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SUPERCRITICAL FLUID EXTRACTED CHESTNUT TANNIN AS NATURAL BIOCIDE FOR SOAKING BEAMHOUSE PROCESS OF LEATHER PRODUCTION

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SUPERCRITICAL FLUID EXTRACTED CHESTNUT TANNIN AS NATURAL BIOCIDE FOR SOAKING BEAMHOUSE PROCESS OF LEATHER PRODUCTION

ABSTRACT. Microorganism activity during soaking beamhouse process of leather production is highly dangerous and should be kept under control. Some chemical components are used for this purpose to protect the hides/skins against microbial activities. Because of the banned and restricted substances due to the environmental and health risks in the recent times, natural active ingredients are considered for industrial production as sustainability. Moreover, supercritical fluid extraction as an environmentally friendly technology is applied to isolate biologically active extracts and supercritical extracted natural compounds are reported as potential antioxidant and antibacterial agent for several applications. In this study, supercritical fluid extracted chestnut tannin was used in soaking process of leather production and was determined as potential natural biocide with very good effect against microorganism activity in the process floats.

KEY WORDS: leather, soaking, chestnut, biocide, bactericide, sustainability

UTILIZAREA TANINULUI DIN CASTAN EXTRAS ÎN FLUID SUPERCRITIC CA BIOCID NATURAL ÎN PROCESUL DE ÎNMUIERE A PIELII

REZUMAT. Activitatea microorganismelor în timpul procesului de înmuiere a pielii este foarte periculoasă și trebuie ținută sub control. Unele componente chimice sunt folosite în acest scop pentru a proteja pieile împotriva activității microbiene. Datorită substanțelor interzise și restricționate care prezintă riscuri pentru mediu și sănătate în ultima vreme, ingredientele active naturale sunt luate în considerare în producția industrială pentru sustenabilitate. Mai mult, extracția în fluide supercritice, fiind o tehnologie prietenoasă cu mediul, este aplicată pentru a izola extractele active din punct de vedere biologic, iar compușii naturali extrași în fluid supercritic sunt raportați ca potențiali agenți antioxidanți și antibacterieni pentru mai multe aplicații. În acest studiu, taninul din castan extras în fluid supercritic a fost utilizat în procesul de înmuiere a pielii și a fost considerat potențial biocid natural cu efect foarte bun împotriva activității microorganismelor în procesele umede.

CUVINTE CHEIE: piele, înmuiere, castan, biocid, bactericid, sustenabilitate

UTILISATION DU TANIN DE CHÂTAIGNIER EXTRAIT PAR UN FLUIDE SUPERCRITIQUE COMME BIOCIDE NATUREL DANS LE PROCESSUS DE TREMPAGE DE LA PEAU

RÉSUMÉ. L'activité des micro-organismes pendant le processus de trempage du cuir est très dangereuse et doit être maîtrisée. Certains composants chimiques sont utilisés à cette fin pour protéger les peaux contre l'activité microbienne. En raison des substances interdites et restreintes présentant des risques pour l'environnement et la santé ces derniers temps, les ingrédients actifs naturels sont envisagés dans la production industrielle pour la durabilité. De plus, l'extraction par fluide supercritique en tant que technologie respectueuse de l'environnement, est appliquée pour isoler des extraits biologiquement actifs, et des composés naturels extraits par fluide supercritique sont signalés comme des agents antioxydants et antibactériens potentiels pour plusieurs applications. Dans cette étude, le tanin de châtaignier extrait par un fluide supercritique a été utilisé dans le processus de trempage de la peau et a été considéré comme un biocide naturel potentiel avec un très bon effet contre l'activité des micro-organismes dans les processus humides.

MOTS CLÉS : cuir, trempage, châtaignier, biocide, bactéricide, durabilité

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INTRODUCTION

Leather production is an ancient technology that has been described as man's first manufacturing process. It comprises the transformation of raw putrescible animal hide/skin into useful materials [1, 2].

Soaking is the first and important beamhouse process of leather production and applied to gain water content to the hides/skins and soften them for preparation of the next operations. In soaking applications, several synthetic antimicrobial agents are mostly used to prevent the hides/skins against microbial activity and growth. On the other hand, considering the toxicity of these chemical components especially in the recent years, it has become an important focus of leather industry players. In the recent scientific studies, natural antimicrobial products were also reported for leather processing steps. Aloe vera was reported as antimicrobial agent for fatliquoring process of leather [3]. Quebracho, mimosa, gall-nut and chestnut vegetable tannins were used as biocide in soaking process of leather as well [4] owing to their antimicrobial properties [5, 6].

Vegetable tannins are complex and heterogeneous group of polyphenolic secondary metabolites of higher plants with molecular weights between 500 and 20,000 Da and soluble in water and polar organic solvents [7]. Although almost all plants contain tannins, only few species have sufficient amounts to be of commercial importance. Many of the commercially most significant tannin materials originate in tropical or sub-tropical climes [8].

Chestnut is a kind of fruit cultivated in Asia, Europe and America continents of the Northern Hemisphere and in South America partially. Asia is the most important region and China is the leader of Asia. Southern Europe and Turkey are the second important regions [9]. According to FAO statistics [10], the world's largest producers of chestnut are China (1,000,000 tons), South Korea (75,000 tons), Italy and Turkey (55,000 tons), and Japan (26,000 tons) [11].

Chestnut shells are the waste product from chestnut food processing and have not been fully utilised to date. The glace chestnut industry produces each year a huge amount of chestnut shells as waste [12]. The shell, which represents around 10% by weight of the chestnut, is removed in the peeling process. Chestnut wood tannins are also of the most common hydrolyzable tannins used in leather industry as vegetable tanning agent [13]. Chestnut bark is rich in tannic acid which is a specific form of tannin, a type of polyphenol [4, 14].

The extraction processes of tannins from natural matrix are currently performed by empirical methods [15, 16], especially for the leather industry. Extraction process is influenced by the chemical nature of the tannins and extraction methods have great influence on the yield, concentration and tanning phenolic content of the extract [17].

Supercritical fluid extraction (SFE) is a technique usually employed for the extraction of relatively high polar compounds by utilizing solvent extraction at elevated pressures and controlled temperatures [18-20]. It serves as a substitute to overcome the drawbacks encountered in other extraction methods such as long extraction time and high solvent consumption in conventional solvent extractions (CSE). SFE also provides advantages as increasing the mass transfer, lowering the costs and being an environmentally friendly solvent. It is called as green separation technology [21].

Supercritical extraction is a superior "green chemistry" technology which is used for the isolation of herbal extracts without the need for organic solvents. The most striking advantage is its low operating temperatures, making it possible to preserve thermo-degradable compounds and easily separate extracts. The superiority of this technology is also selective and reflected in the possibility of modeling the conditions for the production of standardized extracts with desirable attributes. It has been shown that some SFE herbal extracts have extraordinary antibacterial activity [22-25]. In this perpective, our study aims to put forward the effect of supercritical fluid extracted chestnut tannin as natural biocide in soaking process of leather.

MATERIALS AND METHODS

Materials

Chestnut shells as an industrial waste were supplied from a chestnut sweet producing food company in Turkey. Thick dry-salted domestic raw sheepskins were used as the leather material during soaking process.

Extraction of the Chestnut Tannin with SFE/Methanol and SFE/Water Method

Supercritical CO_2 extraction was carried out at 70°C under 70 bar during 60 min. (20% co-solvent: methanol) (SFE/methanol method). Water/CO_2 binary pressurized fluid system as SFE was applied with the parameters of S/F=100 with 100 bar during 3 h and at 80°C (SFE/water method) optimized in the previous study by Onem *et al*. (2015) [26] inside high pressure view cell as shown in Figure 1 below.

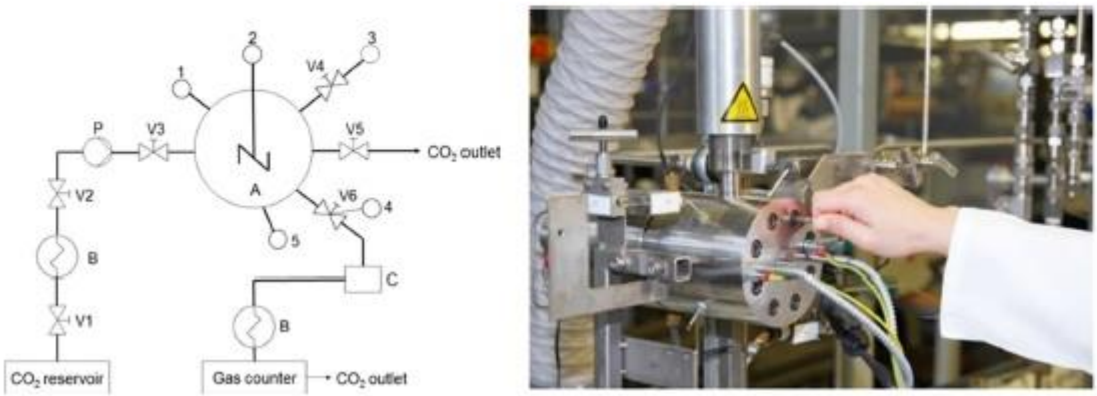

Figure 1. Experimental set-up – high-pressure view-cell for SFE operations

Extraction Yield

10 mL of samples were taken from the SFE extract solution into clean, dried and weighed glass dishes, in order to determine amount of carried solid matter into extraction medium. The extracts were dried in a hot-air oven (100±2°C) until all the water evaporated and only the solid matter was left. The dishes were then cooled in a desiccator and weighed, in this way, the amount of solid matter was determined.

Tannin Content Analysis (Hide-Powder Method)

The tanning phenolic contents of the produced chestnut extracts were determined with hide-powder method according to the official standards of SLC 114, 115, 116 and 117.

Pre-soaking and Soaking Process of Sheepskins with Natural Biocide

Pre-soaking process was applied just with water during 4 hours, then drained. The aim of the pre-soaking process is to allow the skins to take water and to remove the undesired contaminations without any protective additions to the float. The float was drained after pre-soaking, filled water again, then 1.5% SFE chestnut extract was added into the soaking bath for antimicrobial agent against bacteria. Table 1 provides the applied soaking process recipe for production.

Table 1: Applied pre- and soaking process recipe for production of the skins

Process	%	Chemicals	Temperature (°C)	Time (hour)	Remarks
Pre-soaking	500	Water	25	4	
					Drain
Soaking	500	Water	25		
	1.5	Chestnut extract			
	0.5	Soaking agent		48	
					Drain

In order to determine the bacterial growth in soaking, samples were taken from the float at the end of 4, 24 and 48 hours and the microorganisms in the float were counted.

Bacterial Counting Method in Soaking Float

1 mL soaking float sample was obtained from each process to determine the total aerobic bacteria numbers at the end of 4h, 24h and 48h. Floats were introduced into the analysis tubes having 9 mL physiological dilution water. Then, floats were diluted by 10^1 to 10^6. After serial dilutions, samples were prepared according to the pour plate method. The petri dishes were incubated during 48h in owen at 37°C. Calculations of bacterial counts were carried out in petri dishes containing 30-300 bacterial numbers. Following the 48h, total bacteria numbers in each soaking float were calculated from the colony numbers. After that, results were presented as colony forming units per millilitre (cfu/mL).

RESULTS AND DISCUSSION

According to the SFE green separation way applied to the chestnut barks, extraction yield and tannin contents of chestnut extracts produced in different SFE extraction conditions for the SFE/methanol and SFE/water methods were given in Table 2 by comparison.

Table 2: Extraction yield and tannin contents related to SFE methods

Extracts	Extraction yield (%)	Tannin content (%)
SFE/methanol method	35.23	60.10
SFE/water method	25.65	55.35

The maximum extraction yield from chestnut shell with the value of 35.23% was obtained in parameter of SFE/methanol method. Addition of methanol into supercritical CO_2 was increased the solubility and extraction yield was increased. SFE/Water method provided 25.65% extraction yield.
Tannin content of the extract was also higher in SFE/Methanol method compared to the SFE/Water method with 60.10% and 55.35% values. SFE/Methanol way was more advantageous according to the extraction and tannin yield.

Table 3 indicates the biocide application results obtained for 4 h, 24 h and 48 hours soaking time in float with SFE chestnut extract application.

Table 3: Biocide application results obtained for 4 h, 24 h and 48 hours soaking time in float

Biocide application	Colony numbers (cfu/mL), 4h (pre-soaking float)	Colony numbers (cfu/mL), 24h (soaking float)	Colony numbers (cfu/mL), 48h (soaking float)
Control float, without application	4.25×10^7	1.78×10^7	2.56×10^7
Chestnut extract (SFE/Methanol)	-	4.46×10^5	1.02×10^6
Chestnut extract (SFE/Water)	-	1.17×10^6	0.91×10^7

Added chestnut extract into the soaking bath had the superior effect on the microorganisms as biocidal application. Microorganismal activity decreased from 1.78×10^7 to 4.46×10^5 by adding the vegetable extract compared to the control soaking float at the end of 24 h by SFE/Methanol method. The beginning ratio was 4.25×10^7 during pre-soaking process after 4 h. At the end of 48 h, bacterial growth decreased from 2.56×10^7 to 1.02×10^6 by chestnut extract addition in the float compared to the control soaking process at the end of 48 h via SFE/Methanol method. Bacteria number was 1.17×10^6 and 0.91×10^7 during 24 and 48 h, relatively by SFE/Water extraction way. SFE/Methanol method was provided better effect on antibacterial activity than SFE/Water application according to the Table 3. On the other hand, both SFE techniques were determined as effective to block the bacterial growth for chestnut extract as natural biocide. This is because of the high content of the tannic acid in chestnut structure and extracted more active compounds via supercritical extraction way for chestnut compound. It is reported that chestnut bark is rich in tannic acid which is a specific form of tannin, a type of polyphenol and has a potential as biocidal effect [4, 14]. Moreover, supercritical fluid extraction method was reported in many studies that SFE provided more antibacterial and antioxidant effect to the extracted compounds compared to the conventional extraction ways [22, 27-30]. By this way, active ingredients are separated with environmentally friendly green separation way and have superior activity properties. Thus, waste materials have gained economical perspective for the industrial applications. There are also some studies on antibacterial activity of extracted tannins for textile industrial applications which is similar and close to the leather sector. Zhang *et al*. (2014) indicated the natural dye extracted from Chinese gall for the application of antibacterial activity on the wool fabrics [31].

Results proved the superior separation effect of SFE techniques for the both methanol and water ambient and effective results as potential natural biocidal agent of chestnut extracts produced.

CONCLUSIONS

In this study, a waste product, chestnut shells from food industry, was converted to a usable product with an economic value, a natural biocide used in soaking beamhouse process of leather production. 35.23% extraction and 60.10% tannin yield were obtained after SFE green separation way in carbon dioxide and methanol ambient. Environmentally friendly separation application was successfully applied for chestnut tannin. Moreover, the biocidal properties of the extracts in SFE were found acceptable and can be enhanced with different techniques for further studies. Considering that, in recent years there is a demand on usage of ecologic and natural products in the world, usage of environmentally friendly vegetable bactericidal agents and producing natural leathers can be more advantageous to gain the admiration and attention of consumers towards sustainable production, industry, life and world.

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EFFECTS OF CUSTOM-MADE ORTHOTIC INSOLES ON LOWER LIMB BIOMECHANICS IN CHILDREN WITH FLEXIBLE FLAT FEET

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EFFECT OF CUSTOM-MADE ORTHOTIC INSOLES ON LOWER LIMB BIOMECHANICS IN CHILDREN WITH FLEXIBLE FLAT FEET

ABSTRACT. This study aimed to investigate the effect of custom-made orthotic insoles on the lower limb biomechanics in children with flexible flat feet. A sample of 27 children (19 boys and eight girls) aged 7 to 13 years old with flexible flat feet participated in the study. Each custom-made orthotic insole was individually designed based on the foot of each subject, using ethylene-vinyl acetate (EVA) material with medium density. The study was conducted using a gait analysis laboratory, and participants were required to walk randomly at a self-selected speed with one of three insole conditions (custom-made orthotic insole, flat insole, and barefoot). The data was processed using Visual 3D software and analyzed by SPSS software. The results showed that using custom-made orthotic insoles resulted in a significant reduction in the maximum ankle inversion angle and ankle inversion moment while increasing the knee adduction moment during the stance phase when compared to wearing either flat insoles or being barefoot ($p<0.05$). This study found that custom-made orthotic insoles significantly altered the biomechanical characteristics of the lower limbs of children with flexible flat feet, reducing the movement of the ankle joint in the frontal plane, thereby enhancing ankle joint stability, but may increase knee joint load.

KEY WORDS: flexible flat foot; orthotic insole; ankle joint; knee joint

INFLUENȚA BRANȚURILOR ORTETICE PERSONALIZATE ASUPRA BIOMECANICII MEMBRELOR INFERIOARE LA COPIII CU PICIOARE PLATE FLEXIBILE

REZUMAT. Scopul acestui studiu a fost de a investiga influența branțurilor ortetice personalizate asupra biomecanicii membrelor inferioare la copiii cu picioare plate flexibile. S-a luat în studiu un eșantion de 27 de copii (19 băieți și opt fete) cu vârsta cuprinsă între 7 și 13 ani, cu picioare plate flexibile. Fiecare branț ortetic personalizat a fost proiectat individual pe baza formei piciorului fiecărui subiect, folosind material pe bază de etilen-acetat de vinil (EVA) cu densitate medie. Studiul a fost realizat într-un laborator de analiză a mersului, iar participanților li s-a cerut să meargă aleatoriu cu o viteză la alegere, în trei situații (cu branț ortetic personalizat, branț plat și desculț). Datele au fost prelucrate folosind software-ul Visual 3D și analizate cu software-ul SPSS. Rezultatele au arătat că utilizarea branțurilor ortetice personalizate a dus la o reducere semnificativă a unghiului maxim de inversiune a gleznei și a momentului de inversiune a gleznei, în același timp crescând momentul de aducție a genunchiului în timpul fazei de sprijin, în comparație cu purtarea branțurilor plate sau mersul cu piciorul desculț ($p<0,05$). Acest studiu a constatat că branțurile ortetice personalizate au modificat semnificativ caracteristicile biomecanice ale membrelor inferioare ale copiilor cu picioare plate flexibile, reducând mișcarea articulației gleznei în plan frontal, sporind astfel stabilitatea articulației gleznei, însă pot crește încărcarea articulației genunchiului.

CUVINTE CHEIE: picior plat flexibil; branț ortetic; articulația gleznei; articulația genunchiului

L'INFLUENCE DES SEMELLES ORTHÉTIQUES PERSONNALISÉES SUR LA BIOMÉCANIQUE DES MEMBRES INFÉRIEURS CHEZ LES ENFANTS AUX PIEDS PLATS FLEXIBLES

RÉSUMÉ. Le but de cette étude a été d'étudier l'influence des semelles orthopédiques personnalisées sur la biomécanique des membres inférieurs chez les enfants aux pieds plats flexibles. On a étudié un échantillon de 27 enfants (19 garçons et huit filles) âgés de 7 à 13 ans avec des pieds plats flexibles. Chaque semelle orthopédique personnalisée a été conçue individuellement en fonction de la forme du pied de chaque sujet en utilisant un matériau à base d'éthylène-acétate de vinyle (EVA) de densité moyenne. L'étude a été menée dans un laboratoire d'analyse de la marche et les participants ont été invités à marcher au hasard à la vitesse de leur choix dans trois situations (avec semelle orthopédique personnalisée, semelle plate et pieds nus). Les données ont été traitées à l'aide du logiciel Visual 3D et analysées avec le logiciel SPSS. Les résultats ont montré que l'utilisation de semelles orthopédiques personnalisées a entraîné une réduction significative de l'angle maximal d'inversion de la cheville et du moment d'inversion de la cheville, tout en augmentant le moment d'adduction du genou pendant la phase d'appui, par rapport au port de semelles plates ou à la marche pieds nus ($p< 0,05$). Cette étude a révélé que les semelles orthopédiques personnalisées ont modifié considérablement les caractéristiques biomécaniques des membres inférieurs des enfants aux pieds plats flexibles, réduisant le mouvement de l'articulation de la cheville dans le plan frontal, augmentant ainsi la stabilité de l'articulation de la cheville, mais pouvant augmenter la charge de l'articulation du genou.

MOTS CLÉS : pied plat flexible ; semelle orthopédique ; articulation de la cheville ; articulation du genou

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INTRODUCTION

Flexible flatfoot is a common condition in children characterized by the absence or collapse of the medial longitudinal arch of the foot [1]. This condition affects approximately 20% to 30% of the pediatric population [2]. Although it usually does not cause pain, long-term abnormal kinematics and kinetics of the lower extremity joints can lead to muscle fatigue, foot pain, asymmetry of the arches, and knee valgus [3]. If left untreated, flatfoot may result in abnormal movements of the foot and lower extremity joints, leading to continuous flattening of the arch, accelerating soft tissue damage of the foot, joint pain, and even impairing balance, stability, posture, and motor skills development in walking [4].

Non-surgical methods such as foot support, arch training, and foot massage, as well as surgical correction, are used for its treatment. Among them, orthotic insoles are one of the non-surgical methods that provide support and stability to the foot, improving the arch structure and foot function [5]. However, the curative effects of universal orthotic insoles may be limited as they cannot fully adapt to individual differences. In contrast, custom-made orthotic insoles are tailored to the patient's foot shape and condition, which may have better adaptability and treatment effects [5].

Studies have shown that custom-made orthotic insoles with arch support can relieve foot pain, improve foot comfort [6], reduce energy expenditure during movement [7], and enhance gait stability in patients with flatfoot [8]. Karimi *et al*.'s study [9] demonstrated that custom-made orthotic insoles could significantly reduce the energy expenditure of patients during movement and increase stride length and stance time. However, the majority of previous studies on the effect of orthotic insoles on flat feet have focused on gait parameters, stability, and plantar pressure distribution, and little is known about the effect of orthotic insoles on lower limb biomechanics in patients with flexible flat feet. Therefore, the objective of this study was to investigate the effect of custom-made orthotic insoles on lower limb biomechanics in children with flexible flatfoot and provide a theoretical reference for orthotic insole design.

MATERIALS AND METHODS

Participants

Sample size was determined using G*Power Analysis program (G*Power 3.1, The G*Power Team, Belgium [10]). One-way ANOVA with repeated measures was used for statistical analysis. Effect size f was 0.26, which was derived from the pre-experiment. A priori power analysis revealed that 26 subjects would be required to achieve 80% statistical power with an alpha level of 0.05. In order to account for potential drop-out and technical errors during the experiment, 27 children (19 boys and eight girls) with flexible flat feet were recruited to participate in this study. Basic information about the subjects in this study is listed in Table 1. Participation in this study was voluntary, and all participants signed an informed consent form.

Table 1: Basic information of enrolled subjects

Variables	Male	Female
Number	19	8
Age (year)	9.1±1.8	9.1±2.3
Height (cm)	136.8±13.7	134.3±25.9
Weight (kg)	35.4±13.1	34.7±11.7
BMI (kg/m^2)	18.3±3.7	17.4±3.7

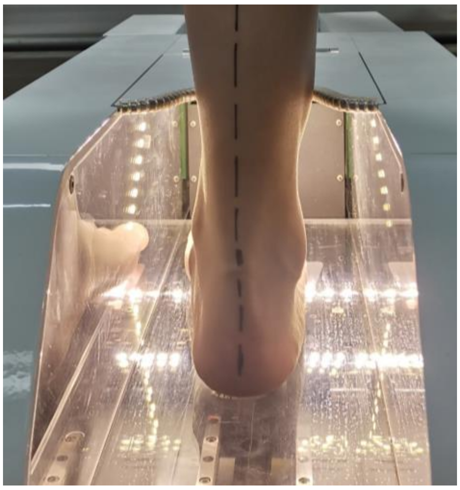
Note: BMI, Body mass index

The inclusion criteria of this study included: (i) children aged 7 to 13 years with flexible flatfoot diagnosed by an orthotic surgeon; (ii) have no history of lower limb injuries or surgeries within the previous six months; and (iii) have no history of wearing custom-made orthotic insole. Exclusion criteria were: (i) rigid flatfoot; (ii) being overweight, (iii) being unable to complete the experimental procedures as required.

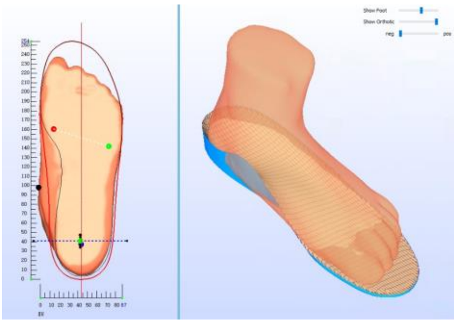
Materials

The custom-made orthotic insole is made of medium-density ethylene-vinyl acetate (EVA). It was designed with two parts to provide both comfort in the forefoot area and support in the rearfoot region. This combination of features helps to promote proper foot alignment and alleviate discomfort for the patient with flexible flat feet. The hardness of the fore and rear parts are Shore C 50° and Shore C 60°, respectively. Each custom-made orthotic insole is custom-made for an individual participant. The process involves taking 3D scans (Upod-s, Carotec Ltd., Guangzhou, China, Figure 1-a) of the subject's feet while they were in a non-weight bearing neutral position [11] and then using these scans to design (IsoleCAD5.4.0, Vismach Technology Ltd., Wuhan, China) and produce a personalized insole for each participant. The design process of custom-made orthotic insoles is shown in Figure 1. This study used the flat insole as a control condition with a thickness of 4mm and a hardness rating of 50° on the Shore C scale. In addition, the same type of children's sports shoes (361° CO. Ltd., Xiamen, China) and cotton socks were used to control for any potential influence they may have on the results. The shoes, orthotic insoles, and flat insoles used in this study are shown in Figure 2. In this study, the orthotic insole's heel cup has a height of 12mm, while the actual thickness of the rear part of the insole remains at 4mm (Figure 2-b). The primary function of the heel cup is to improve the heel's containment and stability within the shoe, without significantly altering the internal dimensions of the shoe cavity or causing discomfort for the subject during wearing.

a. Foot scanner used in this study

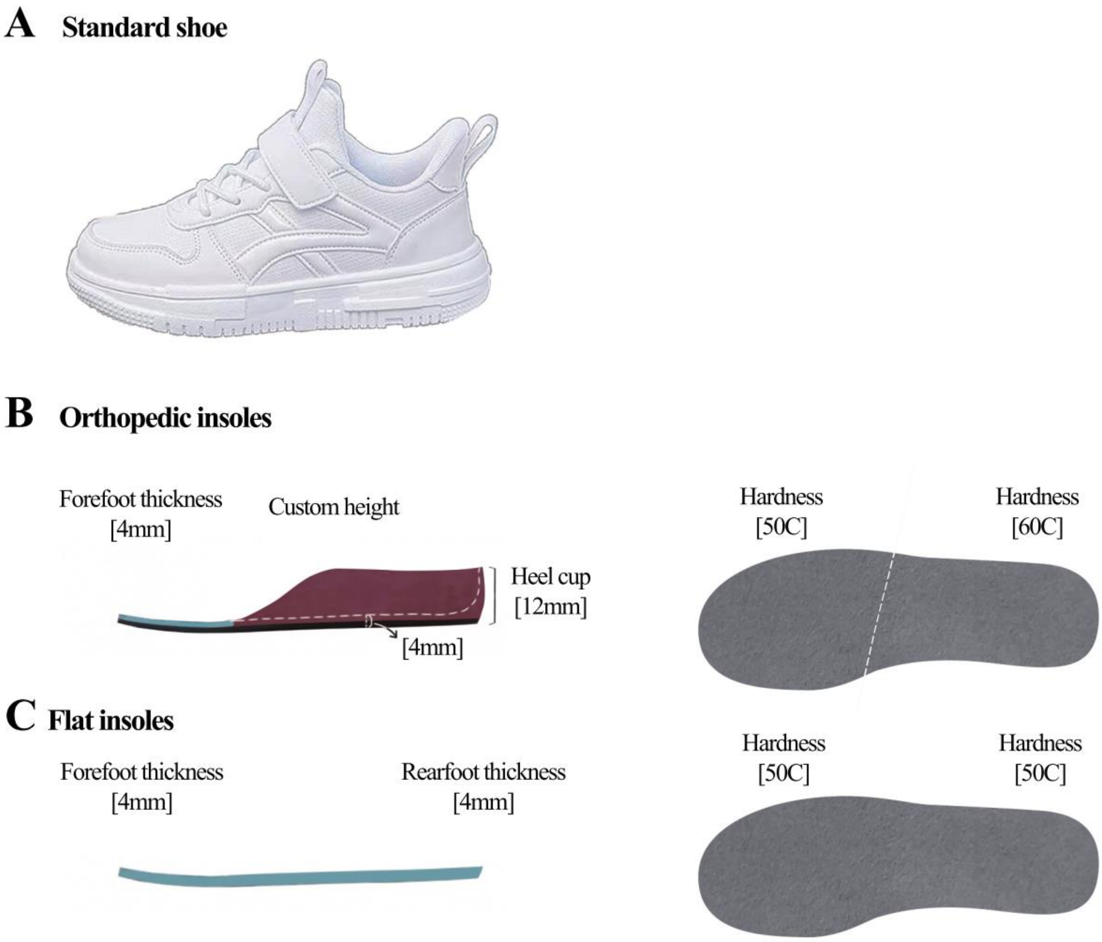


b. Foot scan process



c. Custom-made orthotic insole design process

Figure 1. Foot scan and orthotic insole design process used in this study



Custom-made orthotic insoles

Flat insoles

Figure 2. Shoe and insoles used in this study

Experimental Set-Up and Procedures

Overground gait analysis in the present study was performed in a gait analysis lab. Two force plates (Kistler9260AA6, Winterthur, Switzerland, Figure 3-a) were placed in the middle of a 10 m walkway to acquire ground reaction forces of the left and right lower extremities during the stance phase of a walking cycle. Thirty-seven reflective markers placed bilaterally on the pelvis, thighs, shanks, and feet [12] were tracked using a 12-camera motion capture system (ProReflex MCU 1000, Qualisys AB, Sweden, Figure 3-b). Participants were required to walk randomly at a self-selected speed with one of the three insole conditions (custom-made orthotic insole, flat insole, and barefoot). Kinematics and kinetics data were recorded at 100 Hz and 1000 Hz sampling rates, respectively. For gait analysis, three successful self-selected speed gait trials were collected where the foot naturally landed on the force plate, and the gait cycle was detected.

a. Force plate b. Motion capture system

Figure 3. The force plate and motion capture system used in this study

Data Reduction and Processing

The kinematic and kinetic data were processed using Visual 3D software (C-motion, Rockville, MD, USA) and a fourth-order Butterworth filter was applied with cut-off frequencies of 10 and 50 Hz to filter marker trajectories and ground reaction forces [13]. The 3D joint angles and moments were calculated at the distal segment relative to the adjacent proximal segment using a right-handed orthogonal Cardan x-y-z sequence of rotations. This sequence was chosen to align with the joint coordinate system and included rotations for plantarflexion/dorsiflexion, eversion/inversion, and abduction/adduction [14]. Hip, knee, and ankle joint angles and moments were calculated using inverse kinematics and inverse dynamics for the stance phase. Positive values of the joint angles and moments indicate extension, adduction, internal rotation, dorsiflexion, and inversion, depending on the joint and axis. The time series of each stance phase were normalized to a range of 0% at heel strike on the force plate to 100% at toe-off. Joint moments were normalized by dividing them by body mass and expressed in N·m/kg [15].

Statistical Analysis

In this study, all analyses were performed by SPSS software (version 21.0, SPSS Inc., USA). All data were represented as the mean ± S.D. One-way repeated measures analysis of variance (ANOVA) was performed to examine if there were significant differences in lower limb kinematic and kinetic variables across three insole conditions. Post hoc Bonferroni tests were performed to examine the differences in each dependent variable between insole conditions when a significant difference was observed. The significance level was set at 0.05 ($p < 0.05$).

RESULTS

Kinematics

Table 2 presents the lower limb joint kinematics results for the three insole conditions. The result of this study indicated that different insole conditions significantly impacted ankle joint kinematics ($p<0.05$). The peak hip flexion angle was significantly decreased in custom-made orthotic insoles ($p=0.0003$) and flat insoles ($p=0.0184$)

compared to barefoot. Custom-made orthotic insoles resulted in a significantly greater peak hip adduction angle than flat insoles (p=0.0399).

Results showed a significant increase in peak knee flexion angle (p<0.0001) but a significant decrease in peak knee extension angle (p<0.05) when subjects wore custom-made orthotic insoles compared to being barefoot. No significant differences were observed in knee joint kinematics between custom-made orthotic insoles and flat insoles (P>0.05).

Results showed that different insole conditions significantly impacted ankle joint kinematics (p<0.05). Specifically, the peak ankle dorsiflexion angle significantly increased in both custom-made orthotic insoles (p=0.0001) and flat insoles (p=0.0003) compared to barefoot. Custom-made orthotic insoles resulted in a significantly greater peak ankle dorsiflexion angle than flat insoles (p=0.0247). Additionally, custom-made orthotic insoles and flat insoles resulted in less ankle inversion angle (p=0.0154) and ankle internal rotation angle (p=0.0375) compared to the barefoot condition.

Table 2: Lower limb joint kinematics in three insole conditions during walking (°)

Variables	Barefoot	Flat insoles	Custom-made orthotic insoles	p-value
Peak hip extension angle	36.19±6.41	36.23±6.72	36.16±7.23	0.9909
Peak hip flexion angle	-4.19±6.47	-2.81±6.02*	-2.09±6.62*	0.0002
Peak hip adduction angle	5.69±3.05	5.61±3.35	5.09±3.35#	0.0907
Peak hip abduction angle	-7.90±4.28	-7.64±3.84	-7.81±3.77	0.7270
Peak hip internal rotation angle	-5.92±7.36	-4.61±6.69	-6.08±6.75	0.1922
Peak hip external rotation angle	-15.07±7.28	-13.85±6.08	-14.71±6.48	0.2439
Peak knee extension angle	-12.24±4.64	-9.08±4.31*	-8.64±4.45*	<0.0001
Peak knee flexion angle	-46.72±5.68	-55.13±3.85*	-54.63±4.69*	<0.0001
Peak knee varus angle	3.61±3.81	3.45±3.62	3.58±3.94	0.8855
Peak knee valgus angle	-3.64±3.44	-3.03±2.79	-3.42±3.30	0.3514
Peak knee internal rotation angle	3.39±6.78	3.05±6.40	3.07±6.68	0.8891
Peak knee external rotation angle	-13.80±5.00	-11.88±5.04	-11.86±5.76	0.0521
Peak ankle dorsiflexion angle	14.53±4.39	17.29±4.30*	18.42±4.30*#	<0.0001
Peak ankle plantar Flexion angle	-14.09±6.24	-12.01±5.81	-11.92±5.41	0.0245
Peak ankle inversion angle	6.09±7.54	3.45±4.49*	2.21±4.21*	0.0134
Peak ankle eversion angle	-9.69±7.11	-9.05±3.70	-9.72±3.07	0.6817
Peak ankle internal rotation angle	12.40±5.02	10.82±5.85	9.51±6.30*	0.0274
Peak ankle external rotation angle	-6.69±4.27	-8.66±4.87	-8.71±4.50	0.0377

Note: *indicates a statistically significant difference from the barefoot condition, and #indicates a statistically significant difference from the flat insoles condition.

Kinetics

Table 3 shows the joint moments in the sagittal, frontal, and transverse planes for the hip, knee and ankle joints. Results showed a significant decrease in peak hip flexion moment (p=0.0341) and peak hip extension moment (p=0.0002) when subjects wore custom-made orthotic insoles compared to barefoot. Additionally, the peak hip flexion moment (p<0.0001) and peak hip internal rotation moment (p=0.0441) were significantly lower when subjects wore the flat insole compared to being barefoot.

Results showed a significant increase in peak knee abduction moment (p=0.0399) but a significant decrease in peak knee external rotation moment (p=0.0095) when subjects wore custom-made orthotic insoles compared to barefoot.

Specifically, the peak ankle dorsiflexion moment was significantly increased in both custom-made orthotic insoles (p<0.0001) and flat insoles (p<0.0001) compared to being barefoot. Additionally, custom-made orthotic insoles (p=0.0011) and flat insoles (p=0.0049) resulted in a lesser peak ankle inversion moment than barefoot. Custom-made orthotic insoles resulted in a significantly lesser peak ankle inversion moment than flat insoles (p=0.0047).

Table 3: Lower limb joint kinetics in three insole conditions during walking (Nm/Kg)

Variables	Barefoot	Flat insoles	Custom-made orthotic insoles	p-value
Peak hip extension moment	0.41±0.10	0.39±0.10	0.39±0.10*	0.0237
Peak hip flexion moment	-0.46±0.20	-0.32±0.13*	-0.33±0.15*	<0.0001
Peak hip adduction moment	0.12±0.05	0.11±0.04	0.11±0.04	0.2520
Peak hip abduction moment	-0.3±0.14	-0.31±0.15	-0.31±0.17	0.3792
Peak hip internal rotation moment	0.06±0.03	0.05±0.02*	0.06±0.02	0.0209
Peak hip external rotation moment	-0.10±0.04	-0.10±0.04	-0.10±0.04	0.7415
Peak knee extension moment	0.73±0.39	0.68±0.36	0.73±0.36	0.2138
Peak knee flexion moment	-0.43±0.28	-0.39±0.19	-0.37±0.22	0.2059
Peak knee adduction moment	0.20±0.13	0.17±0.10	0.17±0.10	0.0639
Peak knee abduction moment	-0.37±0.19	-0.39±0.21	-0.41±0.24*	0.0170
Peak knee internal rotation moment	0.17±0.09	0.17±0.09	0.18±0.09	0.2249
Peak knee external rotation moment	-0.10±0.05	-0.09±0.05	-0.08±0.04*	0.0057
Peak ankle dorsiflexion moment	0.35±0.15	0.48±0.20*	0.49±0.21*	<0.0001
Peak ankle plantar flexion moment	-1.44±0.64	-1.43±0.58	-1.40±0.53	0.3427
Peak ankle inversion moment	0.28±0.16	0.22±0.11*	0.19±0.10*#	0.0003
Peak ankle eversion moment	-0.05±0.05	-0.07±0.05	-0.08±0.07	0.0759
Peak ankle internal rotation moment	0.05±0.03	0.05±0.02	0.05±0.02	0.2678
Peak ankle external rotation moment	-0.11±0.04	-0.12±0.06	-0.12±0.05	0.1586

Note: *indicates a statistically significant difference from the barefoot condition, and #indicates a statistically significant difference from the flat insoles condition.

DISCUSSION

Using custom-made orthotic insoles in children with flexible flat feet has been an area of interest for clinicians and researchers. The present study aimed to investigate the effect of custom-made orthotic insoles on the lower limb joint kinematics and moments in children with flexible flat feet during the stance phase of gait. The results showed that custom-made orthotic insoles had a significant impact on hip, knee, and ankle joint biomechanics during gait. The specific effects of custom-made orthotic insoles on each lower limb joint and the study's limitations will be discussed as follows.

Effect of Custom-made Orthotic Insoles on the Hip Joint

The study found that custom-made orthotic insoles could reduce the hip flexion angle, hip extension angle, and hip extension moment, which is consistent with previous research findings [16]. However, the exact mechanism by which custom-made orthotic insoles reduce hip joint flexion and extension is unclear. It could be due to changes in the plantar pressure distribution, alterations in muscle activity, or a combination of both factors. Further research is needed to determine the specific impact of custom-made orthotic insoles on hip joint flexion and extension.

Effect of Custom-made Orthotic Insoles on the Knee Joint

The study showed that knee joint flexion and extension significantly increased in the custom-made orthotic insoles and flat insoles compared to the barefoot condition, which is consistent with the findings of Chen *et al*. [17]. This could be due to the cushioning effect that shoes and insoles provide during walking, which enhances knee flexion and extension mobility. In addition, the internal and external rotation of the knee joint was significantly reduced in both insole conditions, suggesting that wearing both regular and custom-made orthotic insoles reduced the rotational motion of the tibia relative to the femur in children with flexible flat feet. This suggests that wearing the insoles was beneficial in limiting the movement of the knee in the transverse plane.

However, in terms of the knee motion in the frontal plane, results showed that the peak knee abduction moment was significantly increased when subjects wore the custom-made orthotic insole than the barefoot condition. These findings can be explained by the custom-made orthotic insole with arch support shifting the center of pressure inward, resulting in an increased lever arm of the knee joint center to the ground reaction force vector, leading to increased external knee abduction moments. The knee muscles produce more joint muscle moments to balance the increased external knee abduction moment [18]. These findings suggested that wearing custom-made orthotic insoles may increase the stress on the knee joint in the frontal plane. It highlights the need for further research to understand the potential side effects of custom-made orthotic insoles on the knee joint, especially in patients with knee problems or in conditions where the knee joint is already under stress.

Effect of Custom-made Orthotic Insoles on the Ankle Joint

The results of this study indicated that using custom-made orthotic insoles had a substantial impact on ankle joint biomechanics. The study found that the peak ankle dorsiflexion angle and moments were increased, and the peak ankle inversion angle and moments were decreased when subjects wore custom-made orthotic insoles, which is consistent with the finding of Arvin [19] and Hsu *et al*. [20]. The increased ankle dorsiflexion motion in the sagittal plane could enhance the range of motion in the ankle joint and improve its flexion and extension capabilities. The reduced ankle inversion angle and moments observed in children with flat feet who use custom-made orthotic insoles with arch support may be attributed to a shift in the center of pressure towards the inside of the foot. This shift decreases the lever arm between the ankle joint center and the ground reaction force (GRF) vector [21], decreasing the external moment of ankle eversion and a corresponding reduction in the muscle moment responsible for ankle inversion around the ankle joint. The findings in this study suggested that custom-made orthotic insoles could limit the ankle motion in the frontal plane, help improve the stability of the ankle joint, and reduce the strain on the ankle joint during the stance phase, which is consistent with the findings of Karimi *et al*. [22]. These findings suggest that custom-made orthotic insoles may be an effective intervention for children with flexible flat feet. However, more research is required to understand their long-term effects.

It is important to note that the current study has some limitations. It only analyzed the immediate effects of custom-made orthotic insoles on lower limb biomechanics and did not provide information on their long-term effects. Further research that combines surface electromyography tests with kinematic and kinetic variables on the lower limbs could provide valuable insight into the effects of custom-made orthotic insoles. Additionally, it would be helpful to study the impact of other variables, such as different types of movements, movement speeds, insole materials, and hardness, on lower limb biomechanics in children with flexible flat feet.

CONCLUSIONS

Results in this study indicated that custom-made orthotic insoles could effectively reduce ankle joint moments in patients with flexible flat feet during gait. However, the results also suggested that using custom-made orthotic insoles may have an unintended side effect on the knee joint, as the knee abduction moment was significantly increased in the orthotic condition compared to the barefoot condition. This highlights the importance of considering both the positive and negative effects of custom-made orthotic insoles and carefully evaluating their use in each individual case. Further research is needed to fully understand the effects of custom-made orthotic insoles on the mechanics of the ankle and knee joints in patients with flexible flat feet during gait. It is recommended that children with flexible flat feet be given an adjustment period to get used to wearing custom-made orthotic insoles. It can be done by gradually increasing the time the child wears the insoles daily, allowing them to adapt to the insoles and reduce the potential adverse effects on the knee joint.

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EXPERT EXAMINATION OF LEATHER IN INTERNATIONAL TRADE: UKRAINIAN EXPERIENCE

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EXPERT EXAMINATION OF LEATHER IN INTERNATIONAL TRADE: UKRAINIAN EXPERIENCE

ABSTRACT. In the article the state of the market, volumes of export and import of leather are analyzed. The procedure of leather identification expert examination for customs purposes in international trade is showed. Identification expert examination of leather was carried out and its results were issued for customs purposes. The code of leather according to the Harmonized Commodity Description and Coding System (HS) and the Ukrainian Classification of Goods for Foreign Economic Activity (UCGFEA) is defined based on the results of identification expert examination.
KEY WORDS: leather, genuine leather, export, import, identification expert examination, customs purpose, international trade, the HS, the UCGFEA.

EXAMINARE SPECIALIZATĂ A PIEILOR ÎN COMERȚUL INTERNAȚIONAL: EXPERIENȚA UCRAINEI

REZUMAT. Articolul analizează starea pieței, volumele de export și import de piele. Este prezentată procedura de examinare specializată pentru identificarea pieilor în scopuri vamale în comerțul internațional. S-a efectuat examinarea pieilor pentru identificare, iar rezultatele acesteia au fost emise în scopuri vamale. Codul pentru piele în conformitate cu Sistemul armonizat de denumire și codificare a mărfurilor (SA) și Clasificarea ucraineană a mărfurilor pentru activități economice străine (UCGFEA) este definit pe baza rezultatelor examinării specializate.
CUVINTE CHEIE: piele, piele naturală, export, import, examinare specializată, scop vamal, comerț internațional, SA, UCGFEA.

L'EXAMEN D'EXPERTS DU CUIR DANS LE COMMERCE INTERNATIONAL : L'EXPÉRIENCE UKRAINIENNE

RÉSUMÉ. Dans l'article, l'état du marché, les volumes d'exportation et d'importation de cuir sont analysés. La procédure pour l'examen d'experts pour l'identification du cuir à des fins douanières dans le commerce international est présentée. Une expertise d'identification du cuir a été réalisée et ses résultats ont été délivrés à des fins douanières. Le code du cuir selon le Système harmonisé de désignation et de codification des marchandises (SH) et la Classification ukrainienne des marchandises résultant d'une activité économique avec l'étranger (UCGFEA) est défini sur la base des résultats de l'examen d'experts pour l'identification.
MOTS CLÉS : cuir, cuir véritable, exportation, importation, expertise d'identification, des fins douanières, commerce international, le SH, l'UCGFEA.

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INTRODUCTION

Leather goods are articles or materials created from leather. Full-grain leather, top grain leather, corrected grain articles, tanned leather, and more forms of leather exist. Gloves, garments, haberdashery, bags, footwear, furniture, automotive upholstery leathers, and other items made of leather are examples of leather goods. One of the primary driving factors influencing market expansion is an increase in the demand for leather in the creation of garments.

Leather industry is one of the oldest manufacturing sectors. The raw material used in leather industry is derived from the livestock or animal farming industry, then being put through several processes including tanning and finishing etc. Today, leather is still one of the most luxury materials that are used to produce a wide range of high-end products such as leather footwear, leather bags, leather garments, and so on.

The market is mainly driven by rising consumer disposable income, improved living standards, changing fashion trends, and growing domestic and international tourism. The rising demand for comfortable, trendy, and fancy leather apparel, footwear, and accessories, along with growing brand awareness, is expected to have a positive impact on the market. According to the growth of demand for goods, the demand for leather will also increase. With technical development the production of leather substitutes increases, therefore at customs clearance it is important to carry out identification expert examination.

The assortment of leather and leather-like products is quite wide: genuine, artificial and synthetic leather. Depending on the stages of processing, natural leathers are processed hides and usually without hair. These characteristics affect the classification of imported goods, their customs value, duty rate, non-tariff regulation measures and other peculiarities of international trade operations.

In order to levy the correct amount of customs duties, customs officials should check the correctness of leather identification and classification. Leather as object of international trade should be classified according to the Harmonized Commodity Description and Coding System (HS). The Ukrainian Classification of Goods for Foreign Economic Activity (UCGFEA) developed based on the HS and is used in foreign trade activity of Ukraine. The identification expert examination allows to confirm that the goods belong to the UCGFEA corresponding code. So, identification expert examination is very important in leather materials trade and determination compliance of goods to code according to the UCGFEA.

The article aims to develop criteria, indicators, methods and means of expert examination of natural leather and conduct an expert examination of natural leather for customs purposes in international trade. The analysis of leather market related to Ukraine's position in the world was presented.

The World and Ukrainian Market of Leather

The global Leather Goods market was valued at USD 400 billion (bln) in 2021 which is expected to reach USD 703.5 bln by 2027 at a Compound Annual Growth Rate (CAGR) 8.78 % from 2021-2027 [1].

There are significant volumes of leather imports to Ukraine, namely 155.3 million (mln) USD in 2021. Moreover, tanned leather with additional processing was imported in the amount of 141.8 mln USD. Ukraine's imports represent 1.7 % of world imports for this product, and our country ranking in world imports is 18 [2].

In the foreign economic activity of the enterprises of the leather industry, the ratio of import-export depends on the state of the processing material: raw materials, semi-finished products, crust or finished leather.

In order to analyze exports and imports volume as well as for customs clearance purpose it is quite important to correctly classify leather according to the UCGFEA and HS. According to the mentioned documents genuine leather classifiers in section VIII, group 41 Raw hides and skins (other than natural and faux fur), tanned leather (or semi-finished items) and finished leather [3].

In turn, the group is divided into commodity heading. So raw hides have the code 4101-4103, which depends on the type of raw material: 4101 – from the hides of cattle or animals of the equine family; 4102 – from skins of sheep or lambs; 4103 – from other animals. Tanned (semi-finished items) or crusty leather (crust leathers are tanned leathers processed by neutralization and fatliquoring or neutralization, retanning, dyeing and fatliquoring) is classified under codes 4104-4106, which also depends on the type of raw material: 4104 – from the hides of cattle or animals of the equine family; 4105 – from skins of sheep or lambs; 4106 – from other animals. Leather additionally treated after tanning is classified under codes 4107-4115. The code number depends on the type of raw material and method of finishing: 4107 – from the hides of cattle or animals of the equine family; 4112 – from skins of sheep or lambs; 4113 – from other animals; 4114 – suede; patent leather and multilayer patent leather; metallized leather and 4115 – composite leather for comparison of data.

According to the International Trade Center data [2], we will analyze the dynamics and volume of imports to Ukraine in group 41 during 2016-2021 (Fig. 1). In general, in 2021 products of group 41 were imported to Ukraine in the amount of 155.3 mln USD. Headings 4101, 4104 and 4107 are characterized by the same trend of import development during 2016-2021 and occupies the largest shares in the volume of import. The main demand is observed for commodity heading 4107, in 2021 imports of 4107 amounted to 90.2% among group 41. The share of imports of 4101 is 3.5%, 4104-3.2%, 4113-1.1%. The share of imports in commodity headings 4112, 4114, 4115 is less than 1%. Import of product heading 4102, 4103, 4105, 4106, 4108, 4109, 4110, 4111 is absent or in very small volumes, which allows them to be neglected.

There is an increase in imports during 2016-2018, and in 2019-2020 – decline. All commodity headings are characterized by a decline in 2020 due to the global pandemic. Import of leather increased in 2021, with the exception of product headings 4113 and 4114.

Thus, by type of raw material, the greatest demand is for leathers made from the hides of cattle or animals. The largest share is occupied by leather treated after tanning.

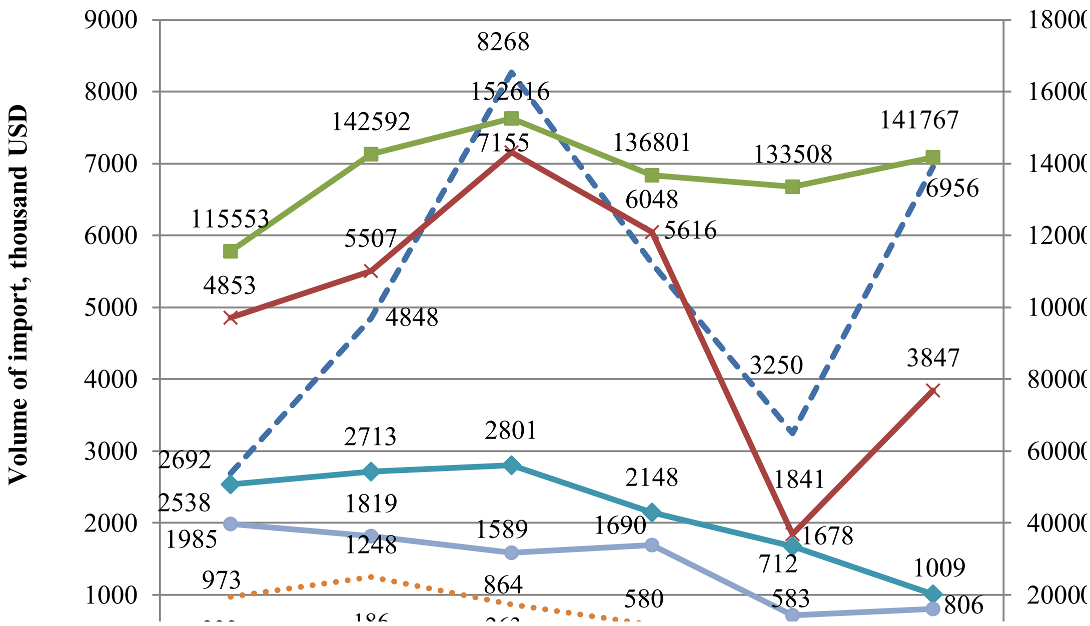


Figure 1. Volume and dynamics of imports of leather during 2016-2021, thousand USD [2]

Then we will analyze the geographical structure of export-import operations with goods of group 41 in the world in 2016-2021. The top exporting countries by the value of exported products are shown in Figure 2. The world leader is Italy with a share of 19%. Next are the USA with a share of 9.9% and Brazil with a share of 7.5%. Ukraine has only 0.5% of the market.

The top importing countries in terms of the imported products value in group 41 in 2021 are shown in Figure 2. The leader among importers is China with a market share of 20%. Also in the top 3 is Italy with a share of 12.2% and Vietnam – 8.9%. Ukraine occupies 0.8% of the import market.

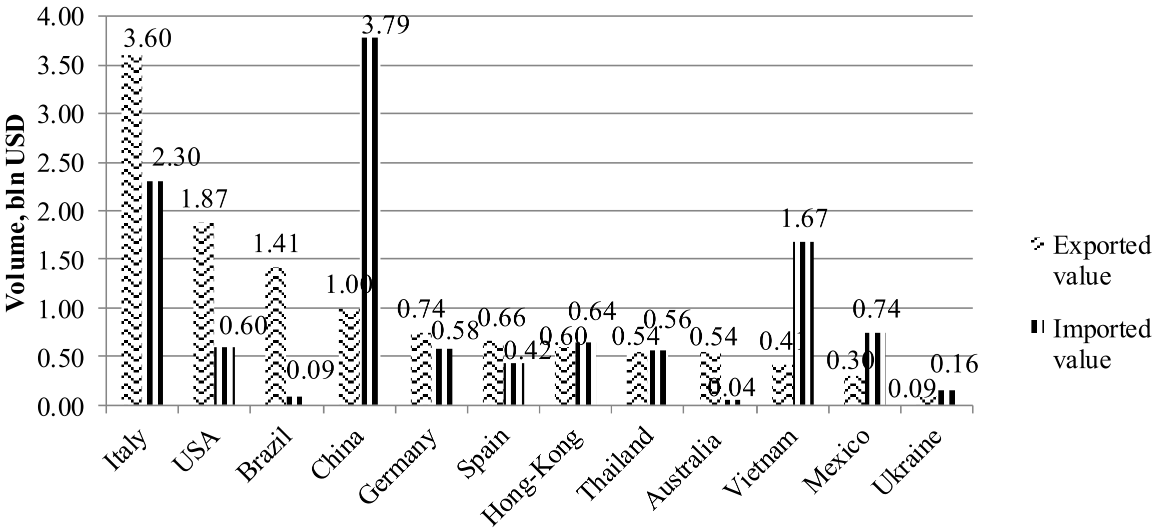


Figure 2. Exporting and importing countries of product of the group 41 in the world in 2021, bln USD [2]

As we have determined, the greatest demand among imported leathers is the leather of heading 4107. Also, we will identify the main counterparties that exported leather of heading 4107 to Ukraine (Fig. 3) [2].

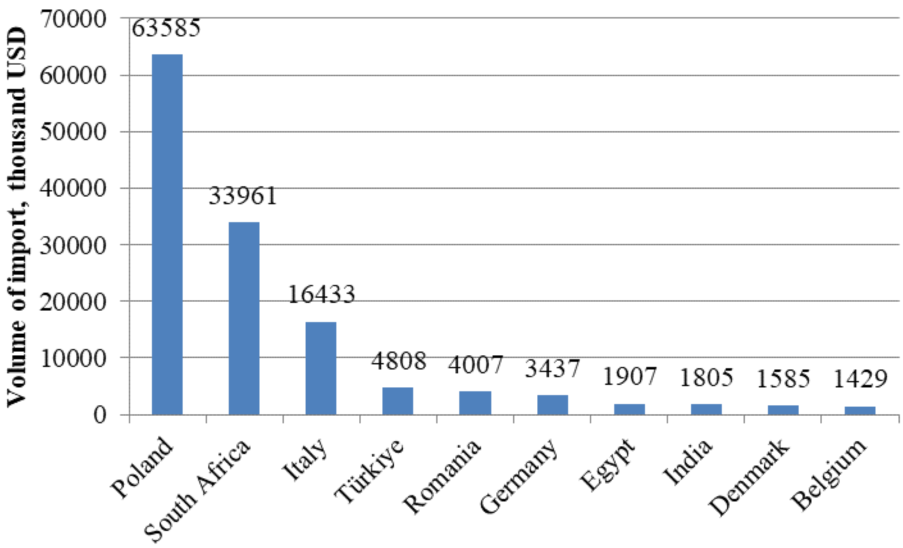


Figure 3. The biggest exporting countries of heading 4107 leather in 2021 to Ukraine, thousand USD [2]

The total import to Ukraine of goods of commodity position 4107 in 2021 amounted to 141.767 mln USD. Ukraine's imports represent 1.7% of world imports for this product, its ranking in world imports is 18. The biggest exporting countries are Poland with a share of 44.9%, South Africa – 24.0%, Italy – 11.6% (Fig. 4).

The most important outlets for European Union (EU) tanners' production (Fig.

4) are footwear, leather goods, furniture and automotive industry [4].

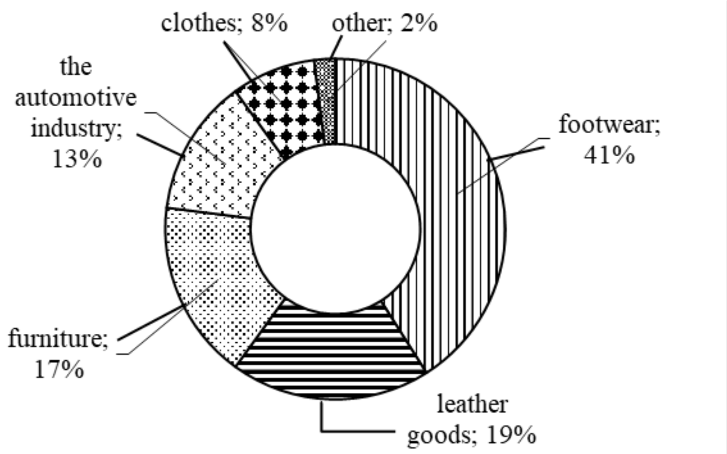


Figure 4. Outlets for EU tanners' production [4]

The processing of hides and skins also generates other by-products, which find outlets in several industry sectors such as pet and animal food production, fine chemicals (including photography and cosmetics), and fertilizers.

Geographically, Asia Pacific is the currently the largest market for leather and leather goods, the region has generated the highest revenue in the world for leather goods over the recent years (Table 1). The developing countries such as India, China and Dubai are expected to pose the highest market demand in the near future, mainly due to the rising disposable income and rising inclination towards designer leather bags and accessories. North America also holds one of the biggest markets for this industry in terms of leather imports and leather products consumption [5, 6].

Table 1: Leading leather production countries of the world

№	Country	Average annual production (million sq. ft)	Share of global production, %
1	China	6170	25
2	Brazil	2360	9.5
3	Russia	1652	7
4	India	1560	6.4
5	Italy	1521	6.3
6	South Korea	1140	4.8
7	Argentina	804	3.4
8	USA	719	3
9	Mexico	642	2.7
10	Turkey	529	2.2

China is the largest leather producing country in the world with over 6.1 bln sq ft leather produced every year, representing more than 25% of the annual global leather production. China's leather industry is renowned for producing heavy leather used for manufacturing belts, straps and soles, while light leather used in shoes, bags, and jackets. Most of China's leather output is derived from light bovine hide which accounts for nearly 40% of the total production per year, making China the biggest producer of bovine leather in the world as well. The light leather of sheep and goats is the second largest leather source in China's leather industry.

Brazil is the second largest leather producing country in the world. Brazil's leather industry produces over 2.3 bln sq ft of leather per annum. Leather production in Brazil is majorly light leather produced from bovine. Leather produced from bovine in Brazil is about 1.8 bln sq ft a year, making it the biggest contributor of Brazil's leather industry. Sheep and goat leathers make about 4 percent of the leather industry in Brazil.

So, the global leather industry is booming due to the growing demand for leather products worldwide. The Asia-Pacific is the biggest region in manufacturing of genuine leather, second – South America, third – Europe. The main countries, which exported genuine leather into Ukraine are Poland, South Africa, Italy and Germany. Over the recent years, the growing demand for luxury fashion products such as handbags, wallets and other fashion accessories driven by the rising spending on personal goods has resulted from the rapid increase in the leather production and leather trade around the world.

EXPERIMENTAL

Materials and Methods

Identification expert examination of leather as well as other goods in international trade is appointed, if for clarifying questions that arise in the case of violation of customs rules, there was a need for special knowledge in different sphere of science, technology, art, etc.

Identification expert examination of goods is an important stage of customs control and clearance, the purpose of which is to establish the conformity of information (properties, qualities, indicators) presented for customs control and customs clearance goods, the information contained in the shipping, commercial and other documents on these goods. The obtained results customs officers use for correct classification code of genuine leather according to the UCGFEA as well as determination belonging to leather analogs.

Thus, the object of the research is genuine leather imported to Ukraine. Identification expert examination for customs purposes were conducted using three samples, described in Table 2.

Table 2: Characteristics of leather samples as object of expert examination

Sample No.	Appearance	Name and address of importer	Manufacturer	Country of origin
1		LLC “TC OKTAVA CENTER” 08153, city Boyarka, 51 Bilogorodska str.	Green point	Italy
2		LLC “FOOD INDUSTRIAL” 61033, Kharkiv, Kyiv district, Vologodskyi entrance 2-y, building 2	No information available	Poland
3		LLC “FOOD INDUSTRIAL” 61033, Kharkiv, Kyiv district, Vologodskyi entrance 2-y, building 2	No information available	Poland

To conduct identification expert examination of leather, the following groups of indicators were determined: labeling conformity assessment; organoleptic; instrumental (physical-chemical) and microscopic. Organoleptic research helped to determine the following leather indicators: color, grain pattern, configuration, additional processing. Instrumental (infrared spectroscopy analysis) – to determine qualitative chemical composition by identification of functional groups. Microscopic – to determine grain pattern and structure identification.

External examination was conducted in accordance with the SSTU 2341:94 and ISO 15115:2019 using organoleptic method [7, 8].

The analysis of the grain pattern was performed by comparing with our database of leather samples.

Depending on the hide section of the leather received for the study, either its area (for whole leather) or its linear size (for part of the leather) is determined.

Analysis by infrared (IR) spectroscopy was performed using the Avatar 370 FT-IR Termo Nicolet with Fourier transform additionally equipped with the ZnSe crystal module, which eliminates the need for special sample preparation. The selected leather sample was placed in the device and was pressed with the ZnSe crystal module during the research. Infrared spectra were obtained in the wavelength range 4000-650 cm^{-1}.

To study the structure of the researched samples, a microscopic examination was performed. Microscopic examination of the leather is used to analyze their structure using optical or electronic microscopes on specially prepared samples. Methods of microanalysis determine the shape of the fibers, in compliance with the morphological features inherent in leather. Microscopic examination was performed according to ISO 17131:2012 [9] using an Olympus CX 31 microscope at 10x and 40x magnification and a SZM-45TL stereoscopic microscope at ×7- ×45 magnification.

The identification expert examination was conducted on the basis of the Specialized Laboratory for Expert Examination and Research (SLEER) of the State Customs Service of Ukraine (SCSU) and laboratories of Commodity Science and Customs Affairs Department of State University of Trade and Economics (SUTE).

RESULTS AND DISCUSSIONS

Identification Expert Examination of Leather for Customs Purposes

For conducting identification expert examination of leather, customs officers must send samples to SLEER of the SCSU in order to establish the characteristics influence to classification in accordance with the UCGFEA.

The identification criteria are the characteristics of the goods, which allow to identify the name of the presented goods with the name indicated on the label or in regulatory, shipping documents. The developed criteria, means and methods of identification expert examination of leather are presented in Table 3.

Table 3: Criteria/indicators, means and methods of identification expert examination of leather

Criteria/indicators	Means	Methods
	General	
Product name	Markings, accompanying documents	Compliance check
Manufacturer’s name	Markings, accompanying documents	Compliance check
Country of origin	Markings, accompanying documents	Compliance check
	Specific	
Color	Goods, accompanying documents	Organoleptic
Grain pattern	Goods, characteristic images of different leather grain patterns	Visual and microscopic
Chemical composition	Goods, Working instructions Qualitative determination of individual substances, polymeric compounds and components in mixtures products by IR spectroscopy [10, 11]	Qualitative by ATR-FTIR
Morphological structure	Goods, ISO 17131:2020 [9]	Qualitative by microscopy

Means of leather identification are regulations that measure quality indicators and can be used for identification. Identification expert examination of leather includes checking the condition of packaging and labeling, identification of leather, sampling, conducting of organoleptic, physical-chemical parameters and microscopic analysis.

Grain pattern is an important identification parameter of leather. Individual features of each animal species are reflected not only in the general structure of the skin, but also in the nature of the pattern, so the leather of each animal corresponds to a certain pattern (Fig. 5), in the most significant features that does not change depending on breed, sex or age. The ability to recognize the origin of the leather by its pattern is based on this. Some types of leather, such as the leather of lizards and snakes, have an extremely unique pattern and color, which gives an original look to shoes and other products made of them.

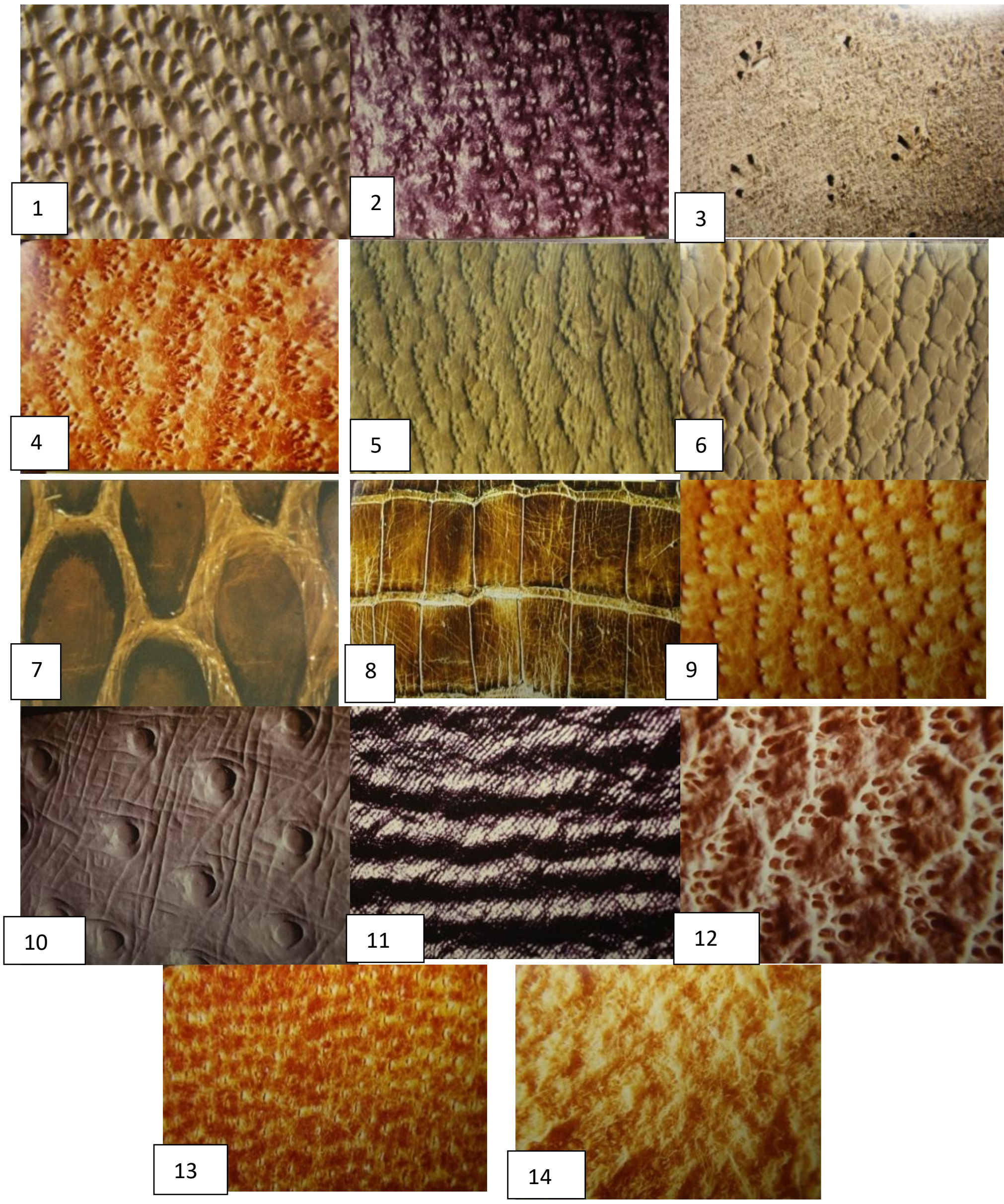
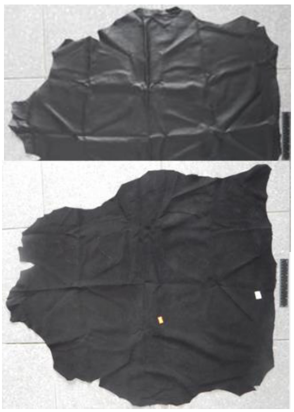


Figure 5. Grain pattern of the different animals' leather: 1-cow; 2-horse; 3-pig; 4-sheep; 5-goat; 6-lamb; 7-snake; 8-alligator; 9-roe deer; 10-ostrich; 11-shark; 12-camel; 13-kangaroo; 14-suede

Obtained results of identification expert examination of leather samples on organoleptic indicators (Table 4) show that the sample 1 is a whole dyed leather. After visual comparison of the sample 1 grain pattern with images shown in Figure 5, we can make a conclusion that this is cow leather. Sample 2 is a whole dyed goat leather. Sample 3 is a whole dyed sheep leather. Sample 1 on the grain side is covered with a silver finishing coat and with flesh side which is dyed in gray. Sample 2 is dyed in black. The grain surface of the sample 3 is painted in black, and the flesh side is finished with a gray coat.

Table 4: Results of identification expert examination of leather on organoleptic indicators

Criteria	Sample 1	Sample 2	Sample 3
Appearance			
Color	Silver	Black	Front side is black, flesh is gray
Grain pattern	Typical for cow leather	Typical for goat leather	Typical for sheep leather
Configuration	Whole leather	Whole leather	Whole leather
Additional processing	The sample on the grain side is finished with a silver coat. Flesh side is dyed in gray and has a homogeneous fibrous structure	The sample is dyed in black on grain and on flesh sides. The grain side is smooth and has a slight sheen	The grain side of the sample is smooth, matte, dyed in black. Flesh side is completely covered with a thin layer of silver coat

The advantage of organoleptic methods is their availability and simplicity, and the disadvantage is the lack of reliability. Therefore, they cannot be the only criteria for identification. So, for higher objectivity we also used physical-chemical and microscopic methods.

To study the origin of different types of finished leather, microscopic research was conducted. The method allows to quickly assess the structure and the origin of leathers.

Animal skin consists of water, proteins, minerals, carbohydrates, fats and fat-like substances. The largest share in the chemical composition of the hide and skin is water (65-75%), the protein content is 28-30%. The main protein of the dermis is collagen, which content range in 93-97% of their total amount of the proteins in the hide. Others include proteins such as reticulin and elastin, as well as water-soluble globular proteins – albumins, globulins, mucoids, mucins and similar others.

Chemical composition was analyzed by method of FTIR spectroscopy. FTIR spectroscopy relies on the fact that most molecules absorb light in the IR area of the electromagnetic spectrum, converting it into molecular vibration. This absorption characterizes the nature of the chemical bonds that are present in the sample. This absorption is measured using a spectrometer as a function of wavelength (as wavenumbers, typically 4000 – 650 cm^{-1}). The result is an IR spectrum that can be used to identify samples.

The analyze of IR spectra are presented in Figure 6. It is advisable to identify natural leather by the most characteristic bands of collagen, as the dermis structure base. The analysis is based on the presence of the main characteristic oscillations bands.

The most characteristic absorption is found in the frequency ranges 3500–3100 cm^{-1} and 1650–1234 cm^{-1}. The first area corresponds to the valence oscillations of the associated NH_2, NH, and OH^- groups. Peaks at 3343, 3276, 3289 cm^{-1} for samples 1, 2 and 3 accordingly refer to the valence oscillations of N-H. This group takes part in the creation of intramolecular hydrogen bonds. The bands at 2920, 2917 cm^{-1} and 2847, 2848 cm^{-1} for samples accordingly refer to asymmetric and symmetric valence oscillations of CH_2 groups [11].

The absorption bands of Amide I, Amide II, and Amide III are observed in the area of 1634–1200 cm^{-1}. This area is associated with absorption of C=O carbonyl groups and deformation oscillations of NH groups, in particular amino-, imino groups, and guanidine groups. Peaks at 1634, 1631 cm^{-1} refer to the valence oscillations of C÷O + C÷N (amide I). Peaks at 1549, 1547, 1544 cm^{-1} for samples accordingly refer to the valence oscillations of C÷N + deformation oscillations N-H (amide II). Peaks at 1334, 1335 cm^{-1} refers to the valence oscillations of C-N. Peak 1278, 1284 cm^{-1} refers to the deformation oscillations of N-H + valence oscillations C-C + deformation oscillations C=O (amide III).

In polypeptides and proteins, the identification of bands 1000–1400 cm^{-1} is difficult due to the interference of oscillations of the side groups of amino acid residues and skeletal oscillations of the carbon chain. The interval 1200–1030 cm^{-1} combines a set of peaks: the band 1082 cm^{-1} and 1030 cm^{-1} are characteristic of valence groups CN, C−O and C=C, and the peaks at 1200, 1202 cm^{-1} and the band 1128 cm^{-1} indicate pendulum oscillations of NH^{3+} groups. The bands present in the spectra at frequencies below 1030 cm^{-1} characterize the “skeletal” oscillations of polypeptides and include, mainly, non-planar deformation oscillations of C−H groups and, to a lesser extent, non-planar deformation oscillations of NH− groups (Amide V).

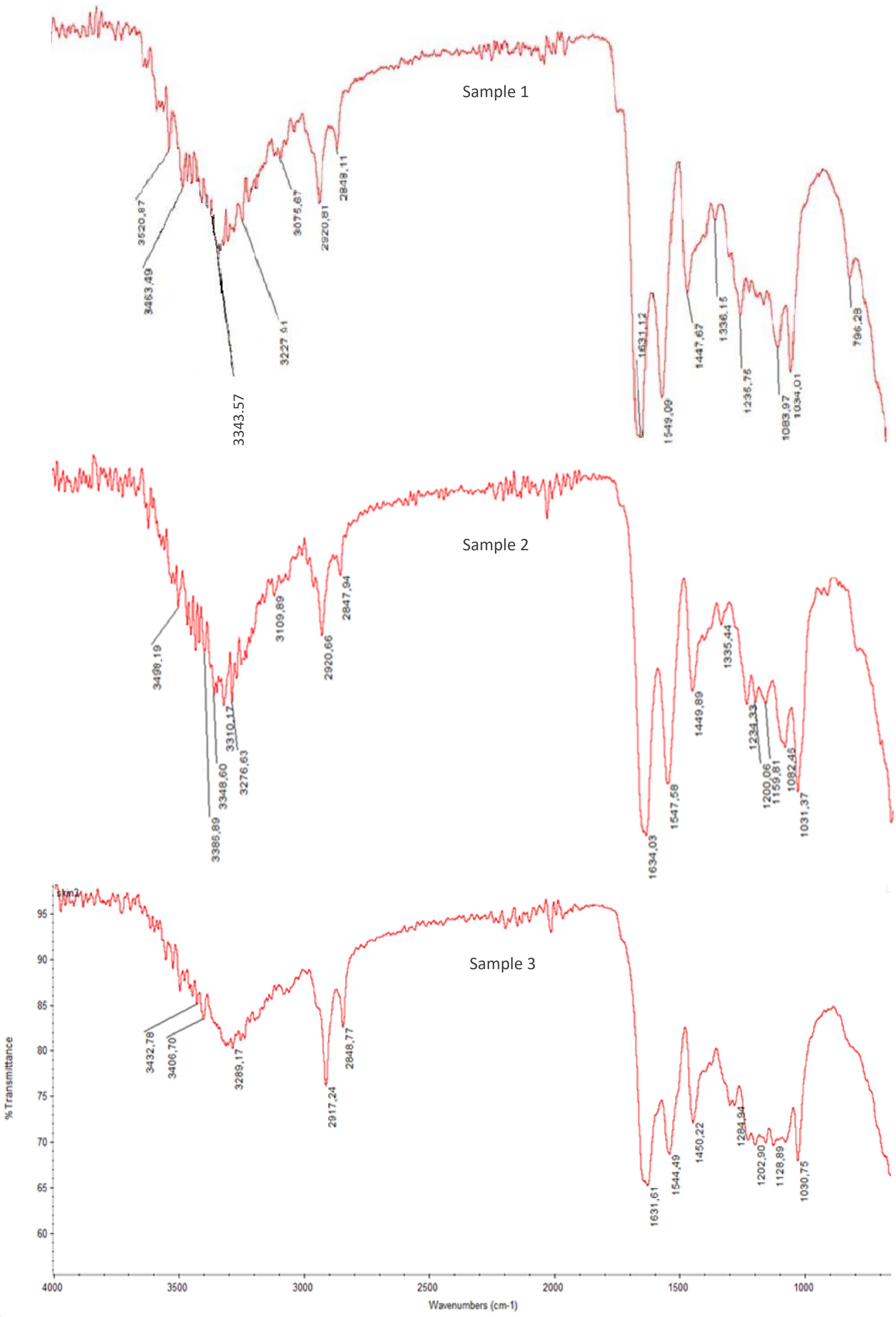


Figure 6. FTIR spectra of the examined leather samples

The main conclusions of FTIR and microscopic analyses regarding the identification of leather samples are presented in Table 5. There are intense absorption bands characteristic of peptide bonds that are present in natural protein (in our case, collagen) in the IR spectrum of the given samples. Structure of all samples has morphological features, characterized for a genuine leather. Taking into account the physical-chemical parameters research results, all test samples are genuine leather.

Table 5: The results of expert examination of leather by FTIR and microscopy

Criteria	Sample		
	1	2	3
Chemical composition	There are intense absorption bands characteristic of peptide bonds that are present in natural protein (collagen, etc.) in the IR spectrum of the given sample (Fig. 6, sample 1).	There are intense absorption bands characteristic of peptide bonds that are present in natural protein (collagen, etc.) in the IR spectrum of the given sample (Fig. 6, sample 2).	There are intense absorption bands characteristic of peptide bonds that are present in natural protein (collagen, etc.) in the IR spectrum of the given sample (Fig. 6, sample 3).
Structure	Has morphological features characteristic of genuine leather.	Has morphological features characteristic of genuine leather.	Has morphological features characteristic of genuine leather.

Based on the results of the identification expert examination, we can make the following conclusions. All samples submitted for examination were in a dry state, soft to the touch. The samples have smooth front surface and flesh surface, which is typical for the natural leather.

The sample 1 is a finished leather in silver color with the flesh side dyed in a similar color. The grain pattern is characteristic for cow leathers.

The front surface of the sample has a relief structure, which is similar in appearance to the surface of leather. The sample on the front side is dried in a silver color.

Examination in the field of view of the microscope revealed that the sample submitted for examination has morphological features characteristic of genuine leather.

There are intense absorption bands characteristic of the peptide bonds that are present in the natural protein (collagen, etc.) in the IR spectrum of the provided sample.

The sample 2 is painted black; has a front and flesh side. The front surface of the sample is smooth, has a slight sheen. Grain pattern is characteristic for goat leather. Microscopic examination revealed that the sample 2 has morphological features characteristic of goat leather.

There are intense absorption bands characteristic of the peptide bonds that are present in the natural protein (collagen, etc.) in the IR spectrum of the sample 2.

The front surface of the sample 3 is smooth, matte, painted black. Flesh side is completely covered with a small layer of silver-thick substance. Pattern is characteristic for sheep leather. Microscopic examination revealed that the sample 3 has morphological features characteristic of sheep leather.

There are intense absorption bands characteristic of the peptide bonds that are present in the natural protein (collagen, etc.) in the IR spectrum of the sample 3.

Therefore, based on the obtained results of identification expert examination and certain characteristics it is possible to determine samples codes according to the UCGFEA. Sample 1 corresponds to the code 4107119000, sample 2 – 4113100000, sample 3 – 4112000000. Appropriate codes according to the HS will be the following: sample 1 – 410711, sample 2 – 411310, sample 3 – 411200.

CONCLUSIONS

Leather and products made of leather such as gloves, garments, bags, footwear, furniture, and many others complete big group of international trade objects of Ukraine as well as many other countries of the World.

Depending on the stages of processing, genuine leather can be untreated with or without hair, tanned or subjected to further processing. These characteristics affect their classification, customs value, duty rate, non-tariff regulation measures and other customs purposes used in international trade operations.

First of all, expert examination allows to determine the correct code of leather and products made of leather according to the Harmonized Commodity Description and Coding System (HS). The Ukrainian Classification of Goods for Foreign Economic Activity (UCGFEA) developed based on the HS and is used in international trade activity in Ukraine.

To conduct identification expert examination of leather, following groups of indicators were determined: labeling conformity assessment; organoleptic; instrumental (physical-chemical) and microscopic. Organoleptic research conducted to determine following leather indicators: color, grain pattern, configuration, additional processing. Instrumental (infrared spectroscopy analysis) – to determine qualitative chemical composition by identification of functional groups. Microscopic – to determine grain pattern and structure identification.

The conducted expert examination results, obtained by using standardized and developed methods, give us opportunity to determine samples codes according to the UCGFEA. Sample 1 corresponds to the code 4107119000, sample 2 – 4113100000, sample 3 – 4112000000. Appropriate codes according to the HS will be the following: sample 1 – 410711, sample 2 – 411310, sample 3 – 411200.

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APPLICATIONS OF ENZYMES AS ECOLOGIC ALTERNATIVES IN THE LEATHER INDUSTRY

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APPLICATIONS OF ENZYMES AS ECOLOGIC ALTERNATIVES IN THE LEATHER INDUSTRY

ABSTRACT. The aim of this paper is to study the applications of enzymes, as ecological alternatives in the leather industry. The research proposes to create ecologic leathers/furs with additional functions (non-toxic, with high-quality of soft, well-degreased surfaces) through the use of original enzyme-based biotechnologies, as new additives for leather/fur processing intended for everyday use. Another application of the enzymes studied in this paper is the development of a novel technology based on enzymes and membrane technique, for purifying wastewaters from the leather industry. We selected and analytically characterized lipases, proteases, and also new enzymes: lysozyme, tryptophan deaminase for use in leather industry. The goal was to obtain leathers/furs for everyday use, with high-performance characteristics, by processing them with enzymes – as new additives that have not been used so far in the leather industry as an alternative to the use of potentially polluting chemical materials. Complex and original technologies of ecological bioprocessing of leather/furs were used. The innovation consists in developing new biotechnologies for leather/fur with enzymatic additives in order to achieve ecologic leathers or fur articles. These biotechnologies provided advanced performances for surface quality: intense and bright colors, soft, well-degreased, and resistant to abrasion and water.

KEY WORDS: original enzyme-based biotechnologies, ecologic leathers or fur articles with additional functions, technology based on enzymes and membrane technique for purifying wastewaters

APLICAȚII ALE ENZIMELOR CA ALTERNATIVE ECOLOGICE ÎN INDUSTRIA DE PIELĂRIE

REZUMAT. Scopul acestei lucrări este de a studia aplicațiile enzimelor, ca alternative ecologice în industria de pielărie. Cercetarea își propune realizarea de piei/blănuri ecologice cu funcții suplimentare (nontoxice, cu suprafețe moi, bine degresate, de calitate superioară) prin utilizarea biotehnologiilor originale pe bază de enzime, ca noi aditivi pentru prelucrarea pieilor/blănurilor de uz zilnic. O altă aplicație a enzimelor studiate în această lucrare este dezvoltarea unei noi tehnologii bazate pe enzime și tehnica membranara, pentru epurarea apelor uzate din industria de pielărie. S-au selectat și caracterizat din punct de vedere analitic lipaze, proteaze, dar și enzime noi: lizozim, triptofan-deaminază pentru utilizare în industria de pielărie. Scopul a fost obtinerea de piei/blănuri de uz zilnic, cu caracteristici performante, prin prelucrarea acestora cu enzime – noi aditivi care nu au fost utilizați până acum în industria de pielărie, ca alternativă la folosirea substanțelor chimice cu potențial poluant. S-au utilizat tehnologii complexe și originale de bioprocesare ecologică a pieilor/blănurilor. Inovația constă în dezvoltarea de noi biotehnologii pentru piele/blană cu aditivi enzimatici în vederea realizării unor articole din piei sau blănuri ecologice. Aceste biotehnologii au condus la performanțe avansate în ceea ce privește calitatea suprafeței: culori intense și strălucitoare, suprafețe moi, bine degresate și rezistente la abraziune și apă.

CUVINTE CHEIE: biotehnologii originale pe bază de enzime, articole din piei sau blănuri ecologice cu funcții suplimentare, tehnologie bazată pe enzime și filtrare prin membrane pentru epurarea apelor uzate

APPLICATIONS DES ENZYMES COMME ALTERNATIVES ÉCOLOGIQUES DANS L'INDUSTRIE DU CUIR

RÉSUMÉ. L'objectif de cet article est d'étudier les applications des enzymes, comme alternatives écologiques dans l'industrie du cuir. La recherche propose de créer des cuirs/fourrures écologiques avec des fonctions supplémentaires (non toxiques, avec des surfaces molles et bien dégraissées, de haute qualité) grâce à l'utilisation de biotechnologies originales à base d'enzymes, comme nouveaux additifs pour le traitement du cuir/fourrure à usage quotidien. Une autre application des enzymes étudiées dans cet article est le développement d'une nouvelle technologie basée sur les enzymes et la technique membranaire, pour purifier les eaux usées de l'industrie du cuir. On a sélectionné et caractérisé analytiquement des lipases, des protéases, mais aussi de nouvelles enzymes : lysozyme, tryptophane désaminase pour utilisation dans l'industrie du cuir. L'objectif est d'obtenir des cuirs/fourrures d'usage courant, aux caractéristiques performantes, en les traitant avec des enzymes – comme de nouveaux additifs qui n'ont pas été utilisés jusqu'à présent dans l'industrie du cuir comme alternative à l'utilisation de matériaux chimiques au potentiel polluant. Des technologies complexes et originales de biotransformation écologique des cuirs/fourrures ont été utilisées. L'innovation consiste à développer de nouvelles biotechnologies pour le cuir/la fourrure avec des additifs enzymatiques afin d'obtenir des articles en cuirs ou en fourrure écologiques. Ces biotechnologies ont apporté des performances avancées pour la qualité de surface : couleurs intenses et lumineuses, surfaces molles, bien dégraissées, résistantes à l'abrasion et à l'eau.

MOTS-CLÉS : biotechnologies originales à base d'enzymes, cuirs ou fourrures écologiques aux fonctions supplémentaires, technologie à base d'enzymes et technique membranaire d'épuration des eaux usées

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INTRODUCTION

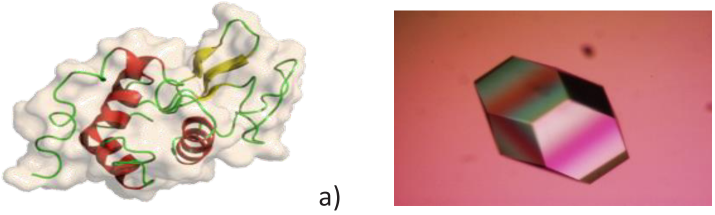
In the leather industry, chemical additives (tannins, retanning products, fatliquors, dyes, finishing products, etc.) play an important role, giving leather/fur properties and characteristics specific to each finite assortment, such as: resistance to mechanical stress (tensile, repeated bending, friction, compression), softness, suppleness, elasticity or stiffness, fullness, touch, color imposed by fashion, impermeability to water, capacity of water vapor absorption, etc.

Enzymes used as additives can have a complex role in the leather industry. Using enzymes to soak high-quality leather leads to more effective rehydration; better opening of the microfibrillar structure of the skin with effects on improving penetration of auxiliary chemicals; degradation and dispersion of fatty substances; more efficient removal of interfibrillar mucopolysaccharides [1-7]. Proteases and lipases are used to soak bovine and sheep skins. The most important effect of enzymes in soaking skins is the solubility of hyaluronic acid and dermatansulphate, basic components of interfibrillar substances from leather [2]. Another important operation in leather processing, in which enzymes are traditionally used, is leather bating. The main function of the bating operation is to eliminate interfibrillar non-collagen substances [3].

The enzymes used are of animal origin – proteases – or of bacterial origin. Bacterial enzymes are cheaper, their sources are more varied, they can degrade elastin and have a greater range of action, pH = 7-9. Bacterial enzymes have a significant action on elastin and may help achieve very soft leather [4].

Leather degreasing is an essential operation for hide processing. Conventional methods of degreasing sheep skins use organic solvents and lead to emissions of volatile organic compounds (VOC) into the atmosphere [2]. The action of the main enzyme types takes place on: hair, non-collagen proteins (proteases); dermis fats, fatty acids from glands in the grain side (lipases); elastin in leather structure and cell membranes (elastase). Lipases are enzymes that can remove fat and fatty acids, particularly those with moderate fat content. The most important effects of using lipolytic enzymes are: the effective removal of fatty substances with positive effects on the uniformity and intensity of leather color; reducing the tenside quantity used; reducing volatile organic compounds (VOC) [1, 4]. Alkaline and acid lipases are used in various stages of the technological process. Alkaline lipases are most often used in decalcifying and bating leather. Lipase activity is increased by the presence of calcium and sodium and is inhibited by the presence of tensides. Acid lipases can be used in degreasing sheep skins preserved by pickling. Research has also shown that degreasing with lipases can lead to an increase in tear resistance of leather, probably because of more uniform distribution of fatty substances in the skin [3]. Another crucial area for the leather industry concerns processing leather waste with proteases for protein separation and recovery of protein component. Lipases hydrolyze triglycerides to mono-and diglycerides, which are more hydrophilic, to fatty acids and glycerin, which are soluble in alkaline medium. In the case of bovine skins, lipases allow full replacement of tensides [1].

In this paper new enzymes, lysozyme and tryptophan deaminase, were selected for use in the leather industry, in purification of wastewaters compared to a bolaform surfactant (sucrose diester). Lysozyme, Fig. 1, also known as muramidase or N-acetylmuramide glycanhydrolase, is a glycoside hydrolase, enzyme that damages bacterial cell walls by catalyzing hydrolysis of 1,4-beta-linkages between N-acetylmuramic acid and N-acetyl-D-glucosamine residues in a peptidoglycan and between N-acetyl-D-glucosamine residues in chitodextrins.



a) b)

Figure 1. Lysozyme: a) structures, b) single crystal [1]

Large amounts of lysozyme can be found in egg white. C-type lysozymes are closely related to alpha-lactalbumin in sequence and structure, making them part of the same family. Lysozyme was the second protein structure and the first enzyme structure to be solved via X-ray diffraction methods, and the first enzyme to be fully sequenced that contains all twenty common amino acids. Since lysozyme is a natural form of protection from pathogens like *Salmonella*, *E. coli*, and *Pseudomonas*, a deficiency due to infant formula feeding can lead to increased incidence of disease [1].

Tryptophan deaminase (Fig. 2) is an enzyme characteristic of groups of organisms belonging to genera *Proteus-Morganella-Providencia*. Tryptophan is one of the 20 standard amino acids, as well as an essential amino acid in the human diet. The distinguishing structural characteristic of tryptophan is that it contains an indole functional group. It is an essential amino acid as defined by its growth effects on rats [1]. Plants and microorganisms commonly synthesize tryptophan from shikimic acid or anthranilate.

a) b)

Figure 2. a), b)-Tryptophan or (2S)-2-amino-3-(1H-indol-3-yl) propanoic acid [1]

This paper also brings a solution to environmental pollution, by introducing membrane technologies (ultrafiltration) for the purification of wastewaters from the leather industry. Leathers and furs were treated with new enzymes and used for medical or everyday use, and the proposed articles are: medical fur belt, leather knee pads.

EXPERIMENTAL

Materials and Methods

The following materials have been used: sucrose diester from SERVA Feinbiochemica GmbH & Co; Pellvit AP from TFL; Esperase, Lipex 100L, NovoCor ABL, NovoCor AX, NovoCor ADL, Greasex from Novozymes; Perizym AFW from Textilchemie Dr. Petry GMBH; Protease produced at ICECHIM; Lysozyme and Tryptophan deaminase from Sigma-Aldrich.

The leathers and furs used come from The Leather and Footwear Research Institute (ICPI).

The experimental techniques used in this work consist in scanning electron microscopy – SEM and UV-VIS spectroscopy tests: a “SEM QUANTA 200” equipment from FEI company, with EDAX coupled. The samples for SEM investigations were prepared by slow evaporation in clean atmosphere at room temperature; a UV-VIS spectrophotometer GBC (model 918).

Ultrafiltration was done with internal cell stirring: Berghoff and with polysulfone membranes from Sigma-Aldrich.

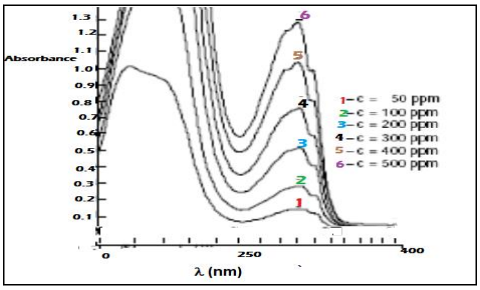
RESULTS AND DISCUSSIONS

The selected lipases and proteases (Pellvit AP, Esperase, Lipex 100L, NovoCor

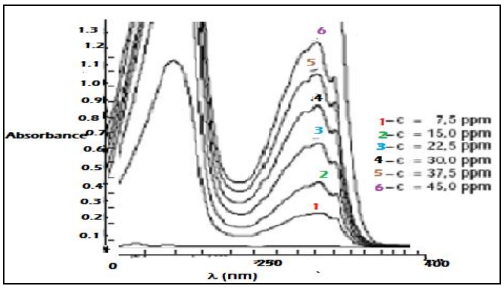
ABL, NovoCor AX, NovoCor ADL, Greasex, Perizym AFW, Protease from ICECHIM) were used to develop a complex and original technology of ecological bioprocessing of leather/furs. In addition, new enzymes used in leather industry (lysozyme, tryptophan deaminase) were selected for their antibacterial effects in order to be used in purification of wastewaters compared to a bolaform surfactant (sucrose diester) by membrane technology.

Characterization of Lysozyme and Tryptophan Deaminase by UV-VIS Spectroscopy

The enzymes were analyzed by UV-VIS spectroscopy. To obtain UV-VIS spectroscopy calibration curves (absorption depending on the concentration of enzyme solutions) we studied a set of two enzymes in a Na_2HPO_4-NaH_2PO_4 buffer solution (pH=7): Tryptophan deaminase (M=204 Da) and Lysozyme (M=14000 Da). This buffer is composed of two salts: monohydrogen phosphate (Na_2HPO_4), which is the basic component and system phosphate dehydrogenase (NaH_2PO_4), the acidic ones. UV-VIS spectra were recorded at room temperature 25°C and showed that certain enzymes are present only in UV absorption and others in visible spectrum. Lysozyme with M = 14 000 Da has four UV absorption peaks at: λ= 209 nm, λ=279 nm, λ =282.5 nm, λ= 292 nm (Fig. 3-a). Absorbance values at 279 nm depend on enzyme's concentration. Tryptophan deaminase has four absorption peaks in UV spectrum at: λ=220 nm, 275 nm, 280 nm and 290 nm (Fig 3-b). Absorbance at λ= 280 nm depends on the concentration of tryptophan deaminase, starting from an initial concentration Ci=45 ppm.



a)



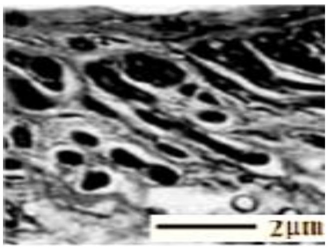
b)

Figure 3. UV spectra of Lysozyme (a) and Tryptophan deaminase (b) in buffer solution, for different concentrations

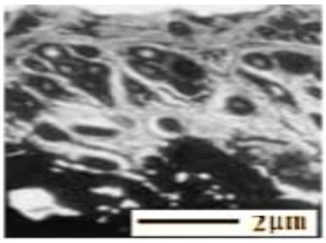
Complex and Original Technologies for Ecological Bioprocessing of Leather/Furs

The processing of hides requires the removal of interfibrillar substances such as soluble proteins, mucopolysaccharides and fatty substances before tanning. Soluble, interfibrillar proteins can be removed with the help of proteases and fats with the lipases or surfactants. Proteases are mainly used in the operations of softening, sanding, and graying of leather (repairing with the help of enzymes). Lipases are used for degreasing the skins by hydrolyzing the fats in the reticular layer and in the structure of the leathers. The use of lipases allows the reduction of pollution with surfactants or organic solvents. The use of lipases for dispersing and removing natural fats helps improve the tanning, retanning and dyeing processes of hides [1]. Lipases hydrolyze triglycerides to mono and diglycerides, which are more hydrophilic, to fatty acids (Fig. 4) and glycerin, which are soluble in an alkaline environment. Ultrafiltration technique was used to remove enzymes (lipases and proteases) from wastewaters from leather and fur industry.

Figure 4 shows SEM images of leathers treated or not with enzymes. Enzymatically hydrolyzed fatty acids from the structure of natural fatty substances in the leather are compared to non-enzymatically treated leather.



a) The structure of non-enzymatically treated leather with a few fatty acids



b) The structure of enzymatically treated leather with many fatty acids, hydrolyzed enzymatically

Figure 4. SEM images of leathers treated or not with enzymes

The dispersion of fats has the effect of a more uniform and brighter coloring of the skins. Lipases also improve the quality of hydrophobized leathers or leathers for car upholstery (by reducing the fogging effect developed on the car windshield by volatile fatty materials). In the case of bovine leathers, lipases allow the complete replacement of surfactants. For sheep leather, which contains up to 40% fatty substances, solvent degreasing can be replaced with an aqueous process in which lipases and surfactants are used. The development of enzyme preparation technologies allowed the creation of enzymes that allow the hydrolysis of non-collagenous proteins in the structure of leather tanned with basic chromium salts. Treating tanned leather with proteolytic enzymes allows better relaxation of the dermis, increased softness, reduced surface defects, and increased usable area by 3-9% (Fig. 5).

Figure 5. Image of leathers, malleable with a larger surface

The use of enzymes allows not only the reduction of polluting chemical materials, but also the reduction of water consumption. The use of surface-active materials requires numerous rinses and high water consumption, while the use of enzymes only requires a simple rinse. Enzymes also allow the elimination of foaming effects, which require the use of silicone-type antifoam materials, allow the reduction of nonyl phenol ethoxylated detergents by 1-3%, which can lead to a 20% reduction of CCO in wastewater.

The main areas of application of enzymatic treatments for fur skins are: when softening furs preserved by drying, washing-degreasing in the float of furs, acid pickling (application of enzymes in the pickling operation), and neutralization of chrome skins. In this paper, the purpose of enzymatic treatments is to replace polluting chemical materials, such as organic solvents, nonylphenolethoxylated detergents (in float degreasing and dry degreasing operations, in solvent), to hydrolyze mucopolysaccharide-type interfibrillar substances, elastin-type proteins, non-hydrolyzable in acid hydrolysis conditions. It is also proposed in this research to replace nonylphenolethoxylated detergents with bolaform sucrose diester, with is non-toxic, ecological.

In the complex technologies for sheep fur bioprocessing the following enzymes were used: Pellvit AP; Esperase, Lipex 100L, NovoCor ABL, NovoCor AX, NovoCor ADL, Greasex; Perizym AFW and protease produced at ICECHIM (Fig. 6).

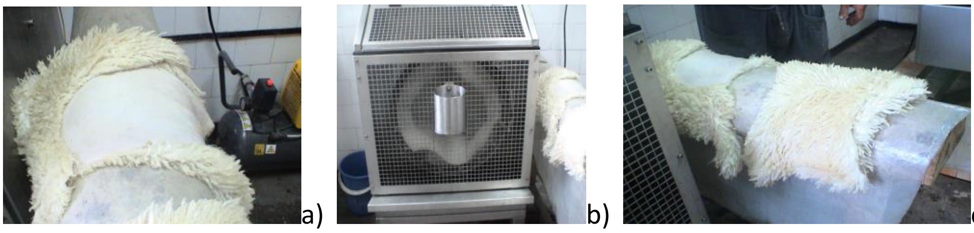


Figure 6. Bioprocessing of furs with enzymes at the micropilot level

Analysis of bioprocessed furs with enzymes (Fig. 7) and of exhausted floats are presented in Fig. 8. Two types of surfactants were used: a classic one, nonylphenol ethoxylate (sample Sc) and a bolaform surfactant, sucrose diester (sample Sb), by comparison. In this research, an original continuous alkaline-enzymatic wool hydrolysis bioprocess was also proposed, where the frequently used alcalase is replaced with lysozyme and the classic surfactant with a bolaform surfactant– sucrose diester. The filtration/separation was done by the membrane technique (ultrafiltration).

Figure 7. Image of fur samples bioprocessed with enzymes: samples P1-P6

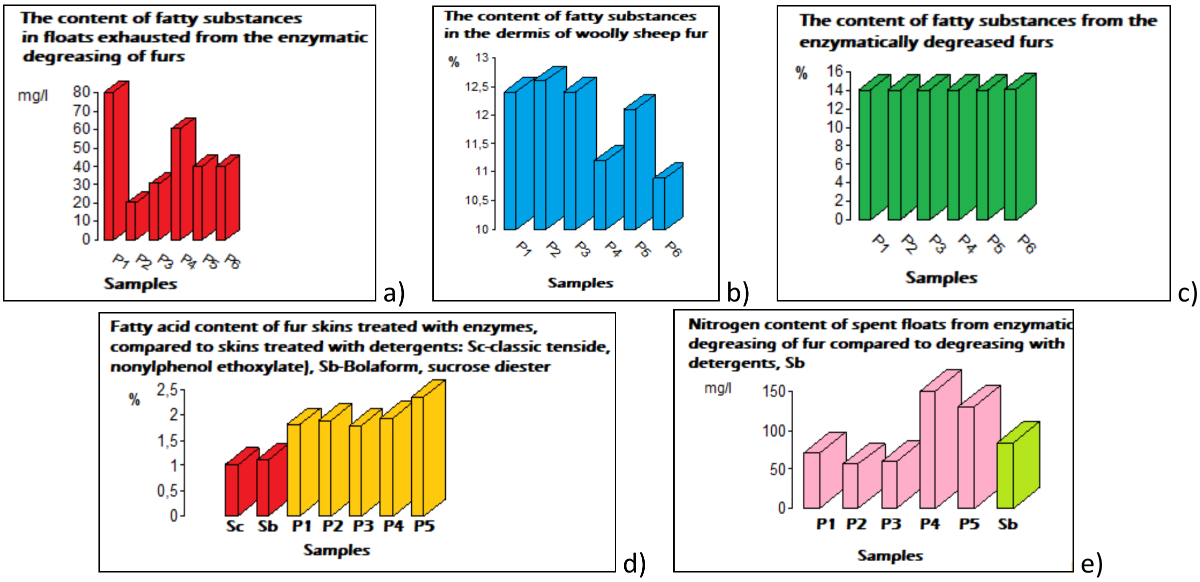


Figure 8. Analysis of bioprocessed furs with enzymes and of exhausted floats

In ultrafiltration an internal cell stirring was used: Berghoff and polysulfone membranes. The new biotechnology proposed for keratin solubilization by alkaline hydrolysis based on lysozyme/sucrose diester/ ultrafiltration is presented in Fig. 9.

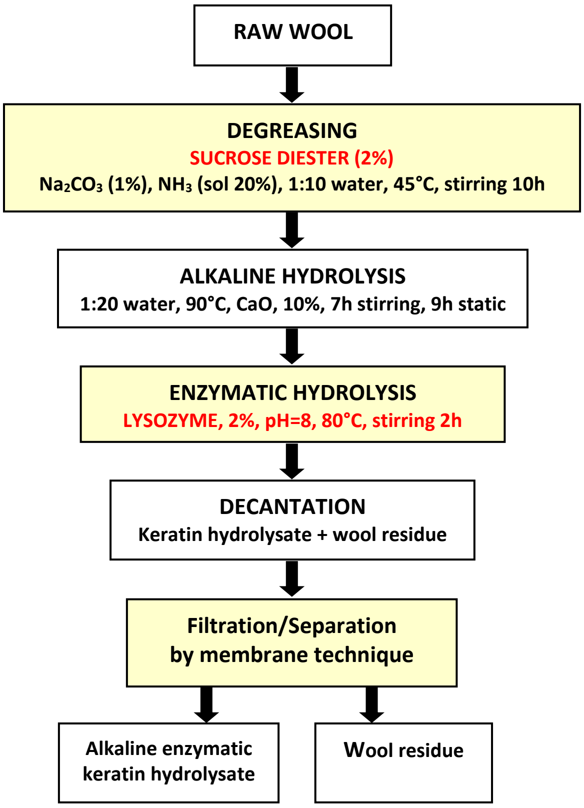


Figure 9. Biotechnology for keratin solubilization by alkaline hydrolysis based on lysozyme/sucrose diester/ultrafiltration

Biotechnology for Purifying Wastewaters from the Leather Industry, Based on Enzymes and Membrane Technique

Membrane techniques (ultrafiltration) can be used to remove lipases and proteases from wastewaters, from the leather industry. If a quantity of c=1% lysozyme or tryptophan deaminase in wastewaters is added before the ultrafiltration process, the glue retentions of enzymes (lipases and proteases) are improved. The addition of enzymes (lysozyme or tryptophan deaminase) improves the purification of wastewaters from the leather industry. Also, the presence of sucrose diester c=1% improves the purification of wastewaters.

There have been UV-VIS spectra of the initial solution and the concentrate, permeate samples,
before and after ultrafiltration. The calibration curves were determined for concentrations of permeate and retention. The retention of each enzyme (lipase or protease) from wastewaters was calculated on the ultrafiltration membrane, using the formula:

$$R = \left(1 - \frac{C_p}{C_c}\right) \cdot 100 \quad (1)$$

Ultrafiltration was done with internal Berghoff cell stirring and the experimental results are given in Table 1.

Table 1: Experimental results

Sample no.	Sample description	Permeate concentration (ppm)	Reject concentration (ppm)	Retention (%)
1	Wastewaters with lipase	95	110	13.63
2	Wastewaters with protease	60	100	40
3	Wastewaters with protease with lysozyme (c=1%)	60	130	53
4	Wastewaters with protease with tryptophan deaminase (c=1%)	30	499	94
5	Wastewaters with protease and lysozyme/ (c=1%)/sucrose diester (c=1%)	29	500	95

CONCLUSIONS

The conducted research has led to the following results:

- Selecting new enzymes with potential applications in the leather industry for their antibacterial effects: lysozyme and tryptophan-deaminase, besides lipases and proteases which are known.
- Selecting enzymes with a potential of stable interaction with collagen and/or keratin, in view of adding new functions to leather and fur.
- Characterization of enzyme solutions and analysis of concentrations by UV-VIS spectroscopy.
- Ultrafiltration technique was used to remove enzymes (lipases and proteases) from wastewaters from leather industry with ~ 13-94 % retention.
- Leathers/furs can be treated with enzymes and used for medical or everyday use and the proposed articles are: medical fur belts and leather knee pads.

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STUDIES REGARDING THE CYTOTOXICITY OF ANTIMICROBIAL GELS FORMULATED WITH NATURAL BIOPOLYMERS

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STUDIES REGARDING THE CYTOTOXICITY OF ANTIMICROBIAL GELS FORMULATED WITH NATURAL BIOPOLYMERS

ABSTRACT. Two antimicrobial gels, formulated with natural biopolymers (collagen and chitosan), limonene, and an imidazole derivative, were subjected to cytotoxicity tests. In these two compositions, the limonene content was 0.4% and 0.67%. The cytotoxicity tests were performed *in vitro*, using MTT methodology, and a standardized human normal cells line, HUVEC type. These cells were exposed to different levels of gel concentrations in the culture media. The final concentration of each gel type in culture media was situated between (0-0.125) µL/mL. The cell viability was determined after 24, 48 and 72h of exposure. The analyses showed that after 24h of exposure, the viability of the cells is greater than 91%, after 48h the viability is greater than 80%; after 72 h of exposure, the viability of the cells is greater than 74%. These values reveal that both selected gels exhibit no cytotoxicity for the normal cell line.
KEY WORDS: antimicrobial gels, biopolymers, cytotoxicity

STUDII PRIVIND CITOTOXICITATEA UNOR GELURI ANTIMICROBIENE FORMULATE CU BIOPOLIMERI NATURALI

REZUMAT. Două tipuri de geluri antimicrobiene, formulate cu biopolimeri naturali (colagen și chitosan), limonene și un derivat de imidazol, s-au testat *in vitro* din punct de vedere al citotoxicității. În aceste două compoziții, conținutul de limonene a fost de 0,4% și 0,67%. Testele de citotoxicitate au fost efectuate folosind metodologia MTT și o linie standardizată de celule umane normale, de tip HUVEC. Celulele normale au fost expuse la diferite niveluri de concentrații de gel, situate între (0-0,125) µL/mL. Viabilitatea celulară a fost determinată după 24, 48 și 72 de ore de expunere. Rezultatele obținute au arătat că după 24 de ore de expunere, viabilitatea celulelor este mai mare de 91%, după 48 de ore viabilitatea este mai mare de 80%; după 72 de ore de expunere, viabilitatea celulelor este mai mare de 74%. Aceste valori arată că niciunul din gelurile selectate nu prezintă citotoxicitate pentru linia celulară normală.
CUVINTE CHEIE: geluri antimicrobiene, biopolimeri, citotoxicitate

ÉTUDES SUR LA CYTOTOXICITÉ DE GELS ANTIMICROBIENS FORMULÉS AVEC DES BIOPOLYMÈRES NATURELS

RÉSUMÉ. Deux types de gels antimicrobiens, formulés avec des biopolymères naturels (collagène et chitosane), du limonène et un dérivé d'imidazole, ont été testés *in vitro* du point de vue de la cytotoxicité. Dans ces deux compositions, la teneur en limonène était de 0,4 % et 0,67%. Des essais de cytotoxicité ont été effectués en utilisant la méthodologie MTT et une lignée cellulaire normale humaine standardisée, HUVEC. Les cellules normales ont été exposées à divers niveaux de concentrations de gel allant de (0-0,125) µL/mL. La viabilité cellulaire a été déterminée après 24, 48 et 72 heures d'exposition. Les résultats obtenus ont montré qu'après 24 heures d'exposition, la viabilité cellulaire est supérieure à 91%, après 48 heures la viabilité est supérieure à 80% ; après 72 heures d'exposition, la viabilité cellulaire est supérieure à 74%. Ces valeurs montrent que les deux gels sélectionnés ne présentent pas de cytotoxicité vis-à-vis de la lignée cellulaire normale.
MOTS CLÉS : gels antimicrobiens, biopolymères, cytotoxicité

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INTRODUCTION

Bioproducts with antimicrobial properties, once developed, must also be evaluated from the point of view of cytotoxicity. The first step for this is represented by the cytotoxicity evaluations performed *in vitro*, on standardized normal cell lines. There are several methods for this, methods that are based both on the evaluation of the cytotoxicity of each individual component and the evaluation of the end bioproduct. Cytotoxicity studies are initiated in the first stage *in vitro*, on normal cells [1-4]. Many natural or synthetic components are used in the development of topical antimicrobial formulations. This category includes citrus essential oils, respectively the predominant compounds of citrus essential oils such as limonene, and some synthetic antibiotics such as imidazole derivatives (i.e., clotrimazole) which cannot be administered intramuscularly or orally due to serious adverse effects [5]. Due to its properties, limonene is intensively used in dermato-cosmetic formulations, but its use in topical formulations is limited due to the fact that it can induce the appearance of epidermal sensitization phenomena. Thus Shah *et al*. [6], in the cytotoxicity studies carried out on standardized cell lines or on primary cell lines initiated from animal tissue, with D-Limonene, reported that at 24 h the cytotoxicity of limonene for the K562 cell line is similar to that of the antitumor reagent doxorubicin (used as a positive control). In this experiment, at concentrations of D-limonene = 4 mM, the viability of K562 cells was 32%, while the viability of cells treated with 0.8μM doxorubicin was 4.1%. If the concentration of D-limonene in the medium increased to 8mM, then cell viability was reduced to 17%. After 48 h of exposure, the best results in terms of cell viability are obtained for doxorubicin. The tests initiated by the same authors on hepatocytes (primary cell line initiated from liver cellular tissue, taken from laboratory animals) showed that for doxorubicin concentrations between (2-32) μM, hepatocyte cell viability is between (100-28)% after 48 h of exposure. If the hepatocyte cells are exposed to concentrations of D-limonene between (2-32)mM, the cell viability obtained is between (100-28)% after 48 h of exposure [6]. The cytotoxicity of limonene can be modified by incorporating it into various lipid formulations such as nanoliposomes or niosomes (niosomes are vesicles made of a nonionic surfactant and cholesterol [7] which are similar to liposomes because they are formations composed of a double lipid start [8]. Another compound often used in topical formulations is clotrimazole, which is a non-biodegradable chemical compound. The half-life of this compound is 60 days, being considered a chemically persistent chemical compound [9]. Regarding clotrimazole, its cytotoxicity can be increased by complexing with ruthenium, gold, or platinum [10]. In the documentary studies carried out by Wang [10], it was reported that the complex combination formed between clotrimazole and ruthenium $[RuCl_2CTZ]_2$ inhibits the proliferation of a parasitic protozoan that causes Chagas disease (i.e., *T. cruzi*) by 90%, in while free clotrimazole does not have this effect [11, 12]. Reducing the cytotoxicity of clotrimazole can also be achieved by microencapsulating it in vegetable oils [13]. Other studies carried out *in vitro* [4, 14-16] showed that the cytotoxicity of some components can be reduced by introducing them into collagen matrices, matrices have the effect of protecting normal cells.

EXPERIMENTAL

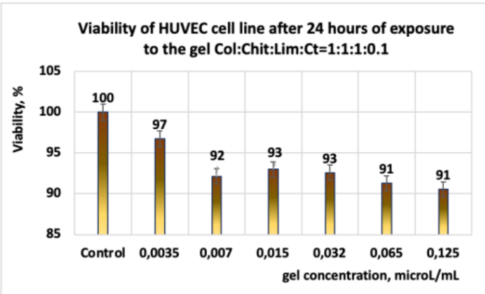
Materials and Methods

Two types of gels formulated with chitosan, collagen (both from Sigma-Aldrich), limonene, and clotrimazole (named Col:Chit:Lim: Ct = 1:1:1:0.1 and Col:Chit:Lim:Ct=1:0:1:0.1, respectively) with antimicrobial activity on microorganisms such as *Staphylococcus aureus*, *Staphylococcus aureus* MRSA, and *Candida albicans*, respectively [17], were tested *in vitro* from the point of view of cytotoxicity in order to further reveal these types of bioproducts, the obtained gels. For this aim, a standardized cell line of human cells named HUVEC ATCC PCS 100-010 was used using the methodology presented by Zaharie *et al*. [1]. The biological determinations were made by exposing the cell line to different concentrations of gels, concentrations that varied between 0.0035 µg/mL and 0.125 µg/mL, for 24h, 48h, and 72h.

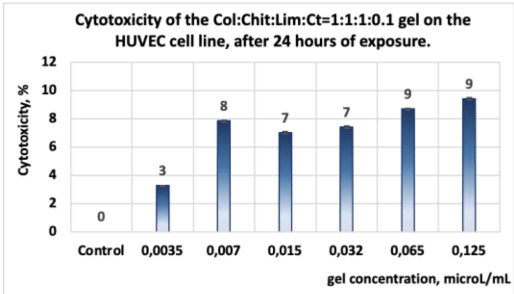
RESULTS AND DISCUSSION

The results obtained in the case of the bioproduct named Col:Chit:Lim:Ct = 1:1:1:0.1 showed the following:

- after 24 hours of exposure to the bioproduct to be tested, for the range of antimicrobial gel concentrations studied, cell viability was between (91-97)% (Figure 1a); the corresponding cytotoxicities obtained for the studied concentration range are below 9% (Figure 1b);



a

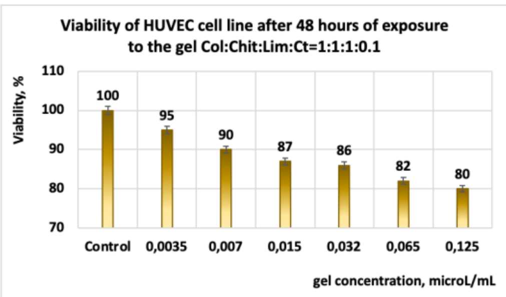


b

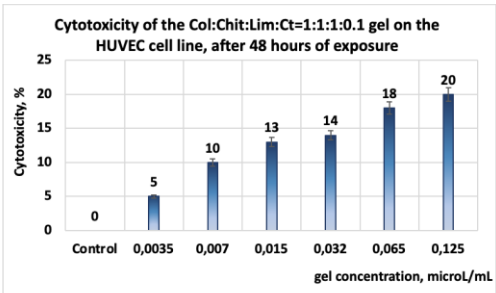
Figure 1. Cytotoxicity studies performed at 24 h on the product Col:Chit:Lim:Ct = 1:1:1:0.1; a) cell viability; b) dead cells (cytotoxicity)

- from a mathematical point of view, the obtained cytotoxicities can be evaluated with a logarithmic function, but in this case, the correlation coefficient is small (Table 1);

- when the normal cells are exposed to the antimicrobial bioproduct for 48 h, their viability ranges between (80-95)% (Figure 2 a, b). The corresponding cytotoxicities were situated between (5-20)% and can be mathematically described by an exponential function (Table 1).



a



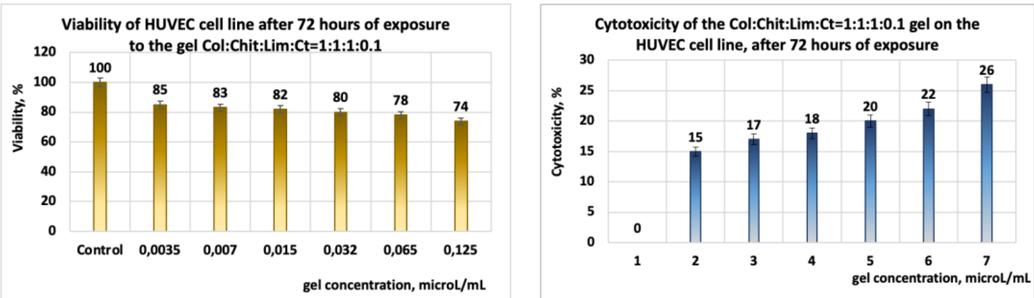
b

Figure 2. Cytotoxicity studies performed at 48h on the product Col:Chit:Lim:Ct = 1:1:1:0.1; a) cell viability; b) dead cells (cytotoxicity)

Table 1: Math function used for cytotoxicity evaluation of antimicrobial gels *in vitro*

Bioproduct Col:Chit:Lim:Ct =1:1:1:0.1		
Time	Math function	Correlation coeficient
24h	$y = 1.2961\ln(x) + 12.15$	$R^2 = 0.6199$
48h	$y = 46.17x^{0.3464}$	$R^2 = 0.915$
72h	$y = 33.51x^{0.1428}$	$R^2 = 0.9706$
Bioproduct Col:Chit:Lim:Ct =1:1:1:0.1		
24h	$y = -90.264x$	$R^2 = 0.9421$
48h	$y=-1.552\ln(x) - 4.1348$	$R^2 = 0.889$
72h	$y = 12.651e^{-14.34x}$	$R^2 = 0.943$

If the exposure of the cell line is prolonged for 72 h, the corresponding cell viability is between (74-85)% (Figure 3a). The corresponding cytotoxicity ranges between (15-26)% (Figure 3b). From a mathematical point of view, the cytotoxicity, in this case, can be characterized by an exponential function (Table 1).



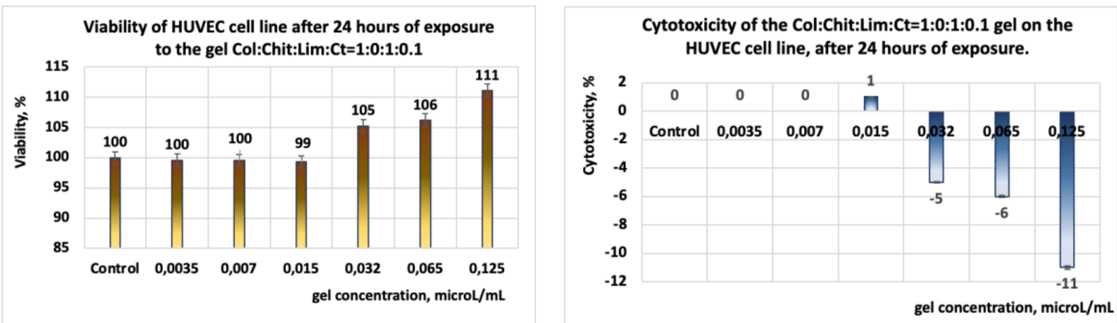
a b

Figure 3. Cytotoxicity studies performed at 72h on the product Col:Chit:Lim:Ct = 1:1:1:0.1; a) cell viability; b) dead cells (cytotoxicity)

The cytotoxicity studies made in the case of the bioproduct named Col:Chit:Lim:Ct = 1:0:1:0.1, showed the following:

- after 24 hours of exposure to the biopreparation to be tested, for the antimicrobial gel concentrations studied, cell viability was between (100-111)% (Figure 4a). The cytotoxicity obtained for the concentration range studied have negative values (Figure 4b), which suggests that this bioproduct favors the proliferation of normal cells and is not cytotoxic;

- when the cells are exposed to the antimicrobial bioproduct for 48h, the cell viability ranged between (95-107)% (Figure 5a). The cytotoxicity obtained after 48 hours of exposure to the antimicrobial gel is below 5% (Figure 5b). If the exposure of the cell line is prolonged up to 72 h, the cell viability was situated between (87-98)% (Figure 6a).



a **b**

Figure 4. Cytotoxicity studies performed at 24h on the product Col:Chit:Lim:Ct = 1:0:1:0.1; a) cell viability; b) dead cells (cytotoxicity)

The corresponding cytotoxicity obtained in this case are below 13% and can be evaluated mathematically with an exponential function (Table 1). Results obtained are in agreement with studies performed by Babeanu *et al*. [4] and Ioan *et al*. [16] which obtain a considerable reduction of cytotoxicity in the case of solid formulation between collagen and clotrimazole, formulation with antimicrobial activity against the same microbial pathogens. Similar results were obtained by Hajidadeh [18] with niosomes based on limonenes, on three types of tumor cells such as HepG2, MCF-7, A549, with the MTT technique. These results showed that at a concentration of 5µM of niosomes with limonenes, in the culture medium, the viability of Hep2G cells is 58%, a much lower value comparison with the viability obtained when the cells were exposed to a culture media in which the D-limonene concentration was 5µM (viability = 95% in this last case). Yip *et al*. [13], in their cytotoxicity studies performed on a normal keratinocyte cells line (HaCaT), exposed to concentrations of 5 µg microcapsules with clotrimazole/mL, reported that after 24 h of exposure, cell viability was 91%, compared to the viability obtained at exposure to solid clotrimazole, when cell viability was 71% after 24 h of exposure.

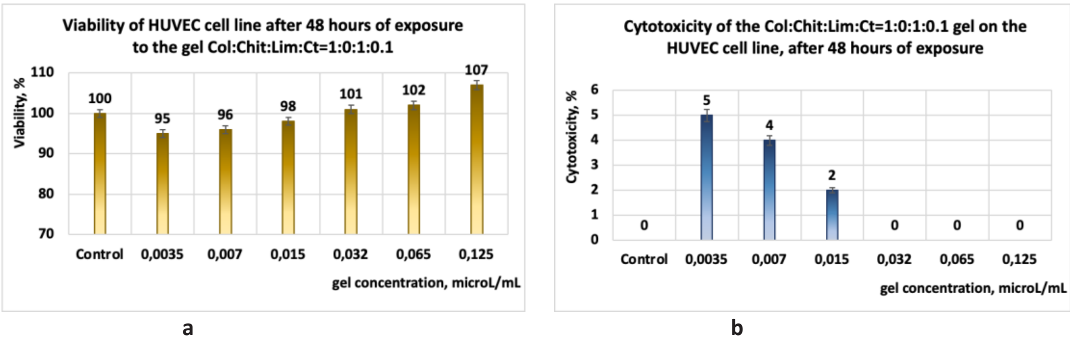


Figure 5. Cytotoxicity studies performed at 48 h on the product Col:Chit:Lim:Ct = 1:0:1:0.1; a) cell viability; b) dead cells (cytotoxicity)

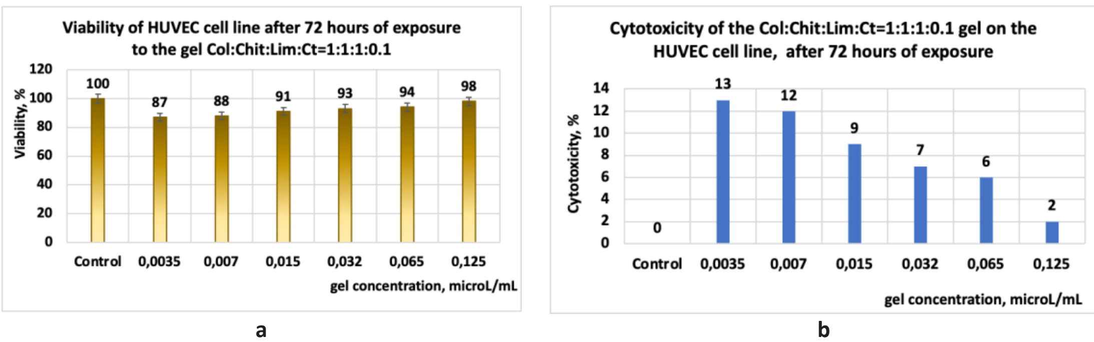


Figure 6. Cytotoxicity studies performed at 72 on the product Col:Chit:Lim:Ct = 1:0:1:0.1; a) cell viability; b) dead cells (cytotoxicity)

The studies carried out *in vitro* by Adinolfi *et al*. [19] on a melanoma cells line and a normal keratinocyte cells line, showed that the exposure of these cells to a concentration of 10µM clotrimazole has the effect of reducing tumoral cell viability up to 50%, while the viability of normal cells had values higher than 85%.

CONCLUSIONS

The cytotoxicity studies made with two gels with antimicrobial properties, formulated with raw materials obtained from by-products from the food industry (collagen obtained from cattle hides, chitosan obtained from crab exoskeletons, citrus peels) and an imidazole derivative (clotrimazole), have demonstrated that the selected antimicrobial biopreparations are not cytotoxic. The cytotoxicity obtained after exposing a normal cell line to the antimicrobial bioproducts for 72h is situated below 25% and is probably due to the more accentuated decomposition of the culture medium as well as the influence of the accumulation in the environment of the products resulting from the cellular metabolism. The cytotoxicity obtained after 72 h of exposure to the antimicrobial bioproducts that do not contain chitosan is 13%, is lower, which suggests a less pronounced decomposition of the culture medium.

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INVESTIGATION OF THE EFFECTIVENESS OF PLANTAR PRESSURE REDISTRIBUTION OF CUSTOMIZED INSOLE STRUCTURE DESIGNED BASED ON PLANTAR STRUCTURE

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INVESTIGATION OF THE EFFECTIVENESS OF PLANTAR PRESSURE REDISTRIBUTION OF CUSTOMIZED INSOLE STRUCTURE DESIGNED BASED ON PLANTAR STRUCTURE

ABSTRACT. Although both fabricated insoles and customized insoles were designed with varied function components, current knowledge approved that customized insoles worked much better. However, how the customized insoles achieved a better performance in term of load re-distribution was still lacking quantitative assessment. The aim was to determine whether the customized insole structure based on a novel customization procedure proposed in this study performs better than the prefabricated insoles in terms of plantar pressure re-distribution efficiency. This study included ten healthy subjects, each wearing four types of insoles accordingly (control insoles; arch support insoles; orthotic insoles; customized insoles), and plantar pressure was collected in a walking state. The custom insoles are made by determining the subject's plantar surface and key plantar points, then following the customization procedure to finish them. The plantar area was divided into eight zones and then a pressure transfer algorithm was used to gain insight into the plantar pressure. Compared to the control group insoles, the arch support of the customized insoles reduced pressure in the hallux region and transferred pressure from the M4-5 to the MH and LH regions, while the metatarsal pad enhanced the pressure transfer in the mid-foot, and the anterior-posterior height difference of the insole plays a role in pressure transfer. Furthermore, the customized insoles performed close to the professional orthotic insoles. Design strategy with accurate insole's component location and reasonably plantar surface matching, our customized insole demonstrated advantages such as a better loading redistribution and significant pressure relieving on the forefoot.
KEY WORDS: customized insole, prefabricated insole, pressure transfer, metatarsal pad, arch support

INVESTIGAREA EFICIENȚEI REDISTRIBUȚIEI PRESIUNII PLANTARE LA UTILIZAREA UNUI BRANȚ CU STRUCTURĂ PERSONALIZATĂ PROIECTAT PE BAZA FORMEI PLANTARE

REZUMAT. Deși atât branțurile fabricate, cât și branțurile personalizate sunt proiectate cu variate componente funcționale, datele actuale confirmă că branțurile personalizate funcționează mult mai bine. Cu toate acestea, lipsește o evaluare cantitativă a modului în care branțurile personalizate ajung la o performanță mai bună în ceea ce privește redistribuirea încărcăturii. Obiectivul lucrării este de a determina dacă structura personalizată a branțului utilizând o nouă procedură de personalizare propusă în acest studiu funcționează mai bine decât branțurile prefabricate în ceea ce privește eficiența redistribuirii presiunii plantare. Acest studiu a inclus zece subiecți sănătoși, fiecare purtând patru tipuri de branțuri (branțuri martor; branțuri de susținere a boltei plantare; branțuri ortetice; branțuri personalizate), iar presiunea plantară a fost măsurată în timpul mersului normal. Branțurile personalizate sunt realizate prin determinarea suprafeței plantare a subiectului și a punctelor plantare cheie, urmând apoi procedura de personalizare pentru a le finisa. Zona plantară a fost împărțită în opt zone și apoi s-a folosit un algoritm de transfer al presiunii pentru a obține o perspectivă asupra presiunii plantare. În comparație cu branțurile martor, suportul plantar al branțurilor personalizate a redus presiunea în regiunea halucelui și a transferat presiunea din regiunile M4-5 în regiunile MH și LH, în timp ce suportul metatarsian a îmbunătățit transferul de presiune în zona mediană a piciorului, iar diferența de înălțime anteroposterioară a branțului joacă un rol în transferul presiunii. În plus, branțurile personalizate au avut o performanță asemănătoare cu branțurile ortetice profesionale. Fiind realizate pe baza unei strategii de proiectare cu localizarea precisă a componentelor branțului și o potrivire rezonabilă pe suprafața plantară, branțul personalizat realizat a demonstrat avantaje precum o redistribuire mai bună a încărcăturii și o reducere semnificativă a presiunii asupra antepiciorului.
CUVINTE CHEIE: branț personalizat, branț prefabricat, transfer de presiune, suport metatarsian, suport plantar

ÉTUDE DE L'EFFICACITÉ DE LA REDISTRIBUTION DE LA PRESSION PLANTAIRE LORS DE L'UTILISATION D'UNE SEMELLE INTÉRIEURE AVEC STRUCTURE PERSONNALISÉE CONÇUE À PARTIR DE LA FORME PLANTAIRE

RÉSUMÉ. Bien que les semelles fabriquées et les semelles personnalisées soient conçues avec divers composants fonctionnels, les données actuelles confirment que les semelles personnalisées fonctionnent bien mieux. Cependant, une évaluation quantitative de la façon dont les semelles personnalisées obtiennent de meilleures performances en termes de redistribution de la charge fait défaut. L'objectif de l'article est de déterminer si la structure de semelle personnalisée utilisant une nouvelle procédure de personnalisation proposée dans cette étude est plus performante que les semelles préfabriquées en termes d'efficacité de redistribution de la pression plantaire. Cette étude a inclus dix sujets en bonne santé, chacun portant quatre types de semelles (semelles de contrôle, semelles de soutien de la voûte plantaire, semelles orthopédiques, semelles personnalisées), et la pression plantaire a été mesurée pendant la marche. Les semelles intérieures personnalisées sont fabriquées en déterminant la surface plantaire du sujet et les points plantaires clés, puis en suivant le processus de personnalisation pour les finir. La région plantaire a été divisée en huit zones, puis un algorithme de transfert de pression a été utilisé pour mieux comprendre la pression plantaire. Par rapport aux semelles de contrôle, le support plantaire des semelles personnalisées a réduit la pression dans la région de l'hallux et a transféré la pression des régions M4-5 vers les régions MH et LH, tandis que le support métatarsien a amélioré le

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transfert de pression dans la zone du milieu du pied, et la différence de hauteur de la semelle antéropostérieure joue un rôle dans le transfert de pression. De plus, les semelles intérieures personnalisées fonctionnaient de la même manière que les semelles orthopédiques professionnelles. Fabriquée à partir d'une stratégie de conception avec un emplacement précis des composants de la semelle intérieure et un ajustement raisonnable sur la surface plantaire, la semelle intérieure personnalisée a démontré des avantages tels qu'une meilleure redistribution de la charge et une réduction significative de la pression de l'avant-pied.
MOTS CLÉS : semelle intérieure personnalisée, semelle intérieure préfabriquée, transfert de pression, support métatarsien, support plantaire

INTRODUCTION

Plantar pressure distribution is a direct index which reflects the foot structure, posture, and function during walking; while wearing insoles is the direct approach to change the foot posture, then to affect its function performance. So, by adjusting and novel designing insole's structure, stiffness and shape, the target of plantar pressure distribution can be achieved. In terms of insole's structure, Tse *et al*. [1] explored the differences in plantar pressure distribution during walking between wedge-only insoles and wedge insoles with arch support, and their results showed that both wedge insoles reduced medial heel and forefoot pressure; further the reduction was more pronounced with supported wedge insoles than unsupported wedge insoles; Farzadi *et al*. [2] used orthotic insoles with medial arch support to reduce the pressure under the hallux and first metatarsal heads, where the load under these two regions shifted to the other regions; meanwhile, Partovifar *et al*. [3] utilized metatarsal pads and medial longitudinal arch support to significantly alleviate the maximum pressure in the heel and metatarsal (MTH) for rheumatoid arthritis patients. In terms of insole's stiffness, Meng *et al*. [4] evaluated the effectiveness of low and high stiffness insoles on plantar pressure during walking and the results showed that the insoles with high stiffness significantly increased plantar pressure of the medial forefoot, but the one with low stiffness increased the loading under mid-foot. In terms of insole's form, Jiang *et al*. [5] designed a plantar pressure redistribution insole based on the characteristic points of plantar pressure redistribution pressure for flat feet adults and they reported that the insole not only reduced the pressure of the second to third metatarsal and medial heel regions, but also it improved gait efficiency.

Most of the existing insoles were prefabricated insoles, which were designed according to the laws of the foot shape from a coherent population. However, foot shape varied through population, a customized design and made insole were considered as the most effective. Miguel Davia-Aracil *et al*. [6] proposed the use of additive manufacturing technology and fused deposition modeling to personalize the material and structure of insoles, while Jandova *et al*. [7] proposed a method that the insole form was 100% matching foot plantar surface to redistribute plantar pressure for patients with flat feet or high arched foot deformities. Korada *et al*. [8] manifested that the customized insoles designed based on plantar form and barefoot plantar pressure could significantly reduce maximum plantar pressure; meanwhile, Stolwijk *et al*. [9] combined the plantar pressure of 204 participants for their individual customized insole design, they observed the effectiveness of pressure redistribution of the customized insole; particularly a significant reduction under the 2^{nd}-5^{th} metatarsal and heel were found. Although positive plantar pressure redistribution was achieved by customized insole, this methodology was still disputed, since 100% of foot matching would ignore the fact that the arch region can change in height during movement, which in turn inhibits the foot's own cushioning capacity.

However, the above studies lack a method to gain insight into the role of insole structure, the pressure transfer algorithm is a method to evaluate the redistribution and transfer of plantar pressure, through this method we can have an insight into the transfer of pressure within foot structure zone, which would indirectly prove the effectiveness of insole structure and form. Bus *et al*. [10] proposed a pressure transfer approach in 2003 and they provided a perspective on changes in foot loading in patients with diabetic neuropathy and foot deformities. Bonanno *et* *al*. [11] executed the pressure transfer analysis for asymptomatic adults wearing insoles with medial heel slopes of 2, 4 and 6 mm; then they disclosed that the insoles with a medial heel slope of 4 mm or 6 mm increased the peak pressure in the medial heel when the subject's foot was in a flat arch or rotated forward position; while plantar pressure in the mid-foot and forefoot did not change. Additionally, Hu *et al*. [12] had an insight into the transfer of plantar pressure in the foot with increasing age in 319 healthy children aged 2-6 years. Nevertheless, the above study assessed the plantar transfer, little literature was focusing on qualitatively assessing pressure transfer caused by customized insoles.

Therefore, the purpose of this study was first to propose a new set of ideas for customized insole design and then to quantitatively evaluate the pressure redistribution efficiency of customized insoles using pressure transfer algorithm. We hypothesize as follows: since the contact area of customized insole was relative larger and the structure design was reasonable and accurate, an optimal pressure distribution, for instance, more anterior and posterior foot pressure transfer, medial and lateral, as well as better forefoot pressure relief would be found.

EXPERIMENTAL

Methods

Participants

Ten participants, two males and eight females (age 22.2±1.0 years, height 166.1±7.8 cm, and weight 59.1±8.7 kg) were recruited. Inclusion criteria were the following: normal BMI; no foot or leg disorders and no history of lower extremity surgery; no abnormal gait pattern. The purpose and procedures of this study were explained to the participants and their formal consent was obtained prior to conducting the test. In addition, the experiment was conducted based on the principles of the Declaration of Helsinki and was approved by the Ethics Committee of Sichuan University.

LuxScan Software Scanning and Modeling of the Foot

LuxScan (v1.1.25, LuxCreo, China) is an application based on the depth-of-field camera of iPhone phones (iPhone X and above) for easy scanning of foot models. The structure-based optical depth camera consists of a point projector, an IR camera and an RGB camera. During the scanning process, the point projector projects special structural patterns onto the object surface; the neural network algorithm in the phone's bionic chip calculates the 3D shape and depth information of the object based on the distortion of the structural light observed by the IR camera on the 3D physical surface, as shown in Figure 1. After obtaining the 3D structure of each subject's foot, the custom design of the insole is performed according to the corresponding algorithm.

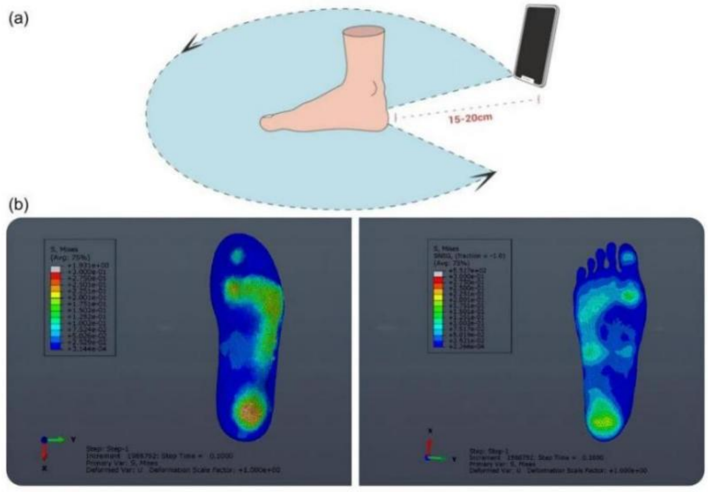

Figure 1. (a) Schematic of 3D scan of the foot (b) 3D plantar structure acquired by LuxScan

Insole Customization Procedure

The design of the key points of the customized insoles in this study followed the following steps, and the specific process is shown in Figure 2.

Insole plane and size design: through the first MTH area (72.5% of foot length), the fifth MTH area (63.5% of foot length) and the heel center key point (18% of foot length).

Arch length: make a medial tangent to the sole of the foot, and determine the arch length of the insole according to the intersection of the first MTH area and the medial heel area point with the medial tangent.

Arch width: determine the arch width according to the vertical distance from the midpoint of the arch (41% of the foot length) to the medial tangent line.

Arch height: divide the outline of the foot into three equal parts (forefoot A, mid-foot B, heel C), excluding the toes, determine the arch height according to the relationship between the arch index x [x=B/(A+B+C)] and the standard navicular bone y (y = -0.3x + 0.2) [13], and adjust the arch height according to the foot type, increasing by 2 mm for high arched feet and decreasing by 2 mm for flat feet.

Metatarsal pad: determine the location of the metatarsal pad according to the intersection area of the line between the first MTH area and the fifth MTH area and the mid-axis of the foot, and adjust the range and height of the metatarsal pad to elevate the transverse arch area of the forefoot according to the sole of the foot.

Heel cup: design the heel bump height of 20mm for both men and women according to the foot survey data [14]. The insole is 3D printed with PLA material, which is a full lattice structure, as shown in Fig. 3, and the insole hardness relies on the lattice for personalized adjustment to the subject.

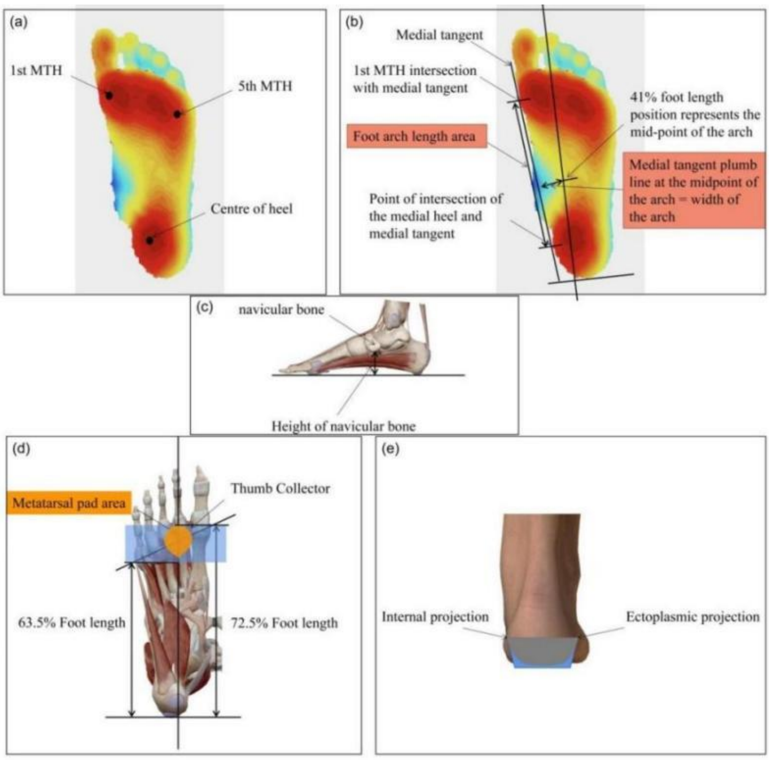

Figure 2. (a) Size design; (b) Arch support area location design; (c) Arch support height design; (d) Metatarsal pad design; (e) Heel cup design

Figure 3. Customized insole structure diagram

Insole for Test

Four insoles with standard test shoes were tested in this study, their details are shown in Table 1.

Table 1: Pictures of shoes and insoles to be tested and their detailed parameters of structure

Insole Type	Front view	Back view	Side view	Arch support	Heel cup	Metatarsal pad	Anterior-posterior height difference	Insole hardness (average)	Remarks
Standard test shoes				×	×	×	×	×	No structure
Control insoles				×	×	×	0mm	46HC	No structure
Arch support insoles				√	√	×	1.8mm	46HC	Bulge
Orthotic Insoles				√	√	×	5.1mm	38HC	Hard support and uneven insole material
Customized insoles				√	√	√	6mm (average)	40HC	Lattice structure

Test Equipment and Test Procedure

The Pedar system (Pedar-X, Novel GmbHgmbh, Germany) was used for the tests. Ramanathan *et al*. [15], Price *et al*. [16] and Hurkmans *et al*. [17] approved good accuracy and repeatability for Pedar insole system. The sole of the foot was divided into eight subdivisions: the hallux (T1), toes 2-5 (T2-5), the 1st metatarsal (MTH1), the 2nd to 3rd metatarsal (MTH2-3), the 4th to 5th metatarsal (MTH4-5), the mid-foot (MF), medial heel (MH), and lateral heel (LH).

Participants warmed up for 3 to 5 minutes; then they wore uniform standardized socks and performed the test on a treadmill with sports shoes. Testing order was determined randomly for each participant. The slope is 0 and speed is 5 km/h for men and 4 km/h for women. Each trail lasted 3 minutes, and they repeated three times for each insole. Data was processed at the end of the experiment. At first, mean pressure (kPa) of each trail was calculated; then, the pressure data of the left and right feet were averaged; finally, the pressure percentage (PP, %) in each area was calculated.

Plantar Pressure Transfer Model

According to the plantar pressure partitioning, four regions were defined from the plantar region: the toe region (level 1), the forefoot region (level 2), the mid-foot region (level 3) and the hind region (level 4) (Fig. 4).

Next, the difference between insoles in each zone was calculated by subtracting the previous insole from the latter, the units of the figures are % (Fig. 4A1).

Then, the arrows from positive to negative marked the transfer of pressure within the level, which means that the loss of pressure in the positive region was transferred to the negative region. The final value of the pressure after the transfer was exhibited in the underlined red color (Fig. 4B1).

After that, the correlation of pressure transfer between adjacent levels was considered and the inter-level and pressure transfer was marked. When an area was faced with a pressure transfer situation to multiple areas, the inside-out and proximity principles need to be considered (Fig. 4C1). Finally, with the longitudinal arch, it was also possible for pressure transfer to occur across the levels (Fig. 4D1).

RESULT

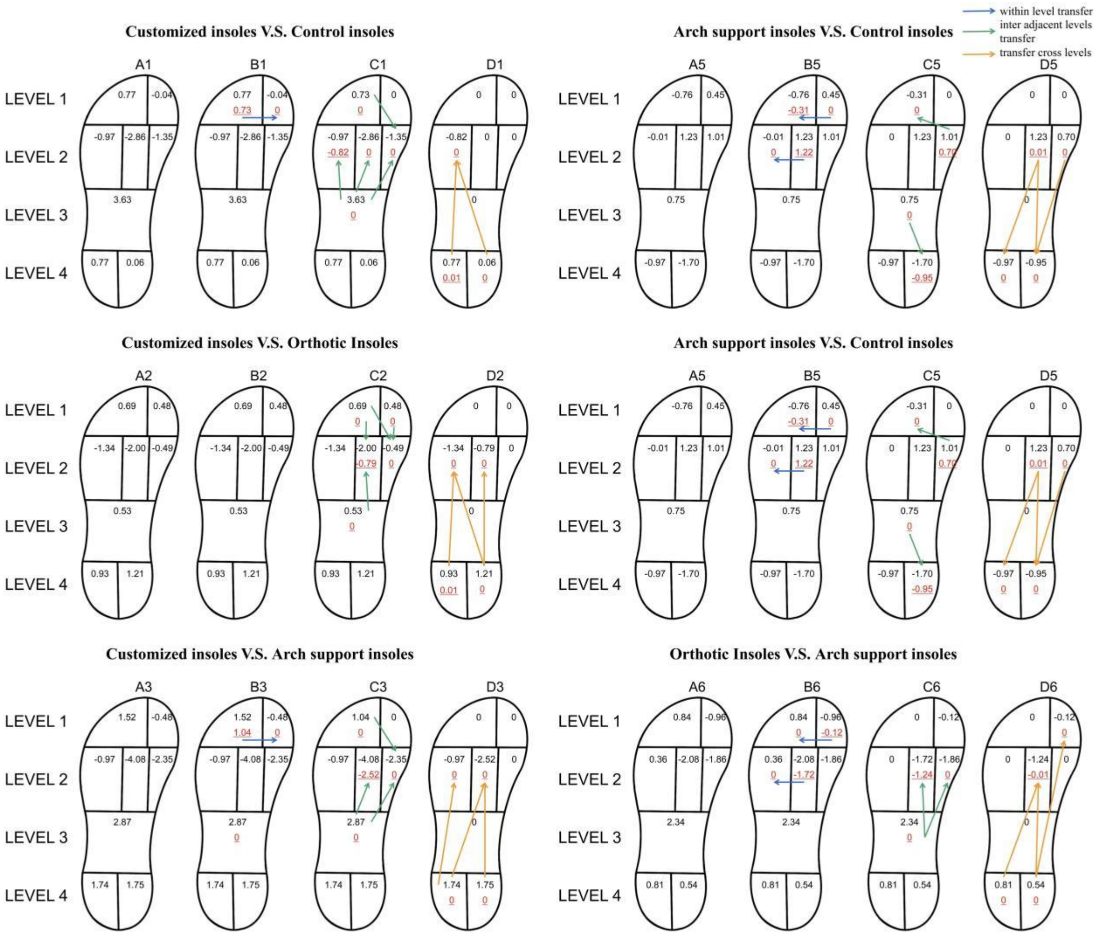

Figure 4. Pressure percentage transfer between insoles (A: PP difference between insoles; B: PP shift within a level l; C: PP transfer between adjacent levels; D: PP transference across levels)

According to Figure 4 we could directly observe what the different insole structures did. The role played by the customized insole in relation to the control group: the arch support adjusted the foot posture and reduced pressure in the T1 region; the metatarsal pad enhanced the transfer of pressure in the mid-foot; the arch support acted as a bridge to transfer pressure from the forefoot region to the mid-foot region on the one hand, and to transfer pressure from M4-5 to the MH and LH regions on the other. Similar results had been found in arch support insoles and orthotic insoles.

In contrast to the arch support insole, the customized insole's metatarsal pad further enhanced the pressure transfer from the mid-foot to the M1-3; its arch support and 6.00mm anterior-posterior height difference further adjusted the foot posture and reduced pressure in the T1 region; the metatarsal pad further enhanced the transfer of pressure from the forefoot to the heel.

The metatarsal pad of the customized insole further took over the pressure transfer in the toe area and mid-foot as opposed to the orthotic insole; personalized fit arch support and a greater anterior-posterior height difference further enhanced pressure transfer from the forefoot to the heel. With orthotic insoles and arch support insoles, there were differences in the role played by the structure: the mid-foot torsional support adjusted the foot posture and also took over the pressure transfer from the forefoot; the fore-and-aft height difference further contributed to the concentration from the forefoot and toe area to the heel area.

DISCUSSION

In this study, firstly, a customized insole design method based on the plantar surface was developed and custom-made insoles were made for ten healthy adults; then the ability of pressure redistribution of four insoles were assessed by the plantar pressure measure and pressure transfer algorithm; finally, the insoles was quantified using a pressure transfer algorithm to characterize the plantar pressure transfer, which not only bridged the anterior and posterior region, but also they apparently reduced load under MTH. Overall performance was similar but superior to that of professional orthotic insoles.

The present study served to reduce mid-foot pressure in terms of arch support. Song *et al*. [18] exhibited that the use of insoles with arch support reduced peak pressure and impact forces in the MTH and heel, and that peak forces of ground reaction were also reduced; Cheng *et al*. [19] conducted a crossover experiment with ten patients with flat feet and showed that the arch support structure significantly increased peak ankle dorsiflexion and peak pressure in the medial mid-foot region, while peak pressure in the heel region was significantly reduced; Abu-Faraj *et al*. [20] showed a significant increase in lateral peak pressure on the plantar aspect of the foot and a significant decrease in peak pressure in the toe and heel regions after the application of arch support. Our result is similar with these reports, but the orthotic insole in the present study did not reduce the pressure in the heel region, which may be related to the custom-made insole metatarsal pad and the 6 mm anterior-posterior height difference in the present study. In addition, the length and height of the arch support are also important factors affecting the plantar pressure, which also provides ideas for the improvement of the heel support structure.

Metatarsal pads usually distribute forefoot pressure and reduce pressure in the metatarsal area. Burgess *et al*. [21] indicated that metatarsal pads shift the peak plantar pressure from the medial forefoot to the lateral forefoot while reducing the peak plantar pressure; Baur *et al*. [22] found that that the use of metatarsal pads or cushion pads can achieve a reduction in peak forefoot pressure and that metatarsal pads can lead to an increase in mid-foot pressure; Chen *et al*. [23] demonstrated that the position of the metatarsal pad directly influenced the effect of pressure distribution, with the best results when the center of the toe pad was positioned 20 mm from the anterior point of the second metatarsal head and 6.5 mm or 13 mm from the anterior point of the second metatarsal head, which would lead to foot tissue damage. These reports are consistent with the findings of this paper that when the metatarsal pad is in the correct position, it can redistribute plantar pressure and transfer the pressure load under the metatarsal region to adjacent areas; however, due to insufficient thickness or improperly low position of the metatarsal pad, the pressure under the metatarsal region may not be reduced, and the proper position and size of the toe pad is important for successful pressure relief under the MH, which explains the importance of customized insoles.

In terms of anterior-posterior height difference, it mainly plays the role of pressure transfer. Shim *et al*. [24] showed that anterior-posterior height difference significantly increased plantar pressure in the metatarsal region and mid-foot region, but there was no significant difference in pressure between foot regions; Zhang *et al*. [25] indicated that after elevating the heel height, the peak pressure in the rear foot decreased, while the peak pressure in the forefoot and mid-foot regions did not increase, indicating an improved plantar pressure distribution. These reports and the results of this study are consistent, but the performance of each insole in this study is consistent, because the anterior-posterior height difference of the customized insole in this study is only 6 mm, and the main role is to cooperate with other structures of the insole to play the role of pressure transfer, while playing a secondary role for the redistribution of plantar pressure. And the study by Koenraadt *et al*. [26] indicated that metatarsal pads produce an increase in the width of the forefoot and an increase in the height of the second metatarsal head, which makes the critical part of the sole of the foot mainly affected by other insole structures in this study.

Our above finding approved the hypothesis, where an optimal pressure redistribution anterior and posterior foot pressure transfer, medial and lateral, as well as better forefoot pressure relief were obtained. However, limitations of this study include the small sample size, the study parameters were only analyzed based on plantar pressure, and the structure of the customized insoles was in a combined form that could not be analyzed against other insoles with a single variable. Future studies should use a larger sample size or even invite patients with flat feet or diabetes to perform the test in order to better analyze the effect of the insole structure; in addition, this study was tested in the walking condition, and future studies could try to analyze the pressure distribution of the customized insole in the exercise condition.

CONCLUSION

Overall, the pressure transfer model was used to insight the plantar pressure redistribution and its path of the customized insole which designed following the foot plantar surface, meanwhile reasonable and accurate insole structure design made our approach function similarly but superior to the professional prefabricated orthotic insole. Therefore, our strategy in custom-design insole would be further utilized orthotic or pressure relieving scenario, such as diabetic patients who need the accurate structure design.

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EFFECT OF CUSTOM-DESIGNED INSOLE WITH ARCH AND METATARSAL SUPPORT ON ADJUSTMENT OF THE LOWER LIMB KINEMATICS

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EFFECT OF CUSTOM-DESIGNED INSOLE WITH ARCH AND METATARSAL SUPPORT ON ADJUSTMENT OF THE LOWER LIMB KINEMATICS

ABSTRACT. Insole construction is considered to be related to stability in sports, and custom-made insoles with their scientifically tailored approach and effective construction are considered to be the main means of adjusting posture and reducing risk in sports. We have designed a non-100% full-fit design insole (CDI) with an arch support insole and metatarsal liner to determine whether this insole can be adapted to lower limb kinematics. Eleven healthy volunteers participated in this randomized crossover test in which a motion capture and 3D gait analysis system was used to measure the subjects' lower limb kinematic data while exercising at different slopes and speeds wearing three different insoles: CDI, control Insole (CI) and arch support insoles (ASI). In addition, the experiments introduced the speed and slope factor, analyzed the extent to which speed slope affects joint angle using UNIANOVA, and compared the performance differences between the three insoles in different planes in pairs. The CDI inhibited ankle dorsiflexion and knee flexion in the sagittal plane; in the frontal plane, CDI reduced knee adduction and hip abduction angles. There was a statistically significant difference ($p < 0.05$) between the change in joint angle and the velocity slope. Slope*speed had a greater effect on the ankle and knee joints (%sig>30%). The results show that the CDI has better kinematic adjustability in the ankle and knee joints due to its superior insole design approach. Therefore, wearing the CDI may be an effective way of reducing risks in sports.

KEY WORDS: customization, insole, sports injury, joint angle, gait kinematic features

EFECTUL BRANȚULUI PERSONALIZAT CU SUPORT PLANTAR ȘI METATARSIAN ASUPRA REGLĂRII CINEMATICII MEMBRELOR INFERIOARE

REZUMAT. Construcția branțului este considerată a fi legată de stabilitate în sport, iar branțurile personalizate cu structură adaptată din punct de vedere științific și construcție eficientă sunt considerate a fi principalul mijloc de ajustare a posturii și de reducere a riscului de accidentare în sport. S-a proiectat un branț cu un design aproape complet personalizat (CDI), cu un suport plantar și căptușeală în zona metatarsiană pentru a determina dacă acest branț poate fi adaptat la cinematica membrelor inferioare. Unsprezece voluntari sănătoși au participat la acest test randomizat încrucișat în care s-a folosit un sistem de captare a mișcării și analiză 3D a mersului pentru a măsura datele cinematice ale membrelor inferioare ale subiecților în timp ce aceștia se antrenau la diferite viteze și înclinații ale planului, purtând trei branțuri diferite: branțul CDI, branțul martor (CI) și branțuri cu suport plantar (ASI). În plus, experimentele au introdus factorul înclinație-viteză; s-a analizat măsura în care înclinația și viteza afectează unghiul articulației folosind testul UNIANOVA și s-au comparat diferențele de performanță dintre cele trei branțuri în perechi, în planuri diferite. Branțul CDI a inhibat dorsiflexia gleznei și flexia genunchiului în plan sagital; în plan frontal, branțul CDI a redus unghiurile de aducție a genunchiului și a șoldului. A existat o diferență semnificativă din punct de vedere statistic ($p < 0,05$) între modificarea unghiului articulației și factorul înclinație-viteză. Factorul înclinație-viteză a avut un efect mai mare asupra articulațiilor gleznei și genunchilor (%sig>30%). Rezultatele arată că branțul CDI are o ajustabilitate cinematică mai bună la nivelul articulațiilor gleznei și genunchiului datorită designului său superior. Prin urmare, utilizarea branțului CDI poate fi o modalitate eficientă de reducere a riscului de accidentare în sport.

CUVINTE CHEIE: personalizare, branț, accidentare sportivă, unghiul articulației, caracteristicile cinematice ale mersului

L'EFFET DE LA SEMELLE INTÉRIEURE SUR MESURE AVEC SOUTIEN DE LA VOÛTE PLANTAIRE ET DU MÉTATARSE SUR L'AJUSTEMENT DE LA CINÉMATIQUE DES MEMBRES INFÉRIEURS

RÉSUMÉ. La construction de la semelle intérieure est considérée comme étant liée à la stabilité dans le sport, et les semelles intérieures personnalisées avec une structure scientifiquement adaptée et une construction efficace sont considérées comme le principal moyen d'ajuster la posture et de réduire le risque de blessure dans le sport. Une semelle au design presque entièrement personnalisé (CDI) a été conçue avec soutien de la voûte plantaire et une doublure métatarsienne pour déterminer si cette semelle pouvait être adaptée à la cinématique des membres inférieurs. Onze volontaires en bonne santé ont participé à cet essai croisé randomisé dans lequel un système de capture de mouvement et une analyse de la marche 3D ont été utilisés pour mesurer les données cinématiques des membres inférieurs des sujets pendant qu'ils s'exerçaient à différentes vitesses et inclinaisons du plan, portant trois semelles différentes : la semelle CDI, la semelle de contrôle (CI) et les semelles avec soutien plantaire (ASI). De plus, les expériences ont introduit le facteur inclinaison-vitesse ; on a analysé la mesure dans laquelle l'inclinaison et la vitesse affectent l'angle de l'articulation à l'aide du test UNIANOVA et on a comparé les différences de performances entre les trois semelles par paires dans des plans différents. La semelle CDI a inhibé la dorsiflexion de la cheville et la flexion du genou dans le plan sagittal ; dans le plan frontal, la semelle CDI a réduit les angles d'adduction du genou et de la hanche. Il y avait une différence statistiquement significative ($p < 0,05$) entre le changement d'angle articulaire et le facteur inclinaison-vitesse. Le facteur inclinaison-vitesse a eu un effet plus important sur les articulations de la cheville et du genou (%sig>30%). Les résultats montrent que la semelle intérieure CDI a une meilleure ajustabilité cinématique au niveau des articulations de la cheville et du genou en raison de sa conception supérieure. Par conséquent, l'utilisation de la semelle intérieure CDI peut être un moyen efficace de réduire le risque de blessure dans le sport.

MOTS CLÉS : personnalisation, semelle intérieure, blessure sportive, angle articulaire, cinématique de marche

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INTRODUCTION

Walking is a common activity in daily life, it refers to two main phases: weight-bearing and ground reaction forces transition; in order to walk in a healthy way, our body and joints in the lower limbs require reasonable posture, to accommodate this loading. As the only part of body contact with the ground, abnormal foot posture will directly and negatively affect the ankle, knee and hip joints, as well as the related muscles and ligaments and other tissues [1]. Thereby, effective insole design is a straightforward method to rectify and guarantee the joints of the foot and lower limb.

The varied insole structure plays a specific role in adjusting foot posture. For instance, the metatarsal pad (MP) functions as supporting the transverse arch of the forefoot, balancing the load under each metatarsal area, and then relieving the pain of the forefoot. Meanwhile, the arch support structure (ASS) has the most direct effect on the adjustment of foot posture, a suitable ASS both ensures its space for movement and allows the arch to release its full cushioning potential, while also helping the arch return to a neutral position after changing plantar flexion [2]. Shaw *et al*. [3] share the same view that the addition of an ASS to a lateral wedge minimises ankle eversion change, and also minimises adduction moment reductions. In this way the stability of standing and walking is improved and the body posture is successfully adjusted. In addition, the calcaneal pad and the heel cup height can successfully minimize the resting calcaneal stance position angle in flat-footed patients [4]. Most of the above insoles use prefabricated insoles, which have the advantages of low price and high applicability.

Although prefabricated insoles have a positive effect on foot posture adjustment, custom insoles are more plausible in literature. Rodrigues *et al*. [5] examined the impact of medially supported custom insoles on lower extremity kinematics and they found that insoles systematically decreased the eversion, where a higher rate of eversion would increase the risk of injury. In another study, they showed a less internal rotation of the tibia when using medially, laterally and posteriorly supported insole [6]. Bonifacio (2018) *et al*. [7] indicated that medially wedged insoles were commonly used to reduce post-patellar stress, limit foot reverse and tibial internal rotation, aimed to stabilize the foot and ankle joint in patients with patellofemoral discomfort. Currently, the custom design was 100% matching foot plantar surface and manufactured by 3D printing or CNC technology. Jin *et al*. [8] applied this method to design insoles for patients with arch deformities and they showed that the distribution of pressure on the bottom of the foot had been improved. In comparison to conventional techniques, the way of CAD increases the design flexibility and variability. But drawbacks, such as high cost and long time waiting, precluded large-scale applications [9].

The effect of custom insoles to modify the foot posture could be quantified by kinematics assessment. According to an analysis of joint angles, angular velocities, and moments, we could quantify this process of modification. Although there is a great deal of research to support the benefits of custom insole construction, most of these custom insoles are designed 100% full-fit to sole, which can limit the arch ability to adjust its balance to a certain extent. As the position of the foot arch changes during exercise, a full-fit insole can inhibit the foot's ability to cushion itself, increasing ankle inversion and leading to postural instability and sports injuries. Knowledge of how a non-100% full-fit design insole with arch and metatarsal support affects the lower limb kinematics was lacking.

Therefore, the purpose of this study was to develop a non-100% full-fit design insole with ASS and MP and then assess its influence on the lower limb kinematics. The hypothesis was made: since CDI has an optimal plantar fit and accurate structure, it would contribute a positive effect on reducing the ankle dorsiflexion and plantar flexion angles and knee inversion compared to the prefabricated insole.

EXPERIMENTAL

Methods

Participants

The study recruited 11 volunteers including 4 males and 7 females, the inclusion and exclusion criteria for all participants are as follows.

The inclusion criteria were:

(1) healthy people aged 18-40 years old; (2) regular exercise habits in daily life.

Exclusion criteria were:

(1) unstable joints of the lower limbs such as hip, knee and ankle; (2) patients with lower limb deformities; (3) surgery on the lower limbs within six months; (4) previous neurological disorders; (5) inability to complete all test movements as required.

The participant's morphological details were: Mean age: 29.36±3.38 years; mean height: 165.54±10.34 cm; mean weight: 59±13.68 kg; male mean shoes size: 42 (eur); female mean shoes size: 37 (eur). Participants were informed of all test content and signed a written informed consent before the measure. The whole process followed the ethical principles of the Helsinki Declaration and was approved by the Institutional Review Committee of Sichuan University.

LuxScan Software Scanning and Modeling of the Foot

LuxScan (v1.1.25, LuxCreo, China) is an application based on the depth-of-field camera of iPhone phones (iPhone X and above) for easy scanning of foot models. The structure-based optical depth camera consists of a point projector, an IR camera and an RGB camera. During the scanning process, the point projector projects special structural patterns onto the object surface; the neural network algorithm in the phone's bionic chip calculates the 3D shape and depth information of the object based on the distortion of the structural light observed by the IR camera on the 3D physical surface, as shown in Figure 1. After obtaining the 3D structure of each subject's foot, the custom design of the insole is performed according to the corresponding algorithm.

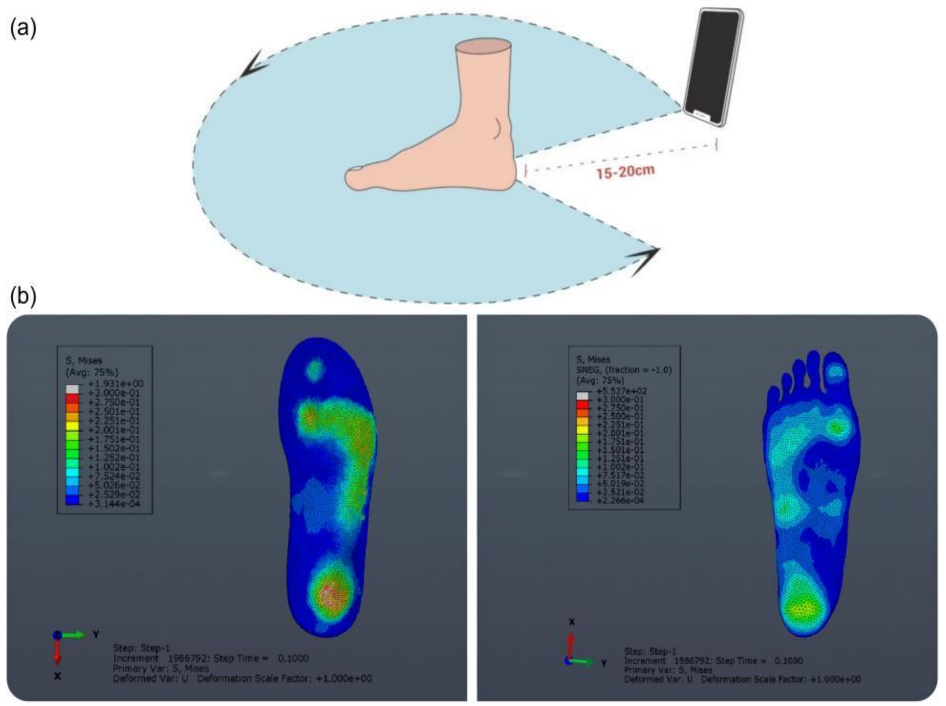

Figure 1. Diagram of LuxScan software operation
(a) Schematic of 3D scan of the foot; (b) 3D plantar structure acquired by LuxScan

Insole Customization Process

The design of the key points for the CDI in this study followed the following steps and the process is shown in Figure 2.

(I) Plane and size design of insole: (Figure 2(a))

The plane of the insole was determined by the first metatarsophalangeal (MTH) joint (72.5% of foot length), the fifth MTH joint (63.5% of foot length) and the key points of the heel (18% of foot length) center.

(II) ASS design of insole: (Figure 2(b))

Arch length: make the medial plantar tangent, and determine the length of the insole arch on the basis of the intersection of the first MTH joint, and the medial heel point with the medial tangent.

Arch width: Using the vertical distance between the medial tangent line and the arch's midpoint (41% of the foot length), calculate the arch width.

Arch height: Excluding the toes, split the foot's shape into three equally sized segments (forefoot A, mid-foot B, and heel C), the height of the arch is determined according to the relationship between the arch index x [x=B/(A+B+C)] and the standard navicular bone y (y=-0.3x+0.2) [10]. The height of the arch is adjusted according to the foot type, the high arch is increased by 2mm, and the flat foot is decreased by 2mm.

(III) MP design of insole: (Figure 2(c))

Determine the position of the MP according to the intersection point between the connecting line of the first MTH joint, the fifth MTH joint and the mid-axis of the foot, adjust the range and height of the MP based on the plantar condition to lift the transverse arch area of the forefoot.

(IV) Heel cup design: (Figure 2(d))

Research on the data from foot type survey in China, the average height of male and female heel bumps is 20mm [11]. The insole is 3D printed with PLA material.

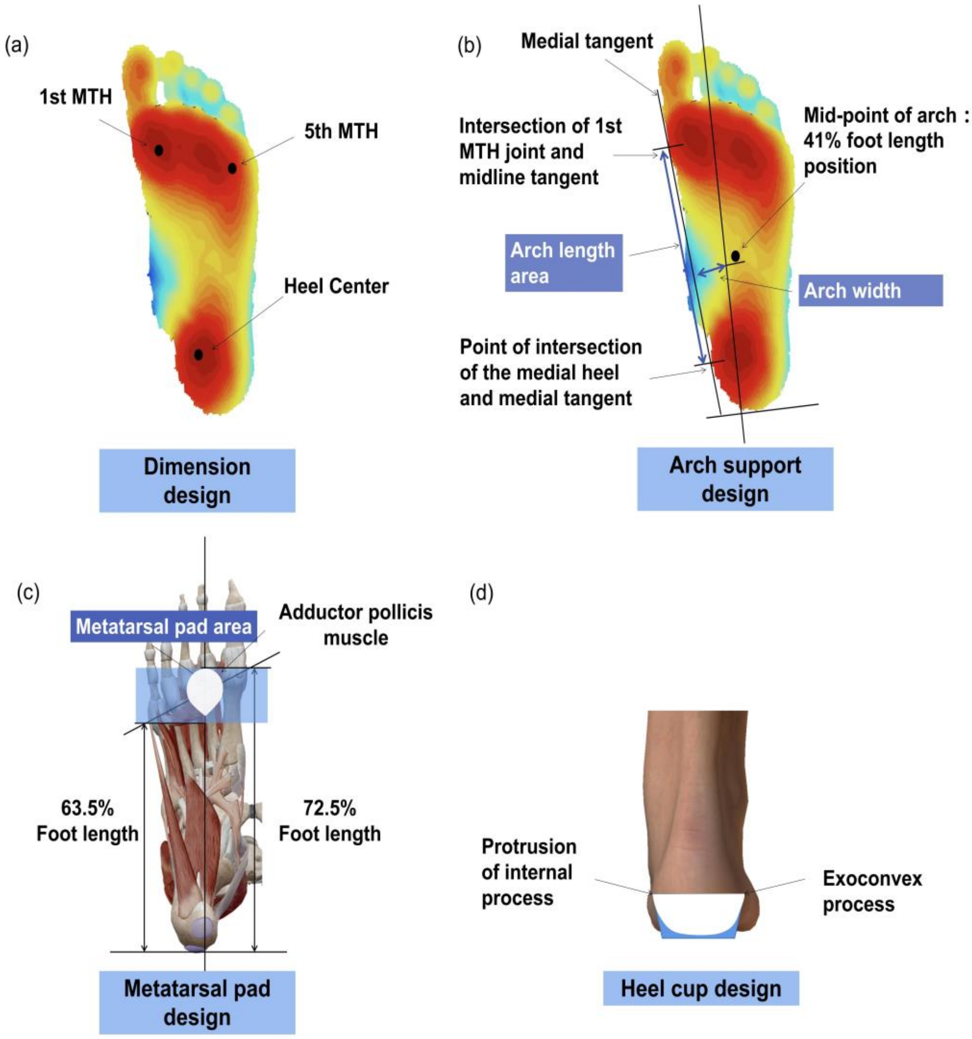

Figure 2. Logic diagram of insole customization
(a) Size design; (b) Arch support area location design; (c) MP design; (d) Heel cup design

Insoles for Measurements

A total of three insoles were tested in this study. The CI had no insole construction and simulated the prefabricated insoles that come with daily purchased shoes. The ASI (with ASS) simulated the more common functional insoles currently available. The three insoles were tested kinematically to investigate whether the CDI we designed positively affected lower limb kinematics.

In this experiment, we used three types of insoles in the same experimental shoe for testing. The hardness was measured using a Shore Digital C hardness tester and the three insoles were tested three times on the forefoot, arch and heel, with the average of the three tests being the final hardness result. For the specific parameters of the insole and the experimental shoes are shown in Table 1.

Table 1: Experimental insole parameters

Types of insoles	Front view	Back view	Profile	AAS	Heel cup	MP	Front rear height difference	Remarks
Custom-designed insole (CDI)				√	√	√	6mm (average)	The insole is a lattice structure
Control Insole (CI)				×	×	×	0mm	Unstructured
Arch support Insole (ASI)				√	√	×	1.8mm	Raised structure of the transverse arch of the foot

Types of insoles	Front view	Back view	Profile	AAS	Heel cup	MP	Front rear height difference	Remarks
Experimental shoe							Front heel height 11mm, heel height 25mm, 13mm height difference between front and rear	Total weight 325g

Types of insoles	Forefoot area hardness	Foot arch area hardness	Heel area hardness	Material	Production Technology
Custom-designed insole (CDI)	42°Shore C	38°Shore C	33°Shore C	polymeric elastic material	Digital Light Procession
Arch support Insole (ASI)	50°Shore C	42°Shore C	46°Shore C	EVA (Ethylene Vinyl Acetate Copolymer), Latex gasket support	Moulding
Types of insoles	48°Shore C	48°Shore C	48°Shore C	EVA	One-piece cutting and gluing

Kinematics Measurement

The Vicon Nexus and Vicon Polygon (Vicon vantage, Vicon, UK) software in the Vicon optical motion capture system was used to collect and process participant kinematic data, maintaining a global coordinate axis system throughout the experiment. Lower extremity reflective points were marked as shown in Figure 3. During the experiment, care should be taken to remove or cover the possible reflective items within the shooting range to avoid the influence of sunlight, lights and other reflective items on the camera identification. After the subjects changed into the experimental clothes as required, 16 reflective points were set on the left and right side of the lower limbs (Figure 4(a)): anterior superior iliac spine, posterior superior iliac spine, thigh, knee, tibia, ankle, toe, heel. The eight Vicon cameras were aligned at 45° angles on all four sides of the 10m square test field, with the treadmill placed in the center of the test field, (Figure 4(b)). To strengthen the experimental effect, we introduced the speed and slope factors, in initial conditions, the male walked at the speed of 5km/h and the female walked at the speed of 4km/h under the condition of 0% slope. Speed simulation: +10% PRS, -10% PRS, grade simulation: +5% grade, +10% grade. A three to five minutes warm-up was first provided to participants, then they were asked to wear three different insoles (CDI, CI, ASI) at random. Participants wearing the insoles were required to perform a randomly assigned ramp speed walking task, maintaining rest intervals of 5 minutes during the test. Eventually, after filtering out incomplete measurements, data of 60 seconds after gait stabilization was selected for acquisition into the database.

Figure 3. Schematic diagram of lower limb model reflective points – front, side, back and detail

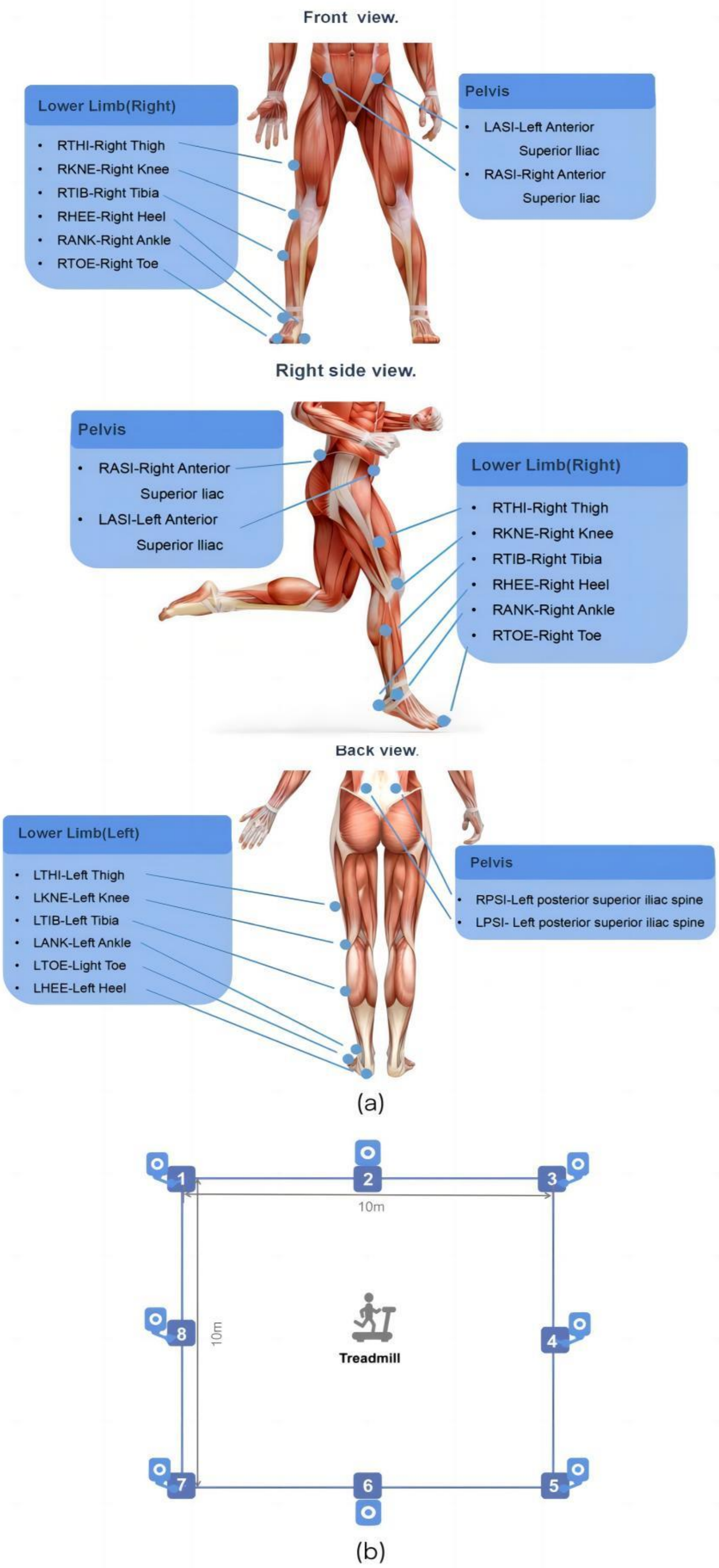

Figure 4. Schematic diagram of insole measurement.
(a) Vicon schematic diagram of reflective points; (b) Schematic diagram of the test site

Data Processing

Using a right-handed coordinate system, each joint is divided into three planes: X (sagittal plane), Y (transverse plane), Z (frontal plane). Since the insole mainly affected motion in X and Z, we chose the sagittal plane and frontal plane for data analysis. Directions were defined for each joint as below: ankles sagittal plane: positive is dorsiflexion and the negative is plantarflexion; ankle's frontal plane: positive is inversion and the negative eversion. The sagittal plane of knee and hip: positive is flexion and the negative is extension; the frontal plane of knee and hip: positive represents adduction and negative represents abduction.

Three complete consecutive strides per phase were used for data analysis, where a complete gait period was defined as from the moment the ipsilateral foot left the ground (the beginning of the gait period) to the moment the second foot left the ground (the end of the gait period) [12]. In terms of the joint angle change evaluation, time-series data were first filtered by a Data filter module of Origin (Origin 8.0, OriginLab, USA) with a 6 Hz cut-off frequency, three complete gait cycles were selected for each case, and then MATLAB (version R2021a, MathWorks, USA) was used to create a 100 points time-normalized gait cycle (GC). The data were normalized for ease of data processing and better observation of data trends, with the algorithm processing logic (Eq. (1)):

$$\alpha=(x-min)/(max-min) \quad (1)$$

where x represents joint angle of each point, min represents valley angle and max represents peak angle.

We first plotted the relationship between the angle and gait cycle in each joint and then UNIANOVA analysis was used at each gait cycle point to explore the influence of slope, speed and slope&speed on the results. Dependent variables were the joint angle, fix factors were insoles, random factors were slope and speed. Within insole's group variances were tested by the Bonferroni model from *post hoc* analysis. Significant differences ($p < 0.05$) from 100 variance analyses were summarized and we used the percentage of significant to indicate the degree of variance. The statistical analyses were performed using SPSS (IBM SPSS Statistics26, SPSS, USA) and α was set as 0.05.

RESULTS

The Insole Lower Limb Kinematic Performance

Similar effects were found for those three insoles in the sagittal plane (Figure 5(a)). During 1–40% GC, CDI and the other two insoles were similar in dorsiflexion and plantarflexion movement, but dorsiflexion of CDI was lower than other groups. In the frontal plane, a higher inverse in the stance phase was found in CDI and ASI during 10% - 50% GC, in contrast with CI; meanwhile, the degree of eversion was lower in CDI and ASI than CI. Variance analyses between CDI and ASI showed that inversion angle of CDI was higher than ASI during single support phase (10-40% GC); further, CDI had minimal ROM (Range of motion) in the frontal plane.

Kinematic parameters of the knee joint in the sagittal plane (Figure 5(b)). CDI and CI showed a similar tendency in flexion angles since the onset of foot contact. During most phases of gait (20-50% GC), the ASI exhibits less knee extension. When heeled off, CDI demonstrated high flexion and continued into the swing phase. In the frontal plane, we observed that CDI was effective in controlling knee adduction mid-to-late stance phase (20-55% GC), where the adductor angle of the knee is noticeably smaller than that of the other two insoles.

In terms of the hip in both the sagittal and frontal plane, variances within three insoles were smaller than the ankle and knee (Figure 5(c)). However, CDI exhibited larger hip extension degrees during early stance of the moment of landing (1-15% GC); while hip extension angle was suppressed during heel off. In the frontal plane, both CDI and ASI showed greater hip abduction angles in the pre-stance phase (1-20% GC), and CDI effectively controlled the degree of hip abduction (21 GC-50% GC) as the gait moved into the mid-stance phase.

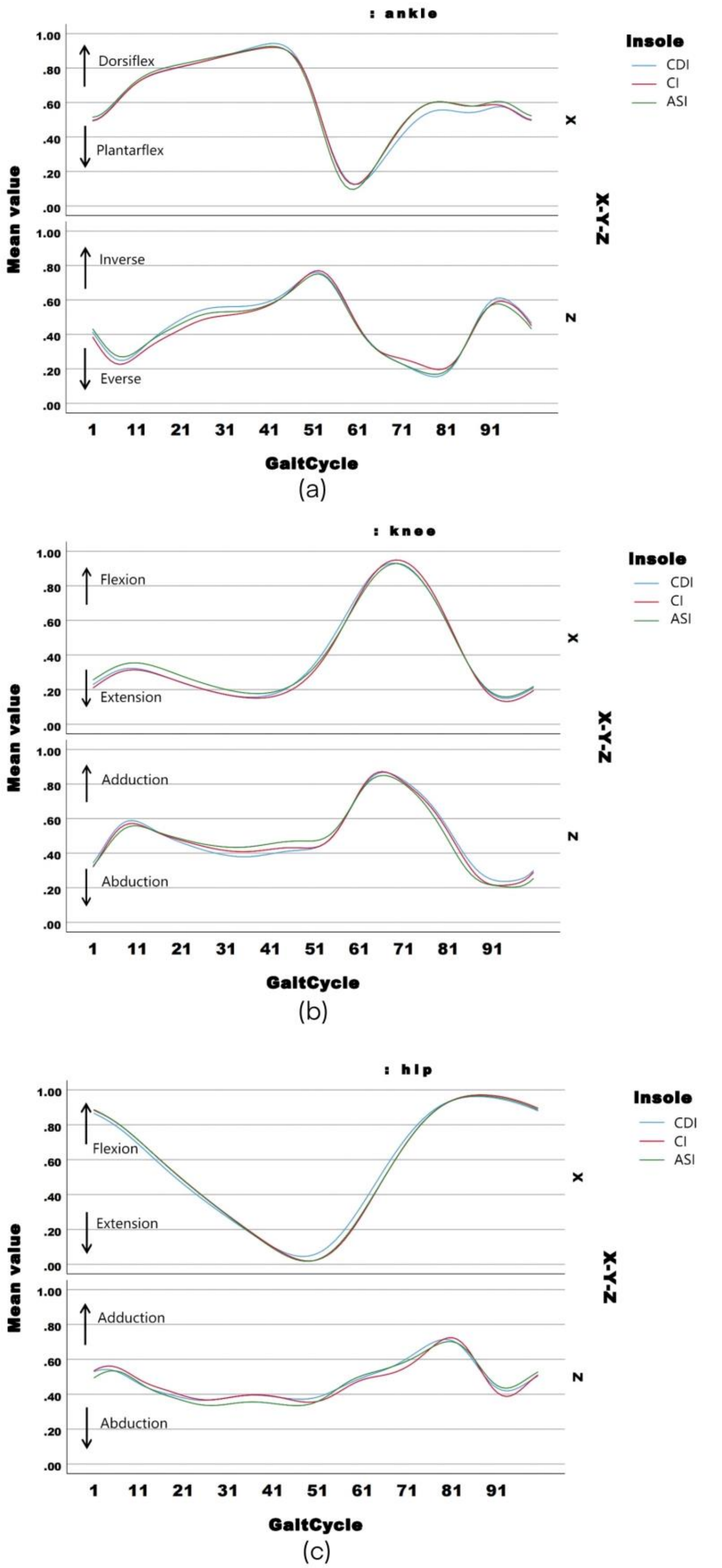

Figure 5. Angle change curve of lower extremity joint.
(a) Sagittal and Frontal plane of ankle joint; (b) Sagittal and Frontal plane of knee joint;
(c) Sagittal and Frontal plane of hip joint

Analysis of Variance

The UNIANOVA results were shown in Table 2. Overall the combined speed*slope factor had a large effect on the lower limb joint kinetic data, particularly for the knee and ankle joints. The effect of slope on sagittal motion of the hip was significant (%sig>30%); speed had a greater effect on sagittal motion of the ankle (%sig>15%), but limited effect on the knee.

The differences between the insoles are mainly between the CDI and the other two insoles. Exceeding 20% differences were found within the two insoles in sagittal plane (CDI V.S CI, CDI V.S ASI), while there were no significant differences in the ASI V.S CI. The results from the frontal plane showed that significant differences in the ankle joint in the CDI V.S CI (%sig>30%) and the knee joint in the CDI V.S ASI.

Table 2: Variance analysis results of lower limb joints

Influence factor		Slope	Speed	Slope*speed
	Ankle	9%	16%	37%
Sagittal plane	Knee	5%	0	37%
	Hip	34%	9%	13%
	Ankle	0%	8%	73%
Frontal plane	Knee	0%	14%	65%
	Hip	0%	19%	39%
Insoles difference		CDI V.S CI	CDI V.S ASI	CI V.S ASI
	Ankle	23%	29%	0
Sagittal plane	Knee	27%	20%	0
	Hip	27%	20%	0
	Ankle	31%	0	0
Frontal plane	Knee	0	15%	2%
	Hip	9%	0	0
	Ankle	31%	0	0
Frontal plane	Knee	0	15%	2%
	Hip	9%	0	0

CDI, Custom-designed insole; CI, Control Insole; ASI, Arch support Insole

DISCUSSION

This study first developed a methodological approach to custom insoles with arch and metatarsal support and then assessed its effectiveness in modifying the lower limb kinematics during walking. Meanwhile, we introduced slope and speed factors to emphasize the effect of the CDI. Our results proved that CDI significantly reduced ankle eversion and dorsiflexion angles during landing; further, we found that CDI not only directly affected ankle motion; but also affected the knee and hip joints, reducing the angle of knee adduction in the frontal plane and effectively controlling the hip abduction angle. Based on the above finding, we postulated that since the CDI with a more precise structural design and better plantar fitting than the other two insoles, a positive effect on lower limb kinematics would be found. In addition, our CDI was combined with 3D scanning, computer modeling [9], and 3D printing technology [13, 14], which made the manufacturing of custom insole more flexible.

Theoretically, reducing ankle valgus contributes to lateral movement of the center of pressure, which reduces medial knee loading and increases body stability. Our findings confirm this, as the CDI effectively reduces the initial valgus angle of the ankle during the stance phase (10-50% GC) and the dorsiflexion angle during the landing phase, and the CDI has a smaller amount of peak ROM during the stance phase than the other two insoles. This is due to the fact that the CDI has structures such as the MP and ASS, which give it a better plantar fit than the control, increasing the contact between the insole and the sole of the foot, thus further improving foot stability during movement. This is consistent with the findings of K.E. Shaw *et al.* [15].

Our finding further approved the significance of ASS in the insole, it functioned by reducing the initial ankle landing angle, maximizing internal, external rotation angle, and angular deflection [16, 17]. Thereby, orthoses with ASS were widely used in clinical practice to limit abnormal hindfoot rotation angles [18]. Similar functions of insole structure were also reported in literature. Hanatsu *et al*. [19] used medial foot support to reduce knee abduction during walking, thereby reducing patellofemoral pain. There had also been investigations into the significant effect during exercise. The ASS could not only mitigate the indirect effects of knee to ankle external rotation, also reduced hip adduction during support by increasing the ROM in the frontal plane [20]. However, it has also been shown that the ASS structure may increase the stress load on the medial and medial longitudinal arches of the ankle. This may be one of the reasons for the discomfort in insole wear found in previous studies [21] and the poor clinical outcomes produced in randomized clinical trials [22].

In addition, our methodology was featured with a 70% fit design for ASS. According to our previous data, 70% fit support allowed the arch to have a flexible amount of movement during exercise, provided better support for walking comfort and foot posture. In the past few years, there have also been studies that confirm this incomplete fitting of ASS is helpful for physical stability and walking comfort. The flexible mesh structure is used instead of a 100% fitted ASS, the elastic activity of the arch provides better static and dynamic balance for walking and arch support [23].

However, most of insoles on the market were designed with a 100% full-fit arch, which leads to excessive arch elevation and knee hyperextension, affecting lower limb joint flexion and extension stability, and may even affect blood flow and have a negative impact on foot ulcers in diabetic foot patients [24]. The full-fit ASS insole resulted in a large initial dorsiflexion angle, which makes it impossible to balance ankle dorsiflexion and knee flexion in the face of a raised slope, thus inhibiting the caching ability of the arch with large joint changes and making it easy to fall and cause bone damage [25, 26]. This makes it easy to fall and causes bone damage.

Other structures such as MP and lateral wedge with ASS also help to stabilize the lower limbs. Gillian *et al*. [27] further concluded that wearing a lateral wedge with ASS reduced knee moment pulses, which had a positive effect on reducing knee pain and improving knee function in patients. Hanatsu *et al*. [28] proposed that the risk of a fall injury is reduced by adjusting the insole height difference and MP design to assist ankle dorsiflexion and eversion angles.

Our study also introduces a speed-slope factor to investigate the effectiveness of the insole structure in dealing with complex environments. Research shows that the effect of slope was directly on the sagittal plane and the speed factor focused on the sagittal plane of the ankle joint; the combined effect of slope and speed was mainly on the frontal plane of the ankle and knee joints. When walking uphill, the body's center of gravity was shifted forward to the front of the foot, and a higher gradient at the same speed had a faster step frequency [29]. Due to the shortened support period during uphill walking compared to horizontal walking, the lower limb joints became more flexed during heel landing and more extended during stance [30]. There is evidence that the use of sloping surfaces has a greater impact on the sagittal joint motion [31]. However, the structure of the insole with 70% arch fitting can effectively regulate the natural dorsiflexion angle of the slope, allowing the ankle joint to remain in a relatively neutral position without excessive dorsiflexion in the face of the slope, thus maintaining walking stability [32].

Speed, and the combined effects of speed and slope, had a positive effect on the lower limb ankle joint in the frontal plane. Although the ankle joint was more affected by speed and slope, the knee joint plays a linking role in the lower limb joints, reasonable knee flexion and ROM could provide stability and relief from osteoarthritic knee pain [33]. Therefore, knee angle control is also particularly important for lower limb stability [34]. Kerrigan *et al*. [36] stated that medially supported insoles of different heights all significantly reduced knee inversion torque and peak angles under uphill walking conditions, theoretically explaining that ASS is biomechanically effective in reducing knee joint loading and adjusting knee posture. Hence, our hypothesis was confirmed.

Several limitations were available in this study: firstly, the sample size of the experiment was small. Future research should increase the sample size, and even invite patients with lower limb diseases to test, to better evaluate the impact of insole structure kinematics. Secondly, there are a few types of insoles involved in the study. In the future, we can increase the types of experimental insoles, compare and analyze them with other single variable insoles, and better analyze the functions of various structures of insoles.

CONCLUSIONS

CDI with a more precise structural design and better plantar fitting than the other two insoles, a positive effect on lower limb kinematics has been found and this structural design of customized insoles (70% fit design for ASS) actually enhances the dynamic stability and comfort of the lower limbs and improves sports safety. Our CDI was combined with 3D scanning, computer modeling, and 3D printing technology, which made the manufacturing of custom insole more flexible. This method would make production of “customized” insoles be actually applied in daily life. According to our methodology, an efficient custom insole would be developed.

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EUROPEAN RESEARCH AREA

COTANCE NEWSLETTERS

Starting with January 2019, the COTANCE Council will issue a monthly ***COTANCE Newsletter*** with the purpose of **promoting an improved image of leather** to relevant decision makers and domestic stakeholders including Members of the European and National Parliament, Governmental authorities, Ministerial officers, Customers of the leather industry, Brands, Retail chains, Relevant NGOs, Designers, etc. The monthly newsletters present topics that tell the truth about a controversial aspect or a fact that is not well known by the general public to bring about a better understanding of leather and the European leather industry, as well as a positive predisposition to legislate in favor of the leather industry. The newsletters are available in seven languages at https://www.euroleather.com/index.php/newsletter, and were also published in the 2019-2022 issues of *Leather and Footwear Journal*. Newsletter 2 of 2023 is given below.

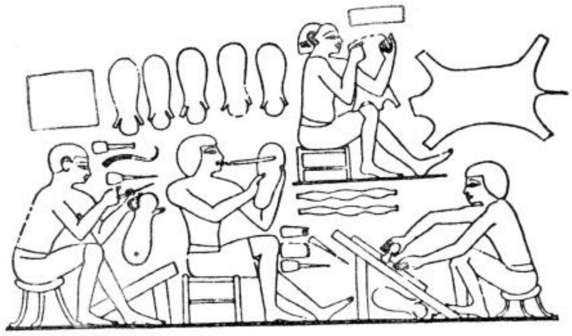
NEWS 2/2023

History and Leather

The archaeological site in Schöningen (Lower Saxony) has already brought back various evidence from the Lower Paleolithic over the years: spears, arrows, sticks. More recently, researchers have focused on some bear remains: they date back 320,000 years and are particularly interesting because they carry engraving traces.

They are evidence, quite rare for that time, that hominids in northern Europe also hunted and skinned bears in order to protect themselves from the cold. Leather features in human history.

Leather has long been in the history of man, and the study of leatherworking methods shows the evolution of traditional methods through to the modern processing methods used today. This is why the discovery in Spain of a hip bone of a large mammal (horse or bison) used for punching hides has aroused some excitement: it dates back 39,600 years and testifies to the method of stitching leather clothing.



Leather is a **noble and versatile material**: there is no civilization that has not made great use of it. But leather is a biological material, as is well known, and therefore certain artifacts have been allowed to reach us only by accident. Certainly, monumental or artistic applications have been preserved and retained, such as Renaissance corami.

Col·lecció A.Colomer Munmany Museu de l'Art de la Pell

But so many everyday objects have been lost. When some of them are found, because the item happened to be in a sufficiently arid or oxygen-deprived environment for it not to come undone, it opens **incredible windows into past eras**. One cannot remain indifferent to the 3,500-year-old Egyptian footwear, or the fourth-century A.D. Viking sandal that ended up who knows how on a Norwegian peak to the two-millennia-old British toy.

Leather manufacturing over the centuries has shaped the territories. Each town had its own production sites and its own typicality, in a pattern of development that harmonized urban design and sense of community. UNIC-Italian Tanneries took charge of the restoration of a tannery in Pompeii, a location as unique as the archaeological excavation that hosts it.

Similar connections between cities and tanneries can be found **all over the world** and cut across eras. Some evidence suggests that from the 18th century onward Belfast grew up around leather laboratories, in Guadalcanal (Spain) an open-air museum has been set up so that recently discovered medieval tanneries can be visited, and in Milan, a city with many waterways that lent itself well to leather-working activities, vestiges of the classical and late antique periods are being studied and celebrated, again with the support of UNIC.

Leather is so much a part of human history that, sometimes, we also come across curious coincidences. It happens, to take one example, that Hermès opens a new atelier in France, where archaeological studies show that hunter-gatherers were already working with animal dermis 13,000 years ago. **But the relationship between leather and history is not only to be read in the past: it is also to the future. The next chapters are yet to be written.**

edited by COTANCE in collaboration with UNIC ITALIAN TANNERIES

News Release from the IULTCS

30th May 2023

Professor Jianzhong Ma Announced as Recipient of the IULTCS Merit Award 2023

It is with great pleasure that IULTCS announces Prof Jianzhong Ma has been chosen as the winner of the prestigious IULTCS Merit Award for Excellence in the Leather Industry. The IULTCS was founded for the purpose of encouraging the technology, chemistry and science of leather on a worldwide basis. It is therefore appropriate that we recognise the achievements of those of stature in our industry who have contributed significantly to our global understanding of the leather industry and its by-products. The IULTCS Merit Award is given biennially by the IULTCS Executive to an individual, whose past or current endeavours have had an extraordinary impact on our industry and provide an example for others to follow. Prof Jianzhong Ma fits this profile perfectly.

Prof Jianzhong Ma has published more than 240 academic articles on prestigious international peer-reviewed journals, such as Advanced Functional Materials, Angewandte Chemie International Edition, Green Chemistry, Carbohydrate Polymers, etc. Seven of them were selected by ESI high citation papers. More than 100 National Invention patents and 7 international invention patents have been authorized. He has published 9 books including 2 monographs, among which “Chemistry of Leather Finishing Materials” has been rated as one of the national top-quality courses. To formulate and revise 7 National and Industrial Standards. Currently, Prof. Jianzhong Ma's H-index (Reported by Web of Science) is 39 and total citation is more than 7056 times.

His extensive list of Practical Achievements is long and varied, with much of his time dedicated to the leather industry, helping to maintain sensible adjustments to testing methods as well as working on new technology for a better future for the leather industry worldwide. He is project leader of 973 pre-research Project of China, 863 Project of China, Key Project of the National Natural Science Foundation of China, General Project of the National Natural Science Foundation of China, National Key Research and Development Plan Project, and over 60 items of University-Enterprise Cooperation Research Projects.

Prof Jianzhong Ma has been actively involved with the development and industrialization of new leather tanning agents, retanning agents, finishing agents, fattening agents, and other chemicals, transferred to more than 40 enterprises at home and abroad.

Functional finishing agents such as cold resistant, hydrophobic and antifouling chemicals have been developed and put into practice. The achievement won the 2nd prize of national technological invention, as well as 2 types of 1st prize of provincial and ministerial technical invention.

He has presided over the national teaching team, national quality courses, national planning textbooks and national experimental teaching demonstration centre etc. He has cultivated more than 100 graduate students, including entrepreneurial talent of the national "Thousand Talents Plan", young top-notch scholar of the "Ten Thousand Talents Plan", and young scholar of Yangtze River Scholars. A large number of outstanding engineering talents have been delivered to the industry. He won the 2nd prize of national teaching achievement.

Notable Conference and workshop presentations from Prof Jianzhong Ma for the leather industry have been:

1. Speaker of 14th National Leather Science and Technology Conference, Plenary presentation, China, 2022

2. Executive Chairman of the Organizing Committee of 11th Asia International Conference of Leather Science and Technology (AICLST), China, 2018

3. Speaker of XXXII Congress of the International Union of Leather Technologists and Chemists Societies (IULTCS), Oral presentation, Turkey, 2013

4. Speaker of XXXI Congress of the International Union of Leather Technologists and Chemists Societies (IULTCS), Oral presentation, Spain, 2011

5. Speaker of 5th Asian International Conference of Leather Science and Technology (AICLST), Oral presentation, Korea, 2002

The award ceremony will be made during the XXXVII IULTCS Congress in Chengdu, China.

Please join us to congratulate Prof Ma for the IULTCS Merit Award.

NATIONAL AND INTERNATIONAL EVENTS

9th WORLD CONGRESS ON RECENT ADVANCES IN NANOTECHNOLOGY (RAN 2024) APRIL, 2024, LONDON, UNITED KINGDOM

The 9th World Congress on Recent Advances in Nanotechnology (RAN 2024) will be delivered in-person in London, United Kingdom and virtually, providing the opportunity of online presentation for the people who can not travel for any reason. Attendees will be able to connect with researchers from across the globe and network in-person or virtually. The registration fee for virtual participation is reduced.

The congress proceedings will be indexed by Scopus, Google Scholar and Semantic Scholar.

RAN is aimed to become one of the leading international annual congresses in the field of nanotechnology. The congress is composed of 2 conferences. While each conference consists of an individual and separate theme, the 2 conferences share considerable overlap, which prompted the organization of this congress.

- **ICNNFC 2024** – 9th International Conference on **Nanomaterials, Nanodevices, Fabrication and Characterization**
- **NDDTE 2024** – 9th International Conference on **Nanomedicine, Drug Delivery, and Tissue Engineering**

This congress will provide excellent opportunities to the scientists, researchers, industrial engineers, and university students to present their research achievements and to develop new collaborations and partnerships with experts in the field.

The **RAN 2024** invites university scholars, scientists and researchers for submission of their papers in the form of extended abstracts, short papers, and full manuscripts. The paper submission deadline for the conferences is **July 21, 2023**, and the early bird registration ends on **September 29, 2023**.

More information: https://rancongress.com/committee/

27th INTERNATIONAL SCIENTIFIC CONFERENCE “COMPETITIVENESS AND INNOVATION IN THE KNOWLEDGE ECONOMY” 22-23 SEPTEMBER, CHISINAU, REPUBLIC OF MOLDOVA

The **Annual International Scientific Conference “Competitiveness and Innovation in the Knowledge Economy”** will bring together many distinguished researchers from all over the world. Participants will find opportunities for presenting new research, exchanging information, and discussing current issues. Although we focus on Europe and all World, all papers from major economics, finance, business fields and IT – theoretical or empirical – are highly encouraged. The conference will be held as a hybrid event which allows the participants to present via Zoom platform and in-person.

Conference Panels:

- Innovative Approaches and New Trends in the Field of Business and Administration
- Fundamental and Applied Economics
- European Integration, Multiculturality and International Economic Relations

- Carpe Scientiam: The Evolution of Social and Human Sciences in the Knowledge Economy
- Innovation and Competitiveness in Accounting and Auditing of Entities
- Informational Technologies and Economic Cybernetics
- Financial Dimensions of the Knowledge Economy
- Current Issues of Upgrading National Law from the Perspective of EU Legal Framework
- Econometrics and Economic Statistics
- Contemporary Libraries: Challenges, Transformations and Premises for Development in the New Socio-Economic Context
- European and Moldovan Procedures on Circular Economy Appropriate for Consumers

Important Dates:

- Deadline for registration and abstract submission – **28 August 2023**
- Deadline for submission of Extended Abstract / Thesis (max. 4 pages) for double peer-review – **20 October 2023**
- Deadline for submission of full Paper (max. 8 pages) for double peer-review – **20 October 2023**

More information: https://conference.ase.md/current-conference/

THE 4TH INTERNATIONAL ELECTRONIC CONFERENCE ON APPLIED SCIENCES (ASEC2023)
27 OCTOBER–10 NOVEMBER 2023, ONLINE

The 4th International Electronic Conference on Applied Sciences (ASEC2023), chaired by Prof. Dr. Nunzio Cennamo, will be held on https://asec2023.sciforum.net/ from 27 October to 10 November 2023.

ASEC2023 offers you the opportunity to participate in this international, scholarly conference without any concerns of expenditures regarding travel—all you need is your computer and access to the Internet. We would like to invite you to “attend” this conference by presenting your latest work. There is **no registration fee**. This online conference will make your presentation accessible to hundreds of researchers worldwide, with the active engagement of the audience in question and answer sessions, in addition to discussion groups that will take place online.

The conference is divided into 10 themes. You are welcome to submit your articles to this conference.

A. Applied Biosciences, Chemistry, Materials Science, and Bioengineering;
B. Computing, Internet of Things, and Artificial Intelligence;
C. Electronics, Optoelectronics and Communications Engineering;
D. Electrical and Energy Sciences, Technology, and Mechanical and Aerospace Engineering;
E. Environmental, Civil Engineering, and Earth Sciences;
F. Applied Nanosciences, Sensors, Instrumentation, and Measurement Science;
G. Food Science and Technology;
H. Applied Dentistry and Oral Sciences;
I. A Student Session;
J. Posters.

Timeline:

- Abstract Submission: **6th July 2023**
- Notification of Acceptance: **27th July 2023**

- Proceedings Paper Submission Deadline: **28th August 2023**
- Conference: **27 October–10 November 2023**

More information: https://asec2023.sciforum.net/

LISBON 2023 CONFERENCES
04-09 DECEMBER, 2023, LISBON, PORTUGAL

The **conferences** will be delivered in-person in **Lisbon, Portugal** and virtually, providing the opportunity of online presentation for the people who can not travel for any reason. Attendees will be able to connect with researchers from across the globe and network in-person or virtually. **The registration fee for virtual participation is reduced.** The **paper submission deadline** for **Lisbon's 2023 conferences** has been **extended to 16 September, 2023.** Accordingly, the **early-bird registration** will end on **30 September, 2023.**

- The 4th International Conference on Civil Engineering Fundamentals and Applications (ICCEFA 2023)
- The 4th International Conference on Environmental Science and Applications (ICESA 2023)
- The 4th International Conference on Fluid Flow and Thermal Science (ICFFTS 2023)
- The 4th International Conference on Advances in Energy Research and Applications (ICAERA 2023)

Important Dates

- Final Extended Paper Submission Deadlines - Sep 16 2023
- Final Extended Notification to Authors - Sep 23 2023
- Final Extended Early-Bird Registration - Sep 30 2023

More information: https://iccefa.com, https://esaconference.com, https://icffts.com, https://icaera.com

NEW TRENDS IN CHEMISTRY RESEARCH
21-22 SEPTEMBER 2023, TIMIȘOARA, ROMANIA

Timisoara's "Coriolan Drăgulescu" Institute of Chemistry will host the 15th edition of the symposium with foreign participation, "New Trends in Chemistry Research", on September 21 and 22, 2023 (Romania).

Through the exchange of scientific knowledge on current developments in Chemistry, particularly those that relate to advanced materials, this international event seeks to establish new collaborations and participation in global projects.

General Topics

- Organic and Polymer Sciences
- Inorganic and Coordination Chemistry
- Applied Biomedical, Electronics, Optoelectronics and Environmental Sciences
- Green chemistry for sustainable development
- Computational chemistry and Chemoinformatics

Deadlines

- 21-07-2023 - Registration form and abstract submission
- 10-08-2023 - Announcement of paper acceptance

More information: https://www.newtrends-timisoara.ro/

THE 27TH INTERNATIONAL SYMPOSIUM ON SEPARATION SCIENCES
24-27 SEPTEMBER 2023, CLUJ-NAPOCA, ROMANIA

ISSS 2023 focuses on fundamentals and applications of separation and detection methods, as well as hyphenated, multidimensional and miniaturized separation techniques applied for analytical, preparative and industrial purposes, including new horizons and challenges in separation sciences.

Topics

1. Gas chromatography
2. Liquid Chromatography
3. Thin-layer chromatography
4. Ion chromatography
5. Supercritical fluid chromatography
6. Capillary electrophoresis
7. Electromigration techniques
8. Hyphenated techniques
9. Multidimensional separations
10. Miniaturized separation techniques
11. Sample preparation techniques
12. Environmental analysis
13. Food analysis
14. Pharmaceutical analysis
15. Clinical analysis
16. Forensic analysis
17. Natural product analysis
18. Industrial analysis
19. Chemometrics
20. Others

Key Dates & Deadlines

• Abstract submission ORAL / POSTER	July 04 / 21, 2023
• Abstract acceptance*	July 21, 2023
• Early bird registration END	July 04, 2023
• Regular registration END	July 31, 2023
• Preliminary Programme	September 01, 2023
• Symposium OPENING	September 24, 2023

More information: https://isss2023.conference.ubbcluj.ro/

THE 22^{ND} INTERNATIONAL CONFERENCE "LIFE SCIENCES FOR SUSTAINABLE DEVELOPMENT" 28^{TH} – 30^{TH} SEPTEMBER 2023, CLUJ-NAPOCA, ROMANIA

The **22^{nd} International Conference "Life Sciences for Sustainable Development"** is organized by the University of Agricultural Sciences and Veterinary Medicine from Cluj-Napoca, will be held on **28^{th} – 30^{th} September 2023.** It includes invited conferences, oral presentations, poster presentations, and round table discussions on recent scientific or technical results, under the European Programme "Horizon 2020" and Romanian Strategy for Research – Technological Development – Innovation in Life Sciences.

The conference programme promotes the dynamic exchange of information, experience, innovative ideas and concepts in the field of agricultural and horticultural sciences, food science and technology, biotechnologies, veterinary medicine, geodesy, as well as other interdisciplinary fields. Participants are invited to present the results of their research within the conference **sessions** as follows:

1. Agriculture
2. Environmental Protection
3. Food Science and Technology
4. Horticulture
5. Economics and Rural Development
6. Animal Science
7. Biotechnology
8. Veterinary Medicine – Fundamental and preclinical sciences
9. Veterinary Medicine – Clinical sciences
10. Geodesy, Forestry and Applied Exact Sciences

The participants have the opportunity to publish for free their presentations (oral or poster) in English, full – paper, after peer-review evaluation, as research article or short communication in Bulletin of UASVM-CN (indexed in prestigious international data bases) – journal series Agriculture, Horticulture, Animal Science-Biotechnology, Veterinary Medicine, Food Science and Technology.

The symposium provides excellent context for scientific exchange, fostering collaboration, and networking, in the meantime with the opportunity of publishing the summaries of the presentations in the "Book of Abstracts" (ISSN 2392 – 6937). The participants registered to our symposium may also publish their presentations, as peer-reviewed full-papers and/or short communications, in the Bulletin of UASVM-CN.

Important Dates

- Deadline for abstract submissions — 27^{th} of August 2023
- Notification of abstract acceptance (as oral or poster presentation) — 3^{rd} of September 2023
- Fee Payment Deadline Before 15^{th} of September 2023

More information: https://symposium.usamvcluj.ro/

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The scientific papers should be presented for publishing in English only. The text of the article should be clear and precise, as short as possible to make it understandable. As a rule, the paper should not exceed fifteen pages, including figures, drawings and tables. The paper should be divided into heads and chapters in a logical sequence. Manuscripts must meet high scientific and technical standards. All manuscripts must be typewritten using MS Office facilities, single spaced on white A4 standard paper (210 x 297 mm) in 11-point Times New Roman (TNR) font.

Paper Format

Title. Title (Centered, 12 pt. TNR font) should be short and informative. It should describe the contents fully but concisely without the use of abbreviations.

Authors. The complete, unabbreviated names should be given (Centered, 10 pt. TNR font), along with the affiliation (institution), city, country and email address (Centered, 9 pt. TNR font). The author to whom the correspondence should be addressed should be indicated, as well as email and full postal address.

Abstract: A short abstract in a single paragraph of no more than 200-250 words must accompany each manuscript (8 pt. TNR font). The abstract should briefly describe the content and results of the paper and should not contain any references.

Keywords. Authors should give 3-5 keywords.

Text

Introduction. Should include the aims of the study and results from previous notable studies.

Materials and Methods. Experimental methods should be described clearly and briefly.

Results and Discussions. This section may be separated into two parts. Unnecessary repetition should be avoided.

Conclusions. The general results of the research are discussed in this section.

Acknowledgements. Should be as short as possible.

References. Must be numbered in the paper, and listed in the order in which they appear.

Diagrams, Figures and Photographs should be constructed so as to be easy to understand and should be named "Figures"; their titles should be given below the Figure itself. The figures should be placed immediately near (after or before) the reference that is being made to them in the text. Figures should be referred to by numbers, and not by the expressions "below" or "above". The number of figures should be kept to minimum (maximum 10 figures per paper).

Tables. Should be numbered consecutively throughout the paper. Their titles must be centered at the top of the tables (12 pt. TNR font). The tables text should be 9 pt. TNR font. Their dimensions should correspond to the format of the Journal page. Tables will hold only the horizontal lines defining the row heading and the final table line. The tables should be placed immediately near (after or before) the reference that is being made to them in the text. Tables should be referred to by numbers, and not by the expressions "below" or "above". The measure units (expressed in International Measuring Systems) must be explicitly presented.

Formulas, Equations and Chemical Reactions should be numbered by Arabic numbers in round brackets, in order of appearance, and should be centered. The literal part of formulas should be in Italics. Formulas should be referred to by Arabic numbers in round brackets.

Nomenclature. Should be adequate and consistent throughout the paper, should conform as much as possible to the rules for Chemistry nomenclature. It is preferable to use the name of the substances instead of the chemical formulas in the text.

References should be numbered consecutively throughout the paper in order of citation in square brackets; the references should list recent literature also. Footnotes are not allowed. If the cited literature is in other language than English, the English translation of the title should be provided, followed by the original language in round brackets. Example: Handbook of Chemical Engineer (in Romanian), vol. 2, Technical Press, Bucharest, 1951, 87.

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