a and 6b). Pyrogallol was a significant product of mimosa tannin, as well (Fig. 5a). Hydroquinone (peak #22) was produced especially by the pyrolysis of hydrolyzable extracts (Fig. 6). Small amount of hydroquinone molecules originated from the decomposition processes of the condensed tannins. Catechol (peak #14) formed by the thermal decomposition of condensed and hydrolyzable tannins except chestnut tanning agent. Methylcatechol (peak #20) was released only from quebracho tannin. Catechol (peaks #14) and methylated dihydroxybenzenes (peaks #20 and 26) can be formed by the scission of pyran rings of the catechin units of condensed tannins.

The mimosa tanning agent was extracted from the wood bark, which contains about 26% organic non-tannin components. As the pyrogram in Fig. 5a indicates, polyols and carbohydrates are also present in the tanning agent. Mono-O-methyl inositol (peak #42) is the characteristic polyol compound and levoglucosan (peak #31) are typical pyrolysis products of carbohydrates [30]. Carbohydrate products were also formed during the thermal decomposition of all hydrolyzable tannins (Fig. 6), which originated from the sugar moieties (peaks #3, #12, #36, #51, #56 and #57). Two isomers of quinic acid (peak #45 and #48) were identified on the chromatograms of tara and valonea hydrolyzable tanning agents. The main decomposition products of sugars in inert atmosphere are anhydrosugars. Levoglucosan (1,6-anhydro-β-D-glucopyranose, peak #31) was formed by the pyrolysis of glucose molecules, so the presence of this decomposition product indicates the glucose content of tannins [31,32]. These results lead to the conclusion that the condensed type mimosa extract contains glucose units and cyclic sugar alcohol, as well. Cyclitol (quinic acid) was identified on the chromatograms of the hydrolyzable tara and valonea tannins.

Fig. 7 shows the pyrograms of gelatin (Fig. 7a) and leathers tanned with mimosa (Fig. 7b), quebracho (Fig. 7c) and tara (Fig. 7d) tanning agents. The decomposition pattern of gelatin and leathers tanned with both condensed and hydrolyzable tanning agents was very similar, indicating the dominance of the decomposition products of amino acid residues in the pyrolyzates.

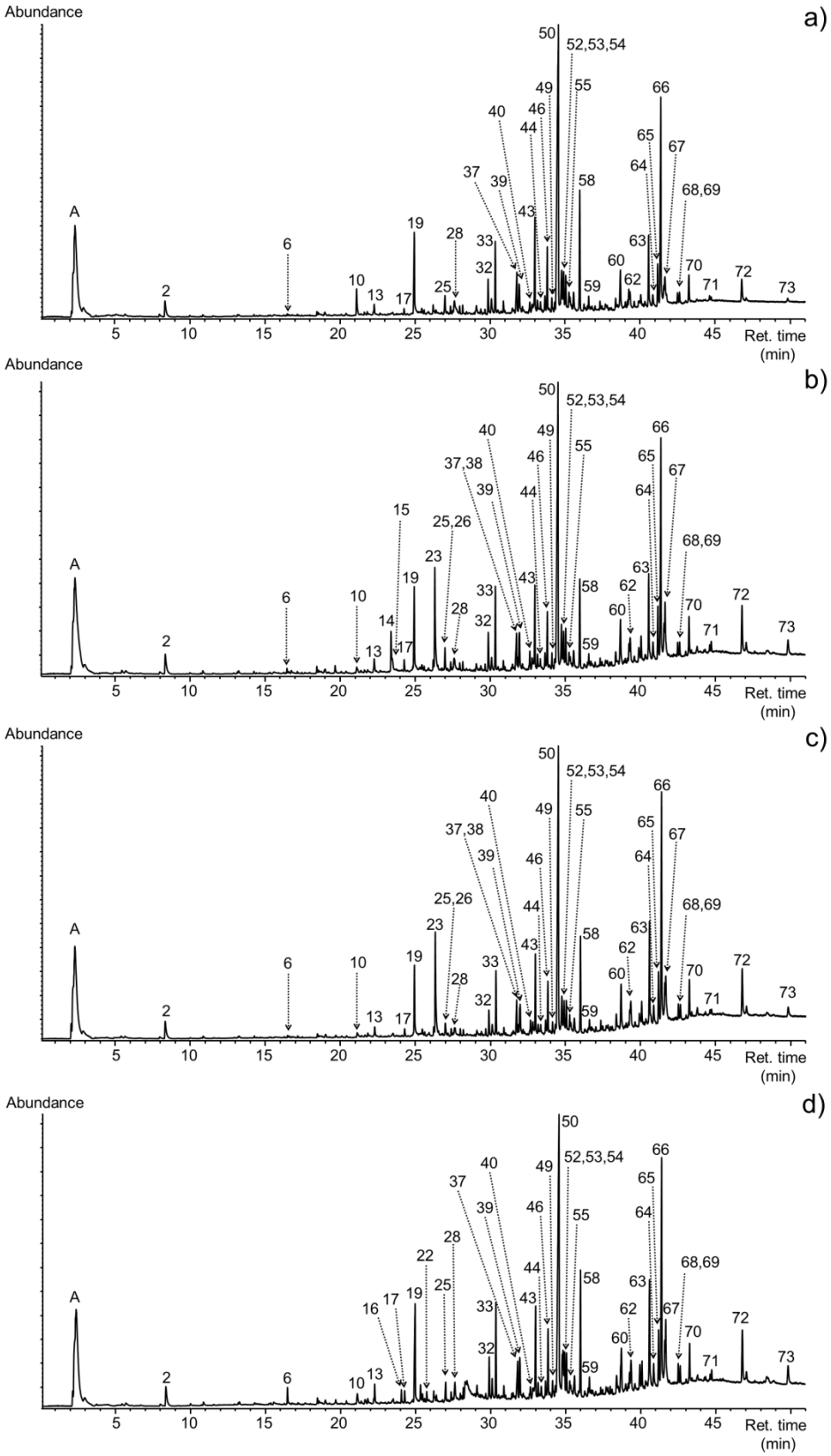

Figure 7 – Pyrograms of gelatin (a) and leathers tanned with mimosa (b), quebracho (c), and tara (d) tannins. Numbered peak identities are given in Table 1.

Table 1 – The main decomposition products released in the Py-GC/MS experiments from vegetable tanning agents, gelatin, and vegetable tanned leathers. Peak numbers refer to Figs. 5, 6 and 7.

Peak #	Ret time (min)	Compound	*m/z*	MW (Da)	Source
A	2.20-2.70	Gaseous products			
		Carbon dioxide	44, 28	44	T, G, L
		Sulfur dioxide	64, 48	64	T, G, L
		Water	18, 17	18	T, G, L
		Ammonia	17, 16	17	G, L
		Hydrogen cyanide	26, 27	27	G, L
1	4.96	Acetic acid	43, 45, 60	60	T
2	8.31	Pyrrole	67, 41, 39	67	G, L
3	9.87	Furfural	96, 95, 39	96	CT, VT
4	12.39	p-Benzoquinone	108, 54, 82	108	VT
5	14.90	3-Methyl-2,5-furandione	39, 68, 40	112	MT
6	16.45	Phenol	94, 66, 64	94	T, G, L
7	16.70	Guaiacol	109, 124, 81	124	T
8	19.23	Succinic anhydride	28, 56, 26	100	VT
9	20.67	Benzoic acid	105, 122, 77	122	TT, VT
10	21.10	Succinimide	99, 56, 70, 42	99	G, L
11	21.21	3-Pyridinol	95, 39, 68	95	CT
12	21.79	1,4:3,6-Dianhydro-α-D-glucopyranose	69, 57, 98	114	CT
13	22.26	1H-Pyrrole-3-carbonitrile	92, 65, 64	92	G, L
14	23.37	Catechol	110, 64. 81	110	MT, QT, QL, TT, VT
15	23.45	Syringol	154, 139, 111	154	CT, MT, QT, QL, TT, VT
16	24.04	2,2-bis(4'-Methoxyphenyl)propane	241, 256	256	TT, TL, CT, CL, VT, VL
17	24.26	1,3-Cyclopentandione	98, 42, 56	98	G, L
18	24.81	2,2'-Bifuran	134, 105, 78	134	MT, QT
19	24.91	Pyrrole-2-carboxamide	110, 94, 93	110	G, L
20	24.95	4-Methyl-1,2-benzenediol	124, 123, 78	124	QT
21	25.06	trans-Isoeugenol	164, 149, 131	164	QT
22	25.72	Hydroquinone	110, 81, 82	110	T, TL
23	26.29	Resorcinol	110, 82, 81	110	MT, ML, QT, QL, TT
24	26.50	Trimethyl-benzaldehyde	147, 148, 119	148	MT, QT
25	26.96	Glycine-glycine DKP	114, 86, 71	114	G, L
26	26.99	Orcinol (5-methyl-1,3-benzenediol)	124, 123, 95	124	MT, ML, QT, QL
27	27.01	Methoxy-1,4-benzenediol	140, 125, 97	140	MT, QT
28	27.58	2,4-Imidazolidinedione	100, 72, 57	100	G, L
29	28.06	Guaiacyl acetone	137, 180, 122	180	CT
30	28.21	Pyrogallol	126, 52, 80	126	MT, CT, TT
31	29.81	Levoglucosan (1,6-anhydro-β-D-glucopyranose)	60, 72, 144	162	T
32	29.87	Unidentified aromatic	136, 107, 108	n.d.	G, L

33	30.34	Pyrocoll	186, 93, 65	186	G, L
34	30.36	Dihydro-coniferyl alcohol	137, 182, 136	182	CT
35	30.51	3-Hydroxy-benzoic acid	138, 121, 93	138	TT
36	31.06	Unknown sugar compound	87, 73, 57	n.d.	MT, TT
37	31.72	2,5-Dioxo-3-methylpiperazine	85, 128, 43, 57	128	G, L
38	31.80	Unidentified aromatic	150, 122, 94	150	MT, ML, QT, QL
39	31.95	2 Unidentified aromatics	94, 164 + 171, 172	n.d.	G, L
40	32.64	Unidentified aromatic	185, 94, 66	n.d.	G, L
41	32.74	Syringyl acetone	210, 167	210	CT
42	32.94	Mono-O-methyl inositol	87, 85, 73	194	MT
43	32.97	Proline-alanine DKP	70, 97, 125, 168	168	G, L
44	33.35	Glycine-valine DKP	114, 85, 57	156	G, L
45	33.56	Quinic acid	60, 57, 70	192	TT, VT
46	33.80	Proline-alanine DKP	70, 97, 125, 168	168	G, L
47	33.83	Unknown sugar compound	87, 85, 41	n.d.	MT, TT
48	34.02	Quinic acid	60, 57, 70	192	TT, VT
49	34.11	Proline-valine DKP	154, 70, 72, 125	196	G, L
50	34.50	Proline-glycine DKP	111, 154, 83	154	G, L
51	34.72	Unknown sugar compound	73, 71, 60	n.d.	TT, VT
52	34.73	4-Methyleneproline	84, 41, 56	127	G, L
53	34.88	5-Methyl-5-(2-methylpropyl)-2,4-imidazolidinedione	114, 113, 59	170	G, L
54	35.03	Proline-arginine-(CN_3H_4) DKP + Unidentified	70, 125, 154 + 94, 204, 189	196 + n.d.	G, L
55	35.25	Proline-proline-H_2 DKP	191, 192, 70, 94	192	G, L
56	35.70	Unknown sugar compound	60, 71, 57	n.d.	TT, VT
57	35.83	Unknown sugar compound	70, 42, 55	n.d.	TT, VT
58	35.97	Proline-proline DKP	70, 194, 96, 138	194	G, L
59	36.57	Proline-isoleucine DKP	154, 70, 125, 86	210	G, L
60	38.67	Unidentified DKP	184, 86, 113, 141	n.d.	G, L
61	39.24	4,4'-Methylenebisphenol	200, 107, 199	200	CT
62	39.32	Unidentified DKP	184, 86, 113, 141	n.d.	G, L
63	40.57	Proline-hydroxyproline DKP	70, 210, 86, 124	210	G, L
64	40.86	2,5-Diisopropylimidazolidin-4-one	170, 86, 140	170	G, L
65	41.17	2,5-Diisopropylimidazolidin-4-one	170, 86, 140	170	G, L
66	41.37	Proline-hydroxyproline DKP	70, 210, 86, 124	210	G, L
67	41.59	2,5-Diisopropylimidazolidin-4-one	170, 86, 140	170	G, L
68	42.49	Proline-phenylalanine DKP	125, 153, 244	244	G, L
69	42.63	Proline-phenylalanine DKP	125, 153, 244	244	G, L
70	43.25	Proline-pyroglutamine DKP	70, 208, 96	208	G, L
71	44.75	δ-(3,5)-Cholestadiene	368, 353, 147	368	G, L
72	46.79	Aromatic compound	162, 134, 94	n.d.	G, L
73	49.85	(3β)-Cholesta-5-en-3-ol	386, 368, 275	386	G, L

n.d.: not determined

DKP: 2,5-diketopiperazine

*T: vegetable tannins, G: gelatin, L: vegetable tanned leathers, TT: tara tannin, QT: quebracho tannin, MT: mimosa tannin, CT: chestnut tannin, VT: valonea tannin, VL: valonea leather, ML: mimosa leather, CL: chestnut leather, TL: tara leather and QL: quebracho leather

The first peak of the pyrograms (marked by #A) comprised simple gaseous molecules and water. Ammonia molecules appeared besides carbon dioxide, sulfur dioxide and water molecules. Ammonia is a typical pyrolysis products of the proteins built of amino acids. The main decomposition products of leather and gelatin samples were 2,5-diketopiperazines (DKP), cyclic dipeptides formed from the amino acid residues during the pyrolysis. These decomposition products are eluting after 27 min (peaks #25, #43, #44, #46, #49, #50, #54, #55, #58-60, #62, #63, #66 and #68-70). The formation mechanisms of these compounds were discussed in detail by Voorhees [33] and Fabbri *et al.* [34]. As previously reported for leather and parchment [18], DKPs are formed from the four most abundant amino acid residues in collagen, i.e. glycine, proline, alanine, and hydroxyproline. The other nitrogen-containing molecules (peaks #2, #10, #13, #19, #28, #33, #37, #52, #53, #64, #65 and #67) are the thermal decomposition products of the amino acid residues, as well. The most likely source of pyrrole (peak #2) and its derivative (peak #19) are proline and hydroxyproline units of collagen. Phenol (peaks #6) originates from both the side groups of amino acid residues and tanning agent compounds. The small peaks (peaks #71 and #73) seen in the chromatograms of gelatin and vegetable tanned leathers (Fig. 7.) at the retention time of 45 and 50 min are δ-(3,5)-cholestadiene and (3β)-cholesta-5-en-3-ol, decomposition products of the cholesterol. The cholesterol content of the gelatin was largely reduced by the preparation process so the peaks of the 2 cholesterol compounds are less intensive on the chromatogram of the gelatin (Fig. 7a).

Significant amount of resorcinol (peak #23) and lower yield of orcinol (5-methyl-resorcinol) (peak #26) were released during pyrolysis of both leather samples tanned with condensed tanning agents, namely mimosa and quebracho (see Fig. 7b and 7c). This decomposition product indicated the presence of tannin, as resorcinol is one of the main pyrolysis products of the condensed quebracho and mimosa tannin extracts.

The pyrograms of the three leather samples tanned with hydrolyzable tanning agents, namely tara, chestnut, and valonea, were found to be almost identical, therefore only the pyrogram of tara tanned leather is presented in Fig. 7d. As a specific decomposition products of the tanning agents 2,2-bis(4'-methoxyphenyl)propane (peak #16) [35] appeared among the volatiles. Syringol (peak #15) can be found among the pyrolyzates of the quebracho tannin and quebracho tanned leather.

4. Conclusions

The thermal decomposition of hydrolyzable (tara, chestnut, and valonea) and condensed (quebracho and mimosa) tannins as well as of calf leathers tanned with these vegetable tanning agents were studied by TG/MS and Py-GC/MS. Calf gelatin was chosen as a reference material for the untanned leather.

Due to their condensed structure, condensed tannins produced lower amount of volatile materials and higher amount of char than hydrolyzable tannins. Comparison of the thermal decomposition temperatures of the vegetable tanning agents and vegetable tanned leathers showed that the lower the thermal stability of the tanning agent, the higher the stability of the tanned leather. The tannins with higher reactivity can establish stronger chemical bonds with collagen producing more stable leather from a thermochemical point of view.

The decomposition of tanning agents started at similar temperatures when analyzed as such (i.e. powder extract) or bound to collagen in leather, as indicated by the evolution curves of water and carbon dioxide and by the DTG curves in TG/MS experiments. The evolution of water and carbon dioxide from leather started at lower temperature than from gelatin pointing out to the decomposition of the tannin moieties at about 200 °C.

The chemical composition of the thermal decomposition products of vegetable tanning agents was studied by Py-GC/MS. Mono-, di-, and tri-hydroxybenzenes were identified as the main decomposition products of all studied tannins during pyrolysis; some of these products were also evolved from the tanned leathers. Polysaccharide derivatives as levoglucosane and furan derivatives were identified among the decomposition products of all hydrolyzable tannins and mimosa tannin. Relatively high amount of monomethyl inositol was released from the mimosa tanning agent, which was apparently present among the non-tannin components of the material.

Resorcinol and orcinol was identified as a marker compound of the condensed tannins among the pyrolysis products of leathers tanned with mimosa and quebracho. Bisphenol derivative (2,2-(4'-methoxyphenyl)propane) can be named as a marker molecule of the hydrolyzable tannins. Py-GC/MS is a feasible analytical technique to identify the type of tanning agents by the analysis of vegetable tanned leather.

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References

1. E. Alptekin, M. Canakci, H. Sanl i, Evaluation of leather industry wastes as a feedstock for biodiesel production, Fuel 95 (2012) 214-220.
2. M.G. Medeiros, A.S. Rodrigues, M.C. Batoréu, A. Laires, J. Rueff and A. Zhitkovich, Elevated levels of DNA–protein crosslinks and micronuclei in peripheral lymphocytes of tannery workers exposed to trivalent chromium, Mutagen. 18 (2003) 19–24.
3. L.S. McNeill, J.E. McLean, J.L. Parks, M.A. Edwards, Hexavalent chromium review, part 2: Chemistry, occurrence, and treatment, J. Am. Water Works Assoc. 104 (2012) E395-E405.
4. B. Basaran, M. Ulaș, B.O. Bitlisi, A. Aslan, Distribution of Cr (III) and Cr (VI) in chrome tanned leather, Indian J. Chem. Technol. 15 (2008) 511-514.
5. P. Frutos, G. Hervás, F.J. Giráldez, A.R. Mantecón, Review. Tannins and ruminant nutrition, Span. J. Agric. Res. 2(2) (2004) 191-202.
6. K. Khanbabaee, T. van Ree, Tannins: Classification and definition, Nat. Prod. Rep. 18 (2001) 641–649.
7. C. Carşote, E. Badea, L. Miu, G. Della Gatta, Study of the effect of tannins and animal species on the thermal stability of vegetable leather by differential scanning calorimetry, J. Therm. Anal. Calorim. 124(3) (2016) 1255-1266.

8. A. Ricci, M.-C. Lagel, G.P. Parpinello, A. Pizzi, P.A. Kilmartin, A. Versari, Spectroscopy analysis of phenolic and sugar patterns in a food grade chestnut tannin, Food Chem. 203 (2016) 425-429.
9. H.R. Tang, R.A. Hancock, A.D. Covington, Study on the composition and structure of commercial chestnut tanning agent, Basic Life Sci. 59 (1992) 221–243.
10. E. Onem, G. Gulumser, M. Renner, A.L. Oelbermann, O. Yesil-Celiktas, Pressurized fluid extraction (PFE) of valonea tannin with binary H_2O–CO_2 and ternary H_2O–CH_3OH–CO_2 systems and phase equilibrium studies, Sep. Purif. Technol. 146 (2015) 101-107.
11. E. Haslam, R.D. Haworth, P.C. Keen, Gallotannins. Part VII. Tara gallotannin, J. Chem. Soc. (1962) 3814-3818.
12. P. Schofield, D.M. Mbugua, A.N Pell, Analysis of condensed tannins: a review, Anim. Feed Sci. Technol. 91(1-2) (2001) 21-40.
13. H. Pasch, A. Pizzi, K. Rode, MALDI-TOF mass spectrometry of polyflavonoid tannins, Polym. 42 (2001) 7531–7539.
14. U.H. Abdullah, A. Pizzi, K. Rode, L. Delmotte, X. Zhou, H.R. Mansouri, Mimosa tannin resins for impregnated paper overlays, Eur. J. Wood Prod. 71 (2013) 153–162.
15. P.B. Venter, M. Sisa, M.J. van der Merwe, S.L. Bonnet, J.H. van der Westhuizen, Analysis of commercial proanthocyanidins. Part 1: The chemical composition of quebracho (*Schinopsis lorentzii* and *Schinopsis balansae*) heartwood extract, Phytochem. 73 (2012) 95-105.
16. A.D. Covington, Tanning chemistry: The science of leather, R. Soc. Chem. Publ., Cambridge (2011) 432-443.
17. E. Badea, C. Şendrea, C. Carşote, A. Adams, B. Blümich, H. Iovu, Unilateral NMR and thermal microscopy studies of vegetable tanned leather exposed to dehydrothermal treatment and light irradiation, Microchem. J. 129 (2016) 158-165.
18. Z. Sebestyén, Zs. Czégény, E. Badea, C. Carsote, C. Sendrea, E. Barta-Rajnai, J. Bozi, L. Miu, E. Jakab, Thermal characterization of new, artificially aged and historical leather and parchment, J. Anal. Appl. Pyrolysis 115 (2015) 419-427.
19. K.G.J. Nierop, C.M. Preston, J. Kaal, Thermally assisted hydrolysis and methylation of purified tannins from plants, Anal. Chem. 77 (2005) 5604–5614.
20. J. Kaal, K.G.J. Nierop, P. Kraal, C.M. Preston, A first step towards identification of tannin-derived black carbon: Conventional pyrolysis (Py–GC–MS) and thermally assisted hydrolysis and methylation (THM–GC–MS) of charred condensed tannins, Org. Geochem. 47 (2012) 99-108.
21. A. Marcilla Gomis, A.N. García, M. León, E. Bañón, P.M. Artínez, Characterization of commercially available leathers using thermogravimetric analysis and PY/GC-MS system, J. Am. Leather Chem. As. 107 (2012) 220-230
22. L. Miu, V. Bratulescu, C. Gaidau, V. Bocu, O. Niculescu, Piele naturala pentru legatorie carte de patrimoniu si procedeu de realizare a acesteia, Romanian patent C14C/2006. Available at p.31 at http://www.osim.ro/publicatii/brevete/bopi_2006/bopi0306.pdf
23. L. Broecke, Cennino Cennini's Il Libro Dell'Arte, Archetype Publications, London 2015.

24. D.H. Chou, C. Morr, Protein-water interactions and functional properties, J. Am. Oil Chem. Soc. 56 (1979) 53-62.
25. E.M. Bradbury, R.E. Burge, J.T. Randallan, D.G.R. Wilkinson. The polypeptide chain configurations of native and denatured collagen fibres, Discuss. Faraday Soc. 25 (1958) 173-185.
26. C. Sendrea, C. Carsote, E. Badea, A. Adams, M. Niculescu, H. Iovu, Non-invasive characterisation of collagen-based materials by NMR-MOUSE and ATR-FTIR, U.P.B. Sci. Bull., Series Chemistry 78 (2016) 27-38.
27. J.M.V. Williams, IULTCS (IUP) Test methods. J. Soc. Leather Technologists Chemists 84 (2000) 359-362.
28. R. Larsen, Experiments and observations in the study of environmental impact on historical vegetable tanned leathers. Thermochim. Acta 365 (2000) 85-99.
29. C. Di Blasi, Modelling and simulation of combustion processes of charring and non-charring solid fuels, Prog. Energy Combust. Sci. 19 (1993) 71-104.
30. O. Faix, I. Fortmann, J. Bremer and D. Meier, Thermal degradation products of wood. A collection of elecfron-impact (EI) mass spectra of polysaccharide derived products, Holz als Roh- und Werkstoff 49 (1991) 299-304.
31. M. Gaugler, W.J. Grigsby, Thermal degradation of condensed tannins from radiate pine bark, J. Wood Chem. Technol. 29 (2009) 305-321.
32. J.M. Garro Galvez, B. Riedl, A.H. Conner, Analytical studies on tara tannins, Holzforschung 51 (1997) 235-243.
33. K.J. Voorhees, W. Zhang, A.D. Hendricker, B. Murugaverl, An investigation of the pyrolysis of oligopeptides by Curie-point pyrolysis-tandem mass spectrometry, J. Anal. Appl. Pyrolysis 30 (1994) 1–16.
34. D. Fabbri, A. Adamiano, G. Falini, R. De Marco, I. Mancini, Analytical pyrolysis of dipeptides containing proline and amino acids with polar side chains. Novel 2,5-diketopiperazines markers in the pyrolysates of proteins, J. Anal. Appl. Pyrolysis 95 (2012) 145–155.
35. S. Tsuge, H. Othani, C. Watanabe, Pyrolysis-GC/MS data book of synthetic polymers, Pyrograms, thermograms and MS of pyrolyzates, Elsevier, Oxford UK (2011)