

Article

Experimental Study on Chrome Tanned Leather Shavings Modification—Properties and Prospective for Future Application

Dorota Gendaszewska * , Paulina Pipiak, Dorota Wieczorek and Katarzyna Sieczyńska

Lukasiewicz Research Network—Lodz Institute of Technology, 73 Zgierska Str., 91-463 Lodz, Poland; paulina.pipiak@lit.lukasiewicz.gov.pl (P.P.); dorota.wieczorek@lit.lukasiewicz.gov.pl (D.W.); katarzyna.sieczynska@lit.lukasiewicz.gov.pl (K.S.)

* Correspondence: dorota.gendaszewska@lit.lukasiewicz.gov.pl

Abstract: Chrome tanned leather shavings (CTLS) are considered one of the more difficult wastes to manage in the tanning industry. At the same time, this waste is an important source of good quality collagen. The few methods described in the literature for the valorization of these wastes usually require pre-treatment or activation. In this study, an attempt was made to see if raw chrome tanned leather shavings (CTLS) could be modified to obtain secondary raw materials with new physical properties. Glutaraldehyde, glycerol, EDCs, pectin, and tannins were selected for modification of the CTLS. The effectiveness of the processes carried out was confirmed by FTIR analysis of the materials obtained. Changes in the intensity of the characteristic collagen peak (amide I, amide II, and amide III) and changes in the position of the bands in the 1200–1000 cm^{-1} region were observed in the recorded spectra. The modifications introduced confirmed changes in the physical properties of samples M1–M5, including an increase in bulk density from 0.15 to 0.59 g cm^{-3} , and improved tensile strength in some cases. The elemental content results of the samples tested showed the resulting modified CTLS were free of Cd, Hg, Pb, and Cr(VI). It was also confirmed tanning waste contained significant amounts of various valuable elements. The research also included preliminary tests to assess the environmental impact of the modified materials. In a phytotoxicity test conducted on modified CTLS, M1 with pectins showed the highest root stimulation (105%), while samples with glutaraldehyde and unmodified CTLS showed significant root inhibition (75%, 74%). Other samples showed moderate plant toxicity and seed germination was not significantly affected.

Keywords: chrome tanned leather shavings; phytotests; heavy metal; FTIR analysis



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1. Introduction

The tanning industry is considered one of the most important industries due to its economic, employment, and export potential. However, it is worth noting the leather industry generates approximately 600,000 tonnes of solid waste per year worldwide [1,2]. Uncontaminated or slightly contaminated waste fractions produced before the tanning process could be recovered in other industrial sectors such as the production of gelatine, sausage casings, yarns, sponges, artificial leather, adhesives, cosmetics, and technical fats for animal feed [3–7]. A much more challenging issue is the management of the wastes generated after the proper tanning process, which are resistant to biological degradation and often contain chemical contaminants. Traditional methods of disposing of these wastes include landfilling and incineration, resulting in a significant waste of resources and potential harm to the environment [8]. Waste leather shavings consist mainly of collagen (at least 90%), which is contaminated with saline compounds (at least 4%) used in the tanning process (e.g., chrome alum pH = 3.5; 20 °C/chrome B pH (1:10) may be 1.5–2.5) [6,9]. Moreover, shavings have a very low bulk density and appear in irregular shapes. As a result, the material has a large volume, which causes problems in transport and storage [10]. The above disadvantages make it difficult to manage the chrome-tanned solid waste from

the leather industry in its untreated form. However, there are a number of reports that describe the direct management of this type of waste [2,6,11–15]. Some of the research focuses on the possibility of energy recovery from these wastes using thermal processes such as combustion, pyrolysis, and gasification [2,6]. The use of tanning industry waste for the production of composite materials is also described in the literature. In this case, CTLS or leather dust is mixed with binders such as: thermoplastic or thermosetting polymers, elastomers [13–15].

On the other hand, there are known solutions for the management of tannery waste that involve the isolation and/or activation of valuable secondary raw materials (e.g., collagen hydrolysate, nutrients, fat). Studies on the modification of collagen derived from tannery waste were reported in the literature, but usually the first step was to carry out acid, alkaline, or enzymatic hydrolyses of the raw materials to obtain a collagen hydrolysate with the desired protein/amino acid content [16]. One possible method often used to modify biopolymers was cross-linking. Additional cross-linking bonds introduced into the biopolymer matrix led to qualitative and/or quantitative, permanent, or reversible interactions that directly affected their new chemical and physical properties [17–19]. Chemical cross-linkers include aldehydes (e.g., glutaraldehyde) [20], glycerol [21], epoxides, acids, acyl azides, and carbodiimides [22,23]. In contrast, compounds of natural origin with cross-linking properties are polyphenols [24]. Sources of polyphenols are usually pectin [25], tannic acids [26], hydrolysed tannin, or fruit pomace. A common feature of polyphenols is the presence of phenolic groups in their molecules. Due to the nature of the chemical reactions that take place, curing agents can be divided into two groups: compounds that are part of the curing bond and compounds that are not incorporated into the system. The first group may include glutaraldehyde, which was the most efficient cross-linking agent among the compounds included in the aldehydes [27]. In a pH-neutral environment, glutaraldehyde reacts violently with amine groups to form thermally and chemically stable bonds [28]. The second group of cross-linking agents are compounds that cause cross-linking bonds to form without being incorporated into the cross-linking. This group includes acyl azides and carbodiimides. The most commonly used is 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDC) [29]. The carbodiimide reagent allows the formation of cross-links between carboxyl groups and amide groups of collagen. This results in a biocompatible and non-cytotoxic composite that greatly enhances the physicochemical properties of collagen [30]. Another potential compound that could alter collagen's properties was glycerol. Its hygroscopic properties and ability to form hydrogen bonds mean it is used as a plasticiser to enhance polymer elasticity [31]. Polyphenols represent another group of compounds that can potentially act as collagen modifiers. They exhibit both antioxidant properties and high compatibility with proteins [25]. Wu et al. 2019 studied how polyphenol derivatives (such as tannic acid, epigallocatechin gallate, and N-2-(3,4-dihydroxyphenyl)ethyl acrylamide) cross-linked with collagen. The results showed all polyphenol derivatives cross-linked collagen mainly by non-covalent bonding under acidic non-oxidized conditions and by covalent bonding under alkaline oxidized conditions [32]. Carrying out these operations in this way increases the energy intensity of the entire waste treatment process and requires the use of many reagents and appropriate equipment. This reduces the potential economic benefits of waste management and the implementation of a closed-loop system for raw materials [17].

Therefore, the aim of this study was to investigate the possibility of modifying chrome tanned leather shavings (CTLS) to obtain secondary raw materials with new physical properties. The essence of the study was to carry out the treatment of collagen-rich CTLS, without their prior isolation of protein components from the waste and without their pre-activation. These objectives set the direction for future research and a strategy for further activities, with the aim of developing an efficient and economical method of waste treatment for use in various areas of the economy, e.g., as an additive to bitumen. Substances commonly used in the literature as collagen cross-linking reagents, i.e., glutaraldehyde, glycerol, EDCs, pectin, and tannins, were selected to test the potential for CTLS modification.

As part of the work, preliminary tests were carried out on the physical and chemical properties of the raw materials obtained. The study also considered the environmental aspect by carrying out phytotoxicity tests, which allowed a preliminary assessment of the level of toxicity of the raw and modified materials considered.

2. Materials and Methods

2.1. Waste Material (CTLS)

Chrome tanned leather shavings (CTLS) were obtained after the wet blue tanning process from the tannery in Poland (M0 sample). The moisture content of the CTLS used in the process was 40–50% with an average of 48% by weight. The fibers of CTLS are between 0.5 and 10 cm long with an average of 4 cm long (fiber fractionation method). The pH in aqueous solution (ratio 1:10) was 3.86 (potentiometric method). CTLS was characterized by the following parameters: the amount of total ash (slowly ashing) at the level of 8.6%, the amount of nitrogen (Kjeldahl method) at the level of 16.5%, and the amount of chromium (III) oxide (total chrome ICP-OES and chromium (VI) spectrophotometric method) at the level of 4.4% according to the literature [5].

2.2. Modification of Waste Material

The following reagents were used to modify the CTLS: pectin (Ecobi, Poland), glutaraldehyde (Pol-Aura, Poland), glycerol (Pol-Aura, Poland), EDC—1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (Pol-Aura, Poland), and tannin (Browin, Poland). Aqueous solutions of each compound were prepared separately. Two liters of each solution were added to 1 kg of wet (48% moisture) chrome-tanned leather shavings (CTLS) and mixed for 30 min. The samples were subsequently dried to a constant weight at 100 °C. Finally, the samples were ground on a laboratory mixer (stainless steel, engine speed: 28,000 r min⁻¹, power: 2000 W, capacity: 1000 g, dimensions: 19 cm × 13 cm × 36 cm). Differences in the amounts of additives resulted from their individual form and data from the literature [20,25,33–36]. Table 1 shows the data on modified samples. Figure 1 shows modified chrome tanned leather shavings under the microscope.

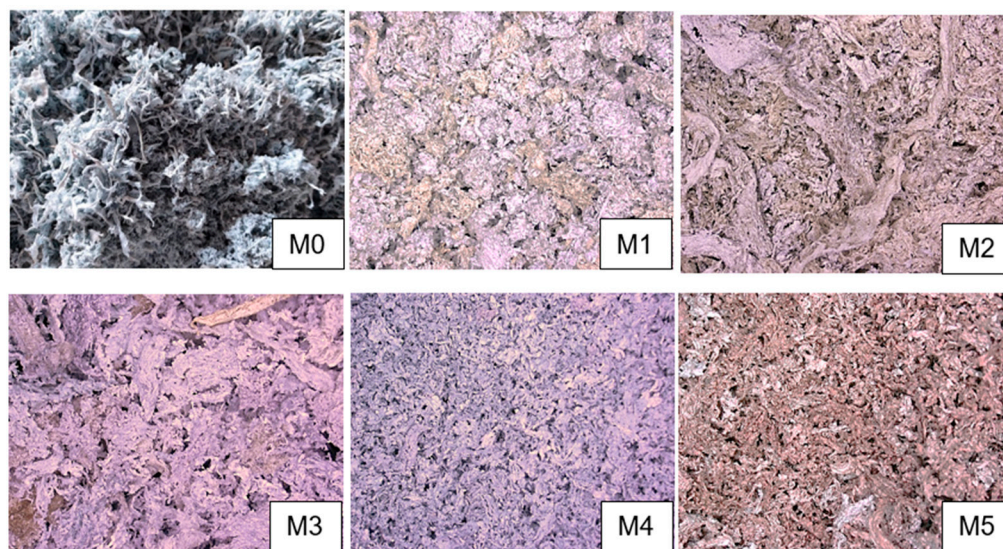


Figure 1. Optical microscope images (20× magnification): M0 unmodified CTLS; M1–M5 modified CTLS [source: own study; KEYENCE VHX 950F].

Table 1. Type and characteristics of CTLS modifying additives.

Acronym of Sample	Raw Material	Type of Additives	Physical State of Additives	pH of Additives	Additives [%]
M0		-	-	-	0
M1	CTLS	Pectin	powder	4.00	11
M2		Glutaraldehyde	liquid	3.70	5
M3		Glycerol	liquid	5.00	50
M4		EDC *	powder	4.00	4
M5		Tannin	powder	2.80	2

* EDC: 1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide.

2.3. Testing of Physical and Chemical Parameters

The pH values were determined by the potentiometric method using an Elmetron CP505 pH meter. Fiber length was determined as the average length of one hundred randomly selected fibers measured manually. The tensile strength was measured using a Zwick/Roell (Z010) tester (ZwickRoell GmbH&Co. KG, Ulm, Germany). During the research, the bulk density was determined by weighing the samples on a digital balance and measuring their bulk volumes using a 250 cm³ glass measuring cylinder. Bulk density values were calculated from Equation (1):

$$\rho = \frac{mc - m}{vc} \quad (1)$$

where

mc is the mass of the sample together with the measuring cylinder [g],

m is the mass of the measuring cylinder [g], and

vc represents the volume of sample in the measuring cylinder [cm³].

The total content of selected elements (metals) in various tanning shavings samples were determined using plasma optical emission spectrometry with an Agilent ICP-OES 5110 spectrometer (Santa Clara, CA, USA). Each 0.2 g sample was mineralized with HNO₃ (65%, wt, Chempur, Piekary Slaskie, Poland) in teflon vessels using a Magnum II microwave mineralizer (Ertec, Wroclaw, Poland). The mineralization process, conducted in 3 cycles lasting a total of 20 min, reached a temperature of 300 °C and pressure of 45 bar, with the microwave power set at 100%. The content of the elements tested in the samples was read from standard curves prepared from standards of individual metals. All elements were determined by inductively coupled plasma optical emission spectrometry using an ICP-OES 5110 spectrometer (Santa Clara, USA) for multi-element analysis. Commercial argon (from Hen-DuKOl, Łódź, Poland) was used to generate the plasma. The sample is introduced into the plasma as an aerosol. The analytical lines and the optimum mode of operation for the determination of individual elements were selected. The linear fit of the calibration curve was used to determine the concentration of elements in the samples. Determinations were carried out using the external standard method. The calibration of the measurement method was performed prior to the measurement using a series of chemical standards (reference materials) with different levels of the content of the tested component. The contents of the tested metals in the samples were read from the standard curves prepared from the ICP standard solution (metals: Ca, Cr, Fe, K, Na, and P in 5% HNO₃, LGC Standard, Manchester, NH, USA) and from the standard of metals (S in H₂O, CPACChem, Stara Zagora, Bulgaria) by appropriate dilution of the standards with 5% HNO₃ (v/v). The tests for Ca, Cr, Fe, K, Na contents were carried out according to the PN-EN ISO 17072-2:2019 standard [37]. The preparation method and measurement of P and S were performed in the same way as for other metals. Chromium (VI) in leather shavings was measured according to the accredited method described in PN-EN ISO 17075-1:2017-05 (UV/VIS spectrophotometric method) [38].

2.4. FTIR Analysis

The study used Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy (ATR-FTIR) to analyze the structural changes exhibited by both modified and unmodified CTLS. First, several fibers were compressed into a disc and then scanned five times in the wavenumber range of 600–4000 cm^{-1} using a Bruker Invenio S spectrometer with a temperature-controlled diamond-window ATR accessory (MIR ATR Diamond). Each sample was scanned with 32 scans per sample and a resolution of 4 cm^{-1} .

2.5. Phytotoxicity Test

2.5.1. Experimental Design in Soil

The short-term experiment was conducted under laboratory conditions for 1 month using identical 2 dm^3 glass containers to which 1 kg of soil and various modified material (M0–M5) were added, at a ratio of 20:1. The reference sample was soil free from the modified material. All samples were mixed homogeneously with soil and incubated at 17–20 °C for 31 days. OECD soil moisture was maintained at 15% w/w (75% of soil water-holding capacity) and evaporated water was replenished every 7 days. All treatments were carried out in triplicate. During sampling for testing, the soil structure was loosened.

2.5.2. Biological Analysis

The effects of the modified samples on plants were assessed using the commercial toxicity bioassay—Phytotoxkit[®] Solid Samples for germination and early plant growth test (ISO Standard 18763; Ghent, Belgium) [39]. In the present study, six samples were tested: control soil and sample M0–M5. Control soil the control soil was without raw and modified materials. First, for the tested soil, 140 g (90 cm^3) was moistened with distilled water to 100% water capacity and covered with filter paper. The tests were made in three replications for each plant. The monocotyledonous plant *Sorghum saccharatum* (sorghum, series no. SOS170524) and two dicotyledonous plants *Lepidium sativum* (garden cress, series no. LES330524), *Sinapis alba* (mustard, series no. SIA291824) were used as model seeds in these experiments. The appropriately prepared soil and ten seeds of one of the higher plants were placed in the special test plate and then, all test plates were incubated for 72 h at 25 °C in the darkness in the incubator (DANLAB, Bialystok, Poland). Results are expressed as % inhibition or stimulation of root elongation (all data averaged) relative to control soil. The response of the plants to the M0–M5 materials was determined by the degree of inhibition or stimulation of seed germination root growth. All measurements were carried out using an “Image J” analysis program (Image Analysis Software; Image J Version 1.53k, National Institutes of Health, USA). The results for plants (different sensitivity) were averaged. During the process, the pH value changes were monitored by potentiometric method, at the beginning and after 31 days.

2.6. Statistical Analysis

The results were statistically analyzed using the Tukey–Kramer test, adjusted for multiple comparisons ($p \leq 0.05$ or $p \leq 0.1$). STATISTICA version 10.0 (Krakow, Poland) was used for all calculations.

3. Results and Discussion

3.1. Testing of Physical and Chemical Parameters

As part of the study, research was conducted to investigate the physico-chemical properties of modified CTLS. Furthermore, initial tests were conducted to evaluate the potential environmental impact. The modified samples obtained differed slightly in the following parameters: pH, bulk density, fiber size, and tensile strength (Table 2). The low pH of the modified materials was due to the initial low pH of the CTLS (3.89) and the low initial pH of the additives used (Table 1). The pH of all the samples ranged from 3.33 to 4.25. In turn, the value of the bulk density of the tested samples increased and ranged from 0.15 to 0.52 g cm^{-3} . The highest value was found for the sample that was

modified with pectin (0.52 g cm^{-3}). For the unmodified CTLS, the value of bulk density was 0.15 g cm^{-3} , which was in agreement with the literature [40,41]. Interestingly, the modifications carried out also resulted in a reduction in fiber length. Changing this physical property of CTLS significantly facilitated the grinding process. This is especially important because the CTLS milling process is difficult due to the length of the fibers, which can twist in the mill. In addition, the low bulk density makes it difficult to carry out other unit operations such as shredding, milling, and screening [10]. The modifications carried out not only increased the bulk density, but in some cases also slightly improved the tensile strength of the fibers (particularly in M1, M2, M4 samples). Changing the unfavorable surface characteristics of collagen fibers has resulted in improved physical properties. Thus, presenting the possibility of utilizing waste material replete with collagen fibers without protein isolation for producing a fully competitive and economically feasible alternative to man-made fiber products [20–22,25,26].

Table 2. Physical parameter data for tested samples before the grinding process. Values designated with the same letter for bulk density or size of fiber were not significantly different ($p \leq 0.05$) according to the Tukey–Kramer test for multiple comparisons, $n = 3$.

Acronym of Sample	Bulk Density [g cm^{-3}]	pH [-]	Size of Fiber [mm]	Tensile Strength [N/mm^2]
M0	0.15 ± 0.02 a	3.86 ± 0.23 a	70 ± 20 a	0.34 ± 0.03 b
M1	0.52 ± 0.03 d	3.83 ± 0.20 a	40 ± 10 a	0.46 ± 0.02 c
M2	0.17 ± 0.01 a	3.33 ± 0.12 a	50 ± 10 a	0.83 ± 0.04 d
M3	0.39 ± 0.04 c	4.16 ± 0.17 a	50 ± 10 a	0.03 ± 0.01 a
M4	0.15 ± 0.03 a	3.89 ± 0.20 a	40 ± 15 a	0.78 ± 0.08 d
M5	0.20 ± 0.02 b	4.25 ± 0.13 a	60 ± 10 a	0.04 ± 0.01 a

The effect of the modification of CTLS on the elemental content of the final product is shown in Table 3. The results of the elemental contents of the samples tested showed the resulting modified CTLS were free of Cd, Hg, Pb, and Cr(VI). The absence of heavy metals was crucial with regard to the environmental aspect discussed later. It was also confirmed tanning waste contained significant amounts of various, environmentally important elements. Modified and unmodified CTLS appear to be potentially valuable waste material due to their high levels of calcium (ranging from 739.24 to $2111.37 \text{ mg kg}^{-1}$), iron (ranging from 295.12 to $813.69 \text{ mg kg}^{-1}$), magnesium (ranging from 405.08 to $1036.33 \text{ mg kg}^{-1}$), sodium (ranging from 8355.70 to $19,813.52 \text{ mg kg}^{-1}$), phosphorus (ranging from 114.10 to $337.62 \text{ mg kg}^{-1}$), and sulphur (ranging from 3145 to $12,848 \text{ mg kg}^{-1}$). It is worth considering these findings in the context of waste management and resource recovery. The levels of elements in the samples demonstrated statistically significant disparities between the untreated and modified material. The incorporation of pectins likely resulted in a minor escalation of calcium and iron concentrations in the material (refer to Table 3) compared to the unmodified sample. Pectins exist in the configuration of plant cell walls and are composed of chains of oligo- and polysaccharides that are conjoined by calcium [16,25]. It was also clear, the modification of the materials reduced the proportion of crude CTLS, which in turn reduced the amount of chromium III present in the final product, as shown in Table 3. It is important to note the modification process does not result in the oxidation of chromium III to chromium (VI), which is a crucial factor regarding the toxicity during the development of novel materials [6,9].

3.2. FTIR Analysis

In the study, we used an ATR-FTIR spectrometer to characterize the materials obtained (M0–M5). In the recorded spectra of the tested samples, we observed bands characteristic of the collagen structure (Table 4). The bands most shifted towards the near infrared were the amide A and B bands. They come from the stretching vibration of the NH group. The position of the amide A band depended on the strength of the hydrogen interaction of the

NH [42] group and was located between 3291 and 3293 cm^{-1} in the samples studied. In contrast, the amide B band, located in the range 3078 to 3083 cm^{-1} , was characterized by lower intensity than amide A. In the wave number range 1628 to 1633 cm^{-1} there is an amide I band with the stretching vibrations $\nu(\text{C}=\text{O})$. The other much smaller contributions come from oscillations: $\nu(\text{CN})$, $\delta(\text{CCN})$, and $\delta(\text{NH})$ in the plane of bonds. The amide I band analysis plays a key role in determining the nature of a protein's secondary structure [43]. The amide II band, occurring in the region of 1540 to 1550 cm^{-1} range, originates mainly from the $\delta(\text{NH})$ vibration in the binding plane and the $\nu(\text{CN})$ vibration, but to a lesser extent to the $\delta(\text{C}-\text{O})$ and $\nu(\text{CC})$ and $\nu(\text{NC})$ vibrations [44]. In the range of wave numbers from 1235 to 1238 cm^{-1} , there is an amide III band resulting from the coupling of $\delta(\text{NH})$ and $\nu(\text{CN})$ vibrations with a small contribution from $\delta(\text{C}-\text{O})$ and $\nu(\text{C}-\text{C})$ vibrations [43]. This band has a lower affinity for the conformation of the polypeptide chain due to the significant influence of C-C backbone vibrations and vibrations of amino acid residues.

Table 3. Content of selected elements in the sample M0–M5 [mg kg^{-1}]. Values designated with the same letter were not significantly different ($p \leq 0.1$) according to the Tukey–Kramer test for multiple comparisons, $n = 3$.

Metal	Sample					
	M0	M1	M2	M3	M4	M5
Ca	1019.78 a	2111.37 b	971.92 a	739.24 a	871.59 a	1297.21 a
Cr	24,844.92 b	17,650.54 a	19,739.65 a	18,540.24 a	21,054.03 a	24,494.17 b
Cr (VI)	un *	un *	un *	un *	un *	un *
Fe	564.24 b	813.69 c	380.3 a	295.12 a	374.20 a	812.18 c
K	34.45 a	50.61 a	41.14 a	14.38 b	33.11 a	110.10 c
Mg	754.44 b	409.49 a	1036.33 c	405.08 a	863.81 b	808.65 b
Na	10,651.40 a	19,813.52 b	13,889.17 b	1070.88 b	8355.70 a	12,028.4 b
P	333.65 d	199.39 b	282.51 c	114.10 a	180.14 b	337.62 d
S	13,825.62 a	11,309.49 a	15,400.63 a	12,234.42 a	14,956.20 a	14,456.39 a
Cd	un *	un *	un *	un *	un *	un *
Hg	un *	un *	un *	un *	un *	un *
Pb	un *	un *	un *	un *	un *	un *

* un—undetectable $< 3 \text{ mg kg}^{-1}$.

Table 4. Position of the amide bands for samples M0–M5 recorded with the ATR-FTIR.

Amide Band Symbol	M0	M1	M2	M3	M4	M5
A	3291	3292	3293	3293	3293	3286
B	2932	2930	2927	2928	2937	2931
I	1631	1628	1631	1634	1630	1630
II	1540	1544	1542	1551	1540	1541
III	1236	1237	1236	1238	1235	1236

The occurrence of all the significant peaks in samples M1–M5, potentially cross-linked collagen, demonstrated the integrity of the collagen was not disturbed in the presence of chemical compounds used (Figure 2). However, for samples M1, M2, M4, and M5, the recorded changes in the position of the bands in the 1200–1000 cm^{-1} region may indicate possible cross-linking of collagen. Moreover, the relative intensity of the collagen characteristic collagen peak (amide I, amide II, and amide III) was changed, suggesting some sites of the collagen amide group was involved in the interaction between M0 and the added chemical compounds [44]. For example, for sample M1 (after pectin modification), the introduction of sugars into the strings affect, among others, the reactive amino acids lysine, hydroxylysine, and histidine, which is reflected in the spectrum by the NH bending vibration coupled with the CN stretching vibration [45,46]. However, in the case of compound M3 (after modification with glycerol), intense peaks from the glycerol molecule are visible in the recorded FTIR spectrum. Their location in the dactyl region interferes with the correct

interpretation of the recorded spectrum. The developed process allows cross-linking CTLS waste without earlier pre-activation. However, further studies are needed to assess the effect of used additives on the cross-linking of collagen present in CTLS.

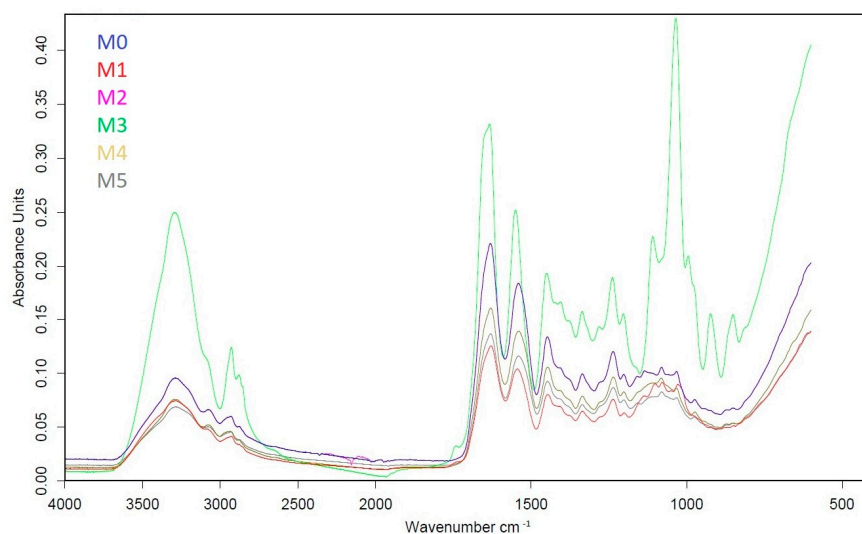


Figure 2. FTIR spectra of samples M0–M5.

3.3. Phytotoxicity Test

The evaluation of new materials on the basis of physico-chemical parameters alone may not be sufficient. Additional tests, such as phytotoxicity tests, are essential to assess their impact on the environment. These tests provide a quick and reliable assessment of the level of toxicity of the raw materials under consideration. Figure 3 depicts the outcomes of a soil phytotest, utilizing sensitive plants to evaluate the effectiveness of modification alternatives of CTLS. Only two variants, control soil and M1, displayed a normal percent stimulation index of root length (refer to Figure 3). The M1 sample exhibited the highest outcome of 105%. The likely cause of this phenomenon was the presence of pectins in the modified material. Pectins are linear combinations of oligosaccharides and polysaccharides. The main components are d-galacturonic acid and saccharides, such as d-galactose, d-xylose, l-fructose, l-rhamnose, and l-arabinose [47]. These compounds are capable of stimulating microorganisms and plants [48,49]. Samples M0 and M2 exhibited the most significant root growth inhibition, with a respective outcome of 74% and 75% (See Figure 3). The presence of chromium III in sample M0 may have resulted in unsuitable conditions for plant growth. The valence state of chromium determines its toxicity to plants: Cr(VI) is highly toxic and mobile, whereas Cr(III) is not toxic at low concentrations [50]. Raw tanning shavings are biologically unstable waste [51]. The degradation of the material may have resulted in the release of chromium, which formed a three-dimensional structure with collagen through cross-linking [52]. In the M2 experiment, glutaraldehyde was used for modification. Glutaraldehyde has antimicrobial and preservative properties and is effective against bacteria, viruses, spores, and fungi in their vegetative forms [53]. However, excessive use of glutaraldehyde for modification could inhibit plant growth. A moderate level of plant toxicity was observed in the other samples, but the effect was slightly less than that observed in the M0 sample, although it was not statistically significant. Seed germination was not considered to assess the effect of the presence of newly modified materials on biological parameters. The number of seeds that did not germinate was similar. It was also found germinating seeds did not guarantee roots would continue to grow. According to the literature, petroleum substances, heavy metals, or other contaminants have little effect on germination. Seed reserves may have more influence on seed germination than other factors [54–56].

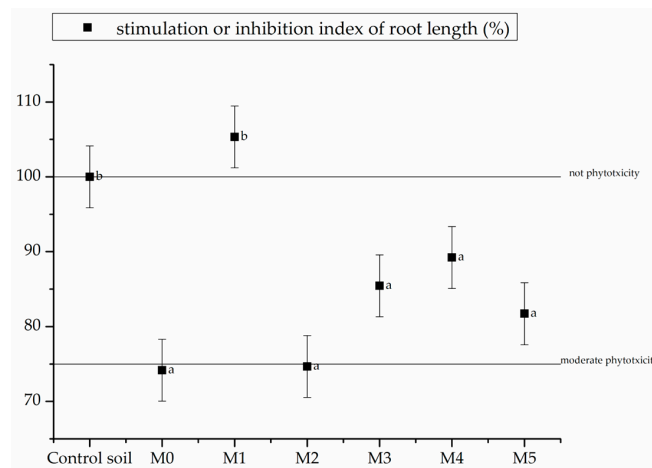


Figure 3. Stimulation/inhibition index of root length values (% of control, mean, and standard deviation) as effects of plant reaction on different materials (M0–M5) in the soil after 4-week experiments. Values designated with the same letter were not significantly different ($p < 0.05$) according to the Tukey–Kramer test for multiple comparisons. Note: The obtained results were averaged, and all treatments were made in triplicate; The y -axis was the calculated seed growth inhibition/stimulation of the plants. Values on the y -axis above 100% indicate stimulation. Values below 100% indicate growth inhibition compared to the control soil.

The pH of the soil in all samples ranged from 5.77 to 6.09 (Figure 4). The highest pH values after 31 days were observed in the control soil without any materials. Conversely, samples M0, M2, and M5 had the lowest pH values after 31 days. The low initial pH of the raw materials used in research may explain the decrease in pH. The raw tanning shavings have a low pH of 3.86 (see Material and Method), 50% glutaraldehyde has a pH not exceeding 3.7. Moreover, a 1% thianine solution has a pH of 2.8, while other additive solutions have a pH in the range of 4–5 (Table 1). However, this decrease in soil pH was not statistically significant (Figure 4). The stability of the modified materials and their slow absorption rate may explain why the soil pH did not change in most samples. This feature is desirable in creating new materials [57].

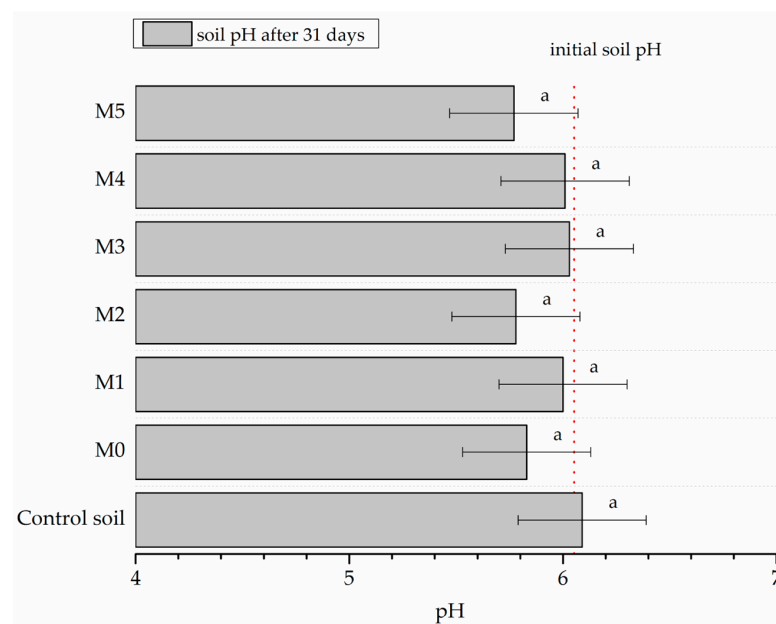


Figure 4. Changes in soil's pH after adding 6 different variants of materials (samples: M0–M5) in a concentration of 5% (w/w) to the environment. Values designated with the same letter were not significantly different ($p < 0.05$) according to the Tukey–Kramer test for multiple comparisons.

4. Conclusions

The present study aimed to innovate the treatment of chrome tanned leather shavings (CTLS) by investigating possibilities for their modifications without prior isolation of protein components. Various substances including glutaraldehyde, glycerol, EDCs, pectin, and tannins were used to evaluate their potential to modify CTLS. The results showed favorable changes in the physical properties of bulk density and tensile strength after modification. Changing these properties may have a positive impact on the possibility of mechanical processing, e.g., grinding. Importantly, elemental analysis confirmed the absence of hazardous elements like Cd, Hg, Pb, and Cr(VI) in the resulting modified CTLS, which potentially indicated no negative impact on the environment. The application of FTIR spectroscopy confirmed the integrity of collagen in the tested samples and simultaneously revealed alterations in their molecular structure confirming modifications. The developed procedure with the application of glutaraldehyde, EDCs, pectin, and tannin allows modifications of CTLS waste without prior preparation. This was confirmed by the changes in the intensity of the characteristic amide bands of collagen observed in the spectrum. Phytotoxicity tests shed light on the varying effects of the modified CTLS on plant growth. The introduction of pectins exhibited the highest root stimulation, presenting a promising avenue for sustainable utilization. Conversely, glutaraldehyde and unmodified CTLS showed significant root inhibition, emphasizing the importance of selecting modification agents judiciously. Despite the modifications, no statistically significant changes in soil pH were observed, indicating the potential for these modified materials to integrate into environmental systems without causing significant disturbances. In summary, this comprehensive study not only demonstrated the feasibility of modifying CTLS to enhance physical properties but also emphasized the need for careful consideration of environmental impacts. The results highlight the potential of modifying CTLS waste, opening the door to different applications in industrial sectors, including road construction, the asphalt industry, and the production of various types of composites.

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