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Alkaline hydrolysis of Zeolite and Chrome Shavings: Investigating the Fate of Their Hydrolysate and Undigested Materials

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1. Introduction

The leather industry generates substantial solid waste, primarily chrome-tanned leather shavings (CTLS) and trimmings, leading to environmental and economic challenges due to rising disposal costs and strict regulations.[1] While CTLS is used in fertilizers, leatherboard, and biopolymer applications, its broader utilization remains limited by environmental concerns, regulatory restrictions, and the risks associated with hexavalent chromium contamination.[2-5] One approach to addressing these challenges is the hydrolysis of CTLS to extract collagen-based hydrolysates for further applications.[6] However, studies have shown that removing more than 95% of the chromium is difficult due to its strong binding affinity to collagen, limiting the potential uses of CTLS hydrolysates as residual chromium raises environmental and health concerns.[2,7] Consequently, disposal has largely relied on landfilling, which not only carries the risk of chromium (III) oxidizing into its more toxic hexavalent (VI) form but also comes at a significant economic cost.[8] In New Zealand, the annual cost of landfilling has risen to approximately $1.5 million, highlighting the urgent need for sustainable waste reduction strategies.

The hydrolysis of tanned leather waste offers a sustainable approach to recovering collagen-derived materials, but its efficiency depends on the tanning agents used.[9] Different tanning methods alter collagen stability, affecting hydrolysis efficiency and the composition of recovered byproducts. For example, wet white tanning, which uses glutaraldehyde and Tetrakis(hydroxymethyl phosphonium sulfate (THPS) to reduce chromium use, still leaves residual agents after hydrolysis.[10,11] These residues can be cytotoxic, environmentally persistent, or affect downstream applications, limiting the usability of the resulting hydrolysates.

Zeolite-based tanning has emerged as a chrome-free alternative in sustainable leather production.[12–15] Zeolites are crystalline, hydrated aluminosilicates with a highly ordered, porous structure that contains water molecules and metal cations.[16] Their low toxicity, structural stability, and environmental benefits make them suitable for various applications, including human and veterinary uses.[17] In the tanning process, functionalized zeolites have been explored for their ability to stabilize collagen fibres while reducing environmental impact.[14,15] This makes zeolite-based tanning a promising alternative to chromium and the conventional wet white methods, addressing key environmental and waste management challenges. While the partial removal of chromium in hydrolysates from CTLS is well-documented, the behaviour of zeolites during the hydrolysis of ZTLS remains largely underexplored. Due to their unique binding characteristics and non-uniform particle size,

zeolites interact with collagen differently from chromium during tanning and hydrolysis.[13] Understanding their behaviour in this process is crucial for assessing their impact on material separation and hydrolysate purity.

This study investigates the hydrolysis of ZTLS and CTLS using a modified alkaline method to assess their interaction with collagen and the efficiency of separation. The primary objectives were:

- To optimize an alkaline hydrolysis protocol for ZTLS.
- To comprehensively characterize and compare the hydrolysis products of ZTLS and CTLS.
- To determine the fate and distribution of the tanning agents (zeolite vs. chromium).
- To evaluate the potential for valorisation of all output streams (soluble hydrolysate and insoluble residual solids).

By analysing the breakdown process and characterizing the resulting fractions, this work evaluates the fate of tanning agents and their potential for reuse. Understanding these interactions will help refine hydrolysis conditions and assess the composition of hydrolysates, particularly their organic and inorganic content, thereby informing the development of more sustainable and circular waste management strategies for the leather industry.

2. Experimental Methods

2.1. Materials

Zeolite-tanned leather shavings and chrome-tanned leather shavings were sourced from a local tannery in New Zealand. All other reagents were from commercial sources and used as received.

2.2. Alkaline Hydrolysis Protocol Optimization

Hydrolysis was carried out using a modified alkaline method based on Mu et al. (2003).[18] Preliminary experiments evaluated different sodium hydroxide (NaOH) concentrations (0.05, 0.1, 0.2, and 0.5 M) and reaction times (4, 6, and 24 hours) at 90°C to determine optimal degradation conditions for ZTLS (Tables 1 & 2).

Table 1: Effect of Different Alkaline Conditions on ZTLS Degradation. Mass balance was used to calculate the percentage of degradation (%). Data are presented as mean ± standard deviation. Statistically significant effects were compared using one-way ANOVA and Tukey's post hoc test. Different letters indicate significant differences between means. ($p < 0.05$; $n = 3$).

NaOH Concentration	Degradation (% w/w)
1. 90°C + 0.05 M for 4 hours	37.90 ± 0.010[a]
2. 90°C + 0.1 M for 4 hours	74.67 ± 0.006[b]
3. 90°C + 0.2 M for 4 hours	76.70 ± 0.012[bc]
4. 90°C + 0.5 M for 4 hours	77.98 ± 0.010[c]

Table 2: Impact of Hydrolysis Duration on ZTLS Degradation. Mass balance was used to calculate the percentage of degradation (%). Data are presented as mean ± standard deviation. Statistically significant effects were compared using one-way ANOVA and Tukey's post hoc test. Different letters indicate significant differences between means. ($p < 0.05$; n = 3).

Condition (90°C + 0.1 M NaOH)	Degradation (% w/DW)
1. **4 hours**	74.67 ± 0.006[a]
2. **6 hours**	76.10 ± 0.010[b]
3. **24 hours**	77.87 ± 0.017[c]

Based on these trials, the selected standard condition for comparative analysis was 0.1 M NaOH, 90°C, 24 hours, with a solid-to-liquid ratio of 1:10 (w/v). Under these conditions, 300 g of ZTLS or CTLS were hydrolysed in an open vessel in a hot-water bath under constant stirring. The initial pH of the 0.1 M NaOH solution recorded was approximately 12.5. Following hydrolysis, the final pH of the ZTLS mixture was approximately 11.6. CTLS was hydrolysed under identical conditions (Table 3).

Table 3: Impact of Hydrolysis Duration on CTLS Degradation. Mass balancing was used to calculate the degradation %. Mean ± standard deviation (n=5).

Condition (90°C + 0.1 M NaOH)	Degradation (% w/DW)
24 hours	83.27 ± 0.013

After hydrolysis, mixtures were centrifuged at 10,000 rpm for 30 minutes to separate the soluble hydrolysate from the insoluble residue. The hydrolysate fraction was frozen, freeze-dried, and milled into powder form. The unhydrolyzed residue was oven-dried at 100°C for 24 hours before being ground into powder for further analysis.

2.3. Preparation of Bovine Hydrolysate (Reference Control)

Delimed bovine hides were diced into approximately 1 cm^3 pieces (reported by volume due to non-uniform hide thickness). A 300 g sample was subjected to enzymatic hydrolysis using trypsin and bromelain (each at 1% w/w relative to wet hide weight) at a solid-to-liquid ratio of 1:5 (w/v).

Hydrolysis was performed in an open vessel in a hot water bath at 55°C for 48 hours under constant stirring. The pH was maintained between 7 and 8 using 0.1 M sodium phosphate buffer. This enzymatic protocol was developed and optimized in-house by LASRA.

Following hydrolysis, the mixture was centrifuged to separate soluble and insoluble fractions. The hydrolysate was frozen, freeze-dried, and milled into a fine powder, which served as a reference material.

2.4. Mass and Protein Balance

Moisture content was determined using a modified version of ISO 4684:2005. Inorganic (ash) content was measured according to ASTM D6716-08.

Total protein content was quantified via nitrogen determination using a modified Kjeldahl method based on ISO 5397:1984. Nitrogen content was converted to protein using an

appropriate conversion factor. These measurements enabled calculation of degradation efficiency through mass balancing of hydrolysed and residual fractions.

2.5. Elemental Analysis

Elemental distribution of aluminium (Al), silicon (Si), and sodium (Na) was quantified using inductively coupled plasma techniques (ICP-OES and ICP-MS).

Total chromium content [Cr(III) and Cr(VI)) was determined in accordance with ISO 5398-1:2018 and ISO 17075-2:2017, respectively.

For ZTLS samples, aluminium and silicon concentrations were measured using modified APHA 3120 B procedures, while sodium content was determined using a modified AOAC 984.27 method. These analyses were used to assess tanning agent distribution between soluble hydrolysates and insoluble residues.

2.6. Fourier Transform Infrared (FTIR) Spectroscopy

Hydrolyzed and unhydrolyzed samples were analysed using a Thermo Scientific Nicolet iS5 FTIR spectrometer equipped with a ZnSe ATR attachment. Spectra were collected over a wavenumber range of 600–4000 cm^{-1}, averaged over 32 scans at 20 ± 1°C.

Amide I and II bands were evaluated to assess collagen integrity, while characteristic functional group vibrations were used to identify tanning agent interactions and confirm the presence of chromium- or zeolite-related species.

2.7. Molecular Weight Distribution (SEC)

Peptide molecular weight distribution was determined using size-exclusion chromatography (SEC) on a BioSep SEC-s2000 column (300 mm × 7.8 mm). The system was calibrated using gel filtration standards ranging from 1,350 to 670,000 g/mol.

Samples (5 mg/mL in 0.1 M phosphate buffer, pH 7.6) were filtered (0.45 μm) prior to injection (10 μL). Isocratic elution was carried out with “mobile phase” at 0.5 mL/min over 40 minutes, and peptide profiles were monitored at 210 nm using UV detection.

2.8. Amino Acid Composition

Amino acid analysis was performed using HPLC following pre-column derivatization with 6-aminoquinolyl-N-hydroxysuccinimidyl carbamate (AQC). Briefly, 10 μL of standard amino acid solution or sample extract was mixed with 70 μL of 0.2 M borate buffer (pH 9.0) and 20 μL of 15 nmol/μL AQC dissolved in acetonitrile. The mixture was vortexed and incubated at 55 °C for 10 min.

Following derivatization, samples were diluted with 400 μL of water and centrifuged at 14,100 × g for 15 min. The supernatant was injected into a Gemini NX-C18 reversed-phase column (150 mm × 3 mm, 3 μm, 110 Å; Phenomenex, USA) fitted with a Phenomenex SecurityGuard™ C18 guard cartridge.

Chromatographic separation was performed at 45 °C using gradient elution. Mobile phase A consisted of 0.4% (v/v) formic acid in water, while mobile phase B consisted of 60% (v/v) acetonitrile in water. The gradient was programmed from 100% to 45% mobile phase A over 16 min. Eluted peaks were monitored at 260 nm.

2.9. Statistical Analysis

All experiments were performed in triplicates, and data are presented as mean ± standard deviation (SD). Statistical significance between sample groups was assessed using one-way analysis of variance (ANOVA) followed by Tukey's post hoc test for multiple comparisons. For amino acid quantification, SEC molecular weight distributions, ICP-MS and ICP-OES elemental analysis, Pearson's correlation was employed to evaluate relationships between different measured parameters. Statistical analysis was performed using JASP version 0.19.3.0 (JASP Team, University of Amsterdam, The Netherlands). A p-value of <0.05 was considered statistically significant.

3. Results and Discussion

3.1. Hydrolysis Efficiency and Mass Balance

Hydrolysis of ZTLS and CTLS was conducted under the optimized standard conditions (0.1 M NaOH, 90°C, 24 h, solid-to-liquid ratio 1:10 w/v). Preliminary optimization (Tables 1 & 2) confirmed that increasing NaOH concentration and reaction time enhanced degradation efficiency.[18] ZTLS degradation reached approximately 78%, whereas CTLS achieved ~83%. Protein recovery into the hydrolysate was 71.7% for ZTLS and 68.2% for CTLS.

Table 4: Total Protein Quantification for ZTLS.

[1] Moisture content calculated using Modified ISO 4684:2005.
[2] Inorganic content determined using ASTM D6716-08 (2020), dry weight basis.
[3] The nitrogen content was measured using a modified Kjeldahl method and converted to protein content using an appropriate conversion factor.
Mean ± standard deviation (n=3). The percentages of protein recovery are presented in brackets relative to the total protein content measured in ZTLS.

Sample	Mass Balance (% w/w)		Protein Content (% w/DW)	
ZTLS	Moisture Content[1]	52.94 ± 0.19	/	
	Inorganic Content[2]	16.88 ± 0.04	/	
	Protein Content[3]	29.93 ± 0.21 (100.0%)	Hydrolyzed ZTLS Fraction	21.45 ± 0.44 (71.7 ± 1.9%)
			Unhydrolyzed ZTLS Fraction	0.63 ± 0.01 (2.1 ± 0.0%)

Table 5: Total Protein Quantification for CTLS.

[1] Moisture content calculated using Modified ISO 4684:2005.
[2] Inorganic content determined using ASTM D6716-08 (2020), dry weight basis.
[3] The nitrogen content was measured using a modified Kjeldahl method and converted to protein content using an appropriate conversion factor.
Mean ± standard deviation (n=3). The percentages of protein recovery are presented in brackets relative to the total protein content measured in CTLS.

Sample	Mass Balance (% w/w)	Protein Content (% w/DW)

CTLS	Moisture Content[1]	56.51 ± 0.15	/	
	Inorganic Content[2]	5.10 ± 0.03	/	
	Protein Content[3]	40.83 ± 1.27 (100.0%)	Hydrolyzed CTLS Fraction	27.83 ± 1.47 (68.2 ± 5.1%)
			Unhydrolyzed CTLS Fraction	1.99 ± 0.55 (4.9 ± 1.3%)

The hydrolyzed ZTLS produced a reddish-brown, slightly cloudy solution, likely due to residual syntans, which upon freeze-drying yielded a fine, chalky powder. CTLS hydrolysis initially produced a moss-green solution, which clarified upon filtration and dried to a pale-yellow powder, suggesting chromium precipitation or oxidation. In contrast, the bovine hide control yielded an off-white hydrolysate, reflecting the absence of tanning agents.

Alkaline hydrolysis effectively liberated protein from ZTLS, demonstrating its potential as a valorisable feedstock. The slightly higher protein recovery from ZTLS compared to CTLS may reflect differences in collagen cross-linking and the binding strength of the respective tanning agents.

3.2. Fate of the Tanning Agent

Chromium in CTLS

FTIR analysis of the CTLS hydrolysate revealed a persistent peak at ~1590 cm^{-1}, indicative of chromium-carboxylate interactions with collagen residues (Figure 1A, stacked with 1B).[19-21] Quantitative analysis confirmed that 37.3% of chromium remained in the solid residue, while a small but notable 0.3% persisted in the hydrolysate (Table 6). These observations indicate that chromium-collagen binding largely persists post-hydrolysis, limiting downstream applications where low metal content is required. Hydrolysate color changes and precipitation behavior also support partial chromium separation during alkaline treatment.

Zeolite in ZTLS

FTIR spectra of the unhydrolyzed ZTLS fraction displayed characteristic Si-O-Si and Si-O-Al vibrations, confirming the integrity and stability of the zeolite framework (Figure 1B, stacked with 1A). Elemental analysis revealed that approximately 80% of aluminium remained in the solid residue after hydrolysis, with silicon largely retained as well (Table 7). This retention suggests that zeolite separates cleanly from the soluble collagen peptides, supporting a valorisation pathway with minimal contamination of the hydrolysate. Partial

dissolution of labile Si-O-Al bonds may release some aluminate ions, but the bulk of the zeolite remains intact, consistent with the FTIR and ICP data.[12,15,22,23]

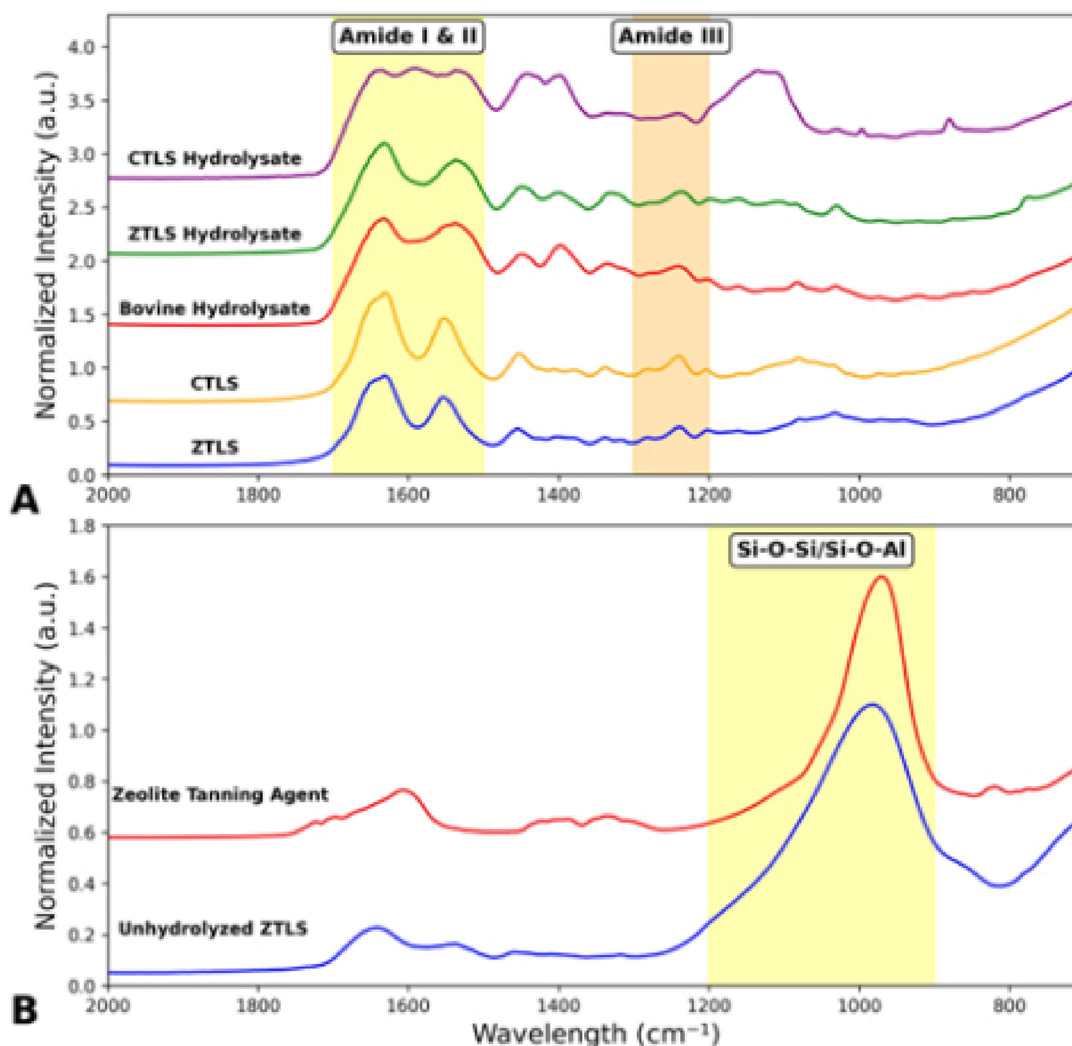

Fig. 1 (A) FTIR spectra comparison of hydrolysates (CTLS, ZTLS and bovine hydrolysate) and the respective intact materials (CTLS and ZTLS); **(B)** zeolite tanning agent vs. unhydrolyzed zeolite fraction from alkaline hydrolysis of ZTLS

Table 6: Quantification of Total Chromium (III) and (VI) in CTLS and Its Derived Hydrolyzed and Unhydrolyzed Fractions. Mean ± standard deviation. One-way ANOVA and post-hoc Tukey's tests revealed significant differences between Unhydrolyzed Fraction (a) and CTLS (b), Unhydrolyzed Fraction (a) and Hydrolyzed Fraction (c), CTLS (b) and Hydrolyzed Fraction (c) ($p < 0.05$; n = 3). Different superscript letters indicate statistically significant differences between groups. n.d. = not detected (below the detection limit of the method).

Sample	Cr (III) (%w/w, on dry weight)	Cr (VI) (%w/w, on dry weight)
CTLS	5.3 ± 0.1^{b}	n.d.
Hydrolyzed Fraction	0.3 ± 0.2^{c}	0.042 ± 0.007
Unhydrolyzed Fraction	37.3 ± 1.3^{a}	-

Table 7. Elemental Analysis of ZTLS and its derived Hydrolyzed and Unhydrolyzed Fractions (*Data normalized to 1 kg of total ZTLS being digested. Mass balancing and moisture content were used to calculate the amounts of hydrolysate and unhydrolyzed matter.) (See Tables 4-5)

Sample Reference	Al (III) (mg/kg)	Si (IV) (mg/kg)	Na (I) (mg/kg)
ZTLS Hydrolysate	3,339	<10	51,983
ZTLS Unhydrolyzed	10,400	<10	5,444
ZTLS	14,600	<10	1,145

3.3. Peptide Profile and Molecular Interactions

Size-exclusion chromatography (SEC) revealed distinct molecular weight distributions across the hydrolysates (Table 8).

Table 8. Comparison of Hydrolysates (CTLS, ZTLS, and Bovine) via Size Exclusion Chromatography, Showing Molecular Weight Distribution and Peak Width at Half Height (PWHH, min)

Sample	Molecular Weight Distribution (Da)	Peak Width at Half Height (min)
ZTLS Hydrolysate	500 – 296,000	6.271
CTLS Hydrolysate	200 – 38,600	3.435
Bovine Hydrolysate	100 – 35,900	2.972

The bovine hydrolysate, produced enzymatically without any tanning agents, exhibited the narrowest molecular weight distribution (0.1–36 kDa). This reflects the mild and selective nature of enzymatic hydrolysis, which breaks down collagen into smaller peptides consistently, without interference from metals or inorganic residues that could promote peptide aggregation.[24-25]

CTLS hydrolysates showed a slightly broader distribution (0.2–38 kDa), which can be attributed to residual chromium interactions. FTIR and elemental analysis confirm that a small fraction of chromium remains bound to carboxylate groups on collagen peptides, which may hinder peptide aggregation or modify solubility, resulting in a population of slightly larger soluble peptides compared to the bovine control.[19-21]

The ZTLS hydrolysate exhibited the broadest molecular weight distribution, ranging from 1 kDa to 300 kDa. This broad profile arises from a combination of factors: partial preservation of large collagen fragments, interactions with aluminosilicate species released from the zeolite matrix, and possible agglomeration of collagen–aluminosilicate complexes. The high alkaline conditions can alter the charge state of peptides, reducing electrostatic repulsion and promoting formation of larger complexes.[12,22,23] FTIR analysis also supports that ZTLS

underwent comparatively less collagen degradation than CTLS, which further explains why larger fragments are retained in the hydrolysate. Collectively, these observations indicate that both the chemical nature of the tanning agent and the hydrolysis environment strongly influence peptide distribution and molecular interactions.

3.4. Amino Acid Composition

Amino acid analysis reflected the influence of tanning agents on hydrolysate composition.

Table 9: Comparison of Amino Acid Composition (mg/g) in CTLS, ZTLS, Bovine Hydrolysates (BH), and Type I Collagen standard (Note: Values are expressed as milligrams of amino acid per gram (dry weight) of each respective hydrolysate).

Amino Acid	ZTLS	CTLS	BH	Type I Std
HyPro	43.7 ± 2.2	64.0 ± 3.0	95.5 ± 3.4	102.0 ± 9.0
His	27.5 ± 1.2	24.6 ± 0.8	7.8 ± 1.0	47.1 ± 3.8
Asp	34.7 ± 2.3	44.9 ± 1.7	56.8 ± 1.7	55.6 ± 4.7
Arg	4.1 ± 1.0	16.8 ± 3.4	63.3 ± 2.9	83.2 ± 8.5
Ser	7.5 ± 0.5	11.0 ± 0.6	28.6 ± 0.8	32.5 ± 2.0
Glu	64.4 ± 3.6	83.4 ± 2.2	103.1 ± 3.1	106.6 ± 8.7
Gly	125.9 ± 6.8	168.7 ± 3.7	204.6 ± 2.3	230.8 ± 11.4
Thr	1.7 ± 0.1	3.2 ± 0.2	17.0 ± 0.5	18.0 ± 1.6
Ala	50.6 ± 2.5	66.2 ± 1.7	81.2 ± 1.9	86.8 ± 5.7
Pro	75.7 ± 3.9	101.6 ± 2.0	115.3 ± 0.8	141.1 ± 8.0
Cys	2.3 ± 0.4	3.0 ± 0.6	0.0 ± 0.0	0.0 ± 0.0
Tyr	4.0 ± 0.3	5.6 ± 0.1	6.6 ± 0.3	4.7 ± 0.4
Val	12.5 ± 0.7	16.3 ± 0.3	20.6 ± 0.3	21.7 ± 1.0
Meth	3.3 ± 0.1	5.0 ± 0.4	6.3 ± 0.4	6.3 ± 1.4
Lys	18.3 ± 1.1	22.4 ± 0.8	35.6 ± 2.2	33.9 ± 3.0
Ile	7.6 ± 0.8	10.8 ± 0.5	13.7 ± 0.1	14.9 ± 0.7
Leu	16.9 ± 1.0	22.0 ± 0.5	26.1 ± 0.3	28.7 ± 1.3
Phe	11.2 ± 0.7	15.1 ± 0.4	15.7 ± 0.1	19.4 ± 1.5
Total	**511.9 ± 24.8^{d}**	**684.5 ± 15.1^{c}**	**897.8 ± 17.2^{b}**	**1033.2 ± 50.8^{a}**

The bovine hydrolysate (BH) exhibited the highest total amino acid content at 897.8 mg/g, with high recoveries of glycine (204.6 mg/g), hydroxyproline (95.5 mg/g), and proline (115.3 mg/g), consistent with mild enzymatic hydrolysis and preservation of collagen structure.[26] Alkaline-sensitive residues such as serine (28.6 mg/g) and threonine (Thr) showed moderate reductions relative to a Type I collagen standard, reflecting partial degradation even under enzymatic conditions.

CTLS hydrolysate had a lower total amino acid content of 684.5 mg/g, reflecting both the effects of alkaline hydrolysis and interactions with residual chromium. Structural residues, glycine (168.7 mg/g) and hydroxyproline (64.0 mg/g), decreased by 17.6% and 33.0%, respectively, relative to bovine hydrolysate. Acidic and basic residues known to interact with chromium, including glutamic acid (83.4 mg/g), aspartic acid (44.9 mg/g), and lysine (22.4

mg/g), were reduced by 19.1%, 21.0%, and 37.1%, respectively. This supports the formation of stable chromium–peptide complexes via coordination with carboxylate (-COO^-) groups and amino (-NH_2) groups, which can limit solubility.[19-21] FTIR analysis corroborated the presence of chromium–carboxylate interactions, confirming partial retention of chromium in hydrolysates. Alkaline-sensitive residues such as serine (11.0 mg/g) and threonine were further degraded, whereas proline (101.6 mg/g) remained largely intact.

The ZTLS hydrolysate exhibited the lowest total amino acid content at 511.9 mg/g, reflecting both lower initial protein content (29.9%) and the higher inorganic fraction (16.9%) in the substrate. Aluminosilicate–collagen interactions, including coordination bonds and hydrogen bonding with peptide groups, likely restricted the release of glycine (125.9 mg/g), proline (75.7 mg/g), and other residues.[12] Acidic residues such as aspartic acid (34.7 mg/g) and glutamic acid (64.4 mg/g) were strongly retained through primary valency interactions with the polyaluminosilicate matrix, while basic residues, including arginine (4.1 mg/g) and lysine (18.3 mg/g), were similarly reduced. Octahedral coordination of aluminium and silicon with carboxyl and peptide groups may have further stabilized the complex, resulting in stronger retention of collagen fragments.[12] These observations highlight the significant influence of tanning agent chemistry on amino acid recovery during hydrolysis.

4. Conclusion

This study highlights the comparative behaviour of ZTLS and CTLS under modified alkaline hydrolysis. ZTLS hydrolysis enabled partial separation of organic and inorganic components, with a significant portion of the zeolite remaining in the unhydrolyzed fraction, supporting its potential for recovery and reuse. In contrast, CTLS hydrolysates retained measurable chromium, limiting downstream applications and emphasizing the challenges of complete metal removal.

FTIR and SEC analyses revealed clear differences between hydrolysates: ZTLS hydrolysates exhibited broader molecular weight distributions, reflecting interactions between collagen fragments and zeolite species. Elemental analysis confirmed partial dissolution of zeolites under alkaline conditions, influencing peptide solubility and contributing to lower total amino acid yields in ZTLS compared to CTLS and bovine hydrolysates. The lower initial protein content of ZTLS also contributed to reduced amino acid recovery, with threonine, serine, and arginine disproportionately affected, indicating selective interactions with the zeolite matrix.

Despite higher protein recovery percentages into ZTLS hydrolysates (Tables 4–5), molecular weight distributions remained broad and shifted toward higher masses, consistent with persistent zeolite-collagen complexes. Overall, these findings demonstrate that zeolite-based tanning may offer a more sustainable alternative to chromium tanning, although complete separation of inorganic components from hydrolysates requires further optimization. Future work should aim to improve zeolite recovery and evaluate the environmental benefits of this approach relative to conventional chromium-based processes.

5. Future work

- Further optimizing alkaline hydrolysis conditions to enhance separation of zeolite from collagen hydrolysates.
- Developing methods for efficient recovery and reuse of zeolite from unhydrolyzed residues.
- Quantitatively assessing the environmental benefits of zeolite-tanned leather processing compared to chromium-based waste streams.
- Investigating the functional properties and potential applications of ZTLS-derived collagen hydrolysates.
- Exploring the influence of hydrolysis parameters on amino acid recovery and peptide molecular weight distribution.

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References

(1) Buljan, J.; Kral, I. The Framework for Sustainable Leather Manufacture; Vienna, 2019.

(2) Zhao, L.; Mu, S.; Wang, W.; Gu, H. Toxicity Evaluation of Collagen Hydrolysates from Chrome Shavings and Their Potential Use in the Preparation of Amino Acid Fertilizer for Crop Growth. Journal of Leather Science and Engineering 2022, 4 (1). https://doi.org/10.1186/s42825-021-00072-1.

(3) Okamura, H.; Shirai, K. Basic Studies on Manufacture of Leatherboard from Chrome Collagen Fiber. Journal of the American Leather Chemists Association 1972, 67 (4), 148–162.

(4) Pati, A.; Chaudhary, R.; Subramani, S. A Review on Management of Chrome-Tanned Leather Shavings: A Holistic Paradigm to Combat the Environmental Issues. Environmental Science and Pollution Research 2014, 21 (19), 11266–11282. https://doi.org/10.1007/s11356-014-3055-9.

(5) Alvarez, C. C.; Bravo Gómez, M. E.; Hernández Zavala, A. Hexavalent Chromium: Regulation and Health Effects. Journal of Trace Elements in Medicine and Biology 2021, 65, 126729. https://doi.org/https://doi.org/10.1016/j.jtemb.2021.126729.

(6) Cabeza, L. F.; Taylor, M. M.; Dimaio, G. L.; Brown, E. M.; Marmer, W. N.; Carrió, R.; Celma, P. J.; Cot, J. Processing of Leather Waste: Pilot Scale Studies on Chrome Shavings. Isolation of Potentially Valuable Protein Products and Chromium. Waste Management 1998, 18 (3), 211–218. https://doi.org/10.1016/S0956-053X(98)00032-4.

(7) Tian, Z.; Wang, Y.; Wang, H.; Zhang, K. Regeneration of Native Collagen from Hazardous Waste: Chrome-Tanned Leather Shavings by Acid Method. Environmental Science and Pollution Research 2020, 27 (25), 31300–31310. https://doi.org/10.1007/s11356-020-09183-4.

(8) Liang, J.; Huang, X.; Yan, J.; Li, Y.; Zhao, Z.; Liu, Y.; Ye, J.; Wei, Y. A Review of the Formation of Cr(VI) via Cr(III) Oxidation in Soils and Groundwater. Science of The Total Environment 2021, 774, 145762. https://doi.org/10.1016/J.SCITOTENV.2021.145762.

(9) Scopel, B. S.; Baldasso, C.; Dettmer, A.; Santana, R. M. C. Hydrolysis of Chromium Tanned Leather Waste: Turning Waste into Valuable Materials - A Review. Journal of the American Leather Chemists Association 2018, 113.

(10) Speer, D. P.; Chvapil, M.; Eskelson, C. D.; Ulreich, J. Biological Effects of Residual Glutaraldehyde in Glutaraldehyde-Tanned Collagen Biomaterials. J Biomed Mater Res 1980, 14 (6), 753–764. https://doi.org/https://doi.org/10.1002/jbm.820140607.

(11) Moiseev, D. V; and James, B. R. Tetrakis(Hydroxymethyl)Phosphonium Salts: Their Properties, Hazards and Toxicities. Phosphorus Sulfur Silicon Relat Elem 2020, 195 (4), 263–279. https://doi.org/10.1080/10426507.2019.1686379.

(12) Zauns, R.; Kuhm, P. An Alternative Approach to Traditional Chrome Tanning. Journal of the American Leather Chemists Association 1995, 90 (6), 177–200.

(13) Costantini, N.; Ciardelli, F.; Covington, A. D. Studies on the Tanning Reactions of Zeolite. Journal of the American Leather Chemists Association 2000, 95 (4), 125–137.

(14) Bacardit, A.; Van Der Burgh, S.; Armengol, J.; Ollé, L. Evaluation of a New Environment Friendly Tanning Process. J Clean Prod 2014, 65, 568–573. https://doi.org/10.1016/J.JCLEPRO.2013.09.052.

(15) Wise, W. R.; Davis, S. J.; Hendriksen, W. E.; von Behr, D. J. A.; Prabakar, S.; Zhang, Y. Zeolites as Sustainable Alternatives to Traditional Tanning Chemistries. Green Chemistry 2023, 25 (4260). https://doi.org/10.1039/d3gc00381g.

(16) Shriver, D. F.; Atkins, P. W. Inorganic Chemistry, 3rd ed.; Oxford University Press BPS, 1999.

(17) Petushkov, A.; Ndiege, N.; Salem, A. K.; Larsen, S. C. Toxicity of Silica Nanomaterials: Zeolites, Mesoporous Silica, and Amorphous Silica Nanoparticles. In Advances in Molecular Toxicology; Elsevier, 2010; Vol. 4, pp 223–266. https://doi.org/10.1016/S1872-0854(10)04007-5.

(18) Mu, C.; Lin, W.; Zhang, M.; Zhu, Q. Towards Zero Discharge of Chromium-Containing Leather Waste through Improved Alkali Hydrolysis. Waste Management 2003, 23 (9), 835–843. https://doi.org/10.1016/S0956-053X(03)00040-0.

(19) Arakawa, H.; Kandadi, M. R.; Panzhinskiy, E.; Belmore, K.; Deng, G.; Love, E.; Robertson, P. M.; Commodore, J. J.; Cassady, C. J.; Nair, S.; Vincent, J. B. Spectroscopic and Biological Activity Studies of the Chromium-Binding Peptide EEEEGDD. JBIC Journal of Biological Inorganic Chemistry 2016, 21 (3), 369–381. https://doi.org/10.1007/s00775-016-1347-x.

(20) Socrates, G. Infrared and Raman Characteristic Group Frequencies, 3rd ed.; Long, D. A., Ed.; John Wiley and Sons Ltd: Chichester, 2001.

(21) Menderes Özlem. Fundamental Aspects of the Chrome Tanning Reaction, University of Leicester, 2002.

(22) Jin, M.; Liu, M.; Nachtigall, P.; Grajciar, L.; Heard, C. J. Correction to Mechanism of Zeolite Hydrolysis under Basic Conditions. Chemistry of Materials 2022, 34 (4), 1986. https://doi.org/10.1021/acs.chemmater.2c00290.

(23) Li, J.; Gao, M.; Yan, W.; Yu, J. Regulation of the Si/Al Ratios and Al Distributions of Zeolites and Their Impact on Properties. Chem Sci 2023, 14 (8), 1935–1959. https://doi.org/10.1039/d2sc06010h.

(24) Denis, A.; Brambati, N.; Dessauvages, B.; Guedj, S.; Ridoux, C.; Meffre, N.; Autier, C. Molecular Weight Determination of Hydrolyzed Collagens. Food Hydrocoll 2008, 22 (6), 989–994. https://doi.org/10.1016/J.FOODHYD.2007.05.016.

(25) Tawalbeh, D.; Muhammad Izzuddin, K.; Noraizah, M. S.; and Sarbon, N. M. Techno-Functional and Bioactivity Properties of Collagen Hydrolysate and Peptide: A Review. Food Reviews International 2025, 1–29. https://doi.org/10.1080/87559129.2025.2450052.

(26) Bowes, J. H.; Elliott, R. G.; Moss, J. A. The Composition of Collagen and Acid-Soluble Collagen of Bovine Skin. Biochemical Journal 1955, 61 (1), 143–150. https://doi.org/10.1042/bj0610143.

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Alkaline hydrolysis of zeolite and chrome shavings: Investigating the fate of their hydrolysate and undigested materials

As the supervising scientist for this project, I would like to express my sincere appreciation to the IULTCS Research Commission for supporting Mr Zoheb Akhter's work through the Young Leather Scientist Grant programme. This funding enabled the systematic investigation of the hydrolytic behaviour of zeolite-tanned leather shavings, an area of growing importance as our industry transitions toward sustainable leather production.

The study provided the opportunity to examine how zeolite-based tanning agents interact with collagen during alkaline hydrolysis, and how these interactions differ from those observed in chrome-tanned substrates. Through a combination of analytical methods (e.g., FTIR spectroscopy, HPLC-SEC profiling, elemental analysis, etc.), the project generated new insights into the fate of aluminosilicate tanning components, the stability of aluminosilicate structures under alkaline conditions, and the resulting peptide distributions in hydrolysates. These findings contribute to a broader understanding of waste valorisation pathways and the potential for recovering both inorganic and organic fractions from alternative tanning systems.

The support from IULTCS not only enabled access to analytical instrumentation and materials but also provided an important platform for advancing sustainable leather science. This project has strengthened our capability to evaluate emerging tanning technologies and their downstream environmental implications. I am confident that the outcomes will inform future research into circular processing strategies and contribute meaningfully to the global leather research community.

I want to take the opportunity to thank the IULTCS selection committee once again for recognising the value of this work and for their continued commitment to fostering scientific innovation in the leather sector.

Yours sincerely,

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