Extract Of Tannins Of The Plant Rumex Confertus, As An Active Component Of Wound Dressing

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ABSTRACT:From the aboveground and root parts of Rumex confertus plants, the method of triple ethanol extraction was used to extract the polyphenol tannic compounds extract - ERc and evaluate its biological activity. It was established that ERc with a single oral administration at a dose of 5000 mg/kg does not cause toxic effects in experimental animals and their death, with prolonged oral administration at a dose of up to 500 mg/kg also does not cause animal death and pathological changes in their organs. At concentrations of 50-100 µg/ml, it inhibited the growth of Bacillus subtilis, Pseudomonas aeruginosa and Staphylococcus aureu. At a concentration above 100 µg/ml, it showed a cytotoxic effect on normal fibroblast cells, and at a concentration of 50 µg/ml ERc significantly stimulated the proliferation of these skin cells, which allowed us to choose this concentration as effective and introduce it into a collagen based RP. As a result, the obtained RP under the name CC-Rc accelerated the time of complete epithelialization of wounds by 1.5 times. At the same time, the commercial NeuSkin-F comparison drug (Eucare Pharmaceuticals (P) Limited, India) consisting of collagen without drug additives showed very low wound healing properties in our studies due to poor adhesion. In this regard, our CC-Rc, which has significantly better congruence and adhesion to the wound, showed an excellent result, did not require additional fixation.

Key words: extract of polyphenolic tannins from Rumex confertus, collagen, wound dressing, proliferative, antimicrobial activity.

1. INTRODUCTION

Natural polymers are widely used in regenerative medicine because of their biocompatibility, biodegradability, biological characteristics and structural similarity with human tissues. Materials of natural origin derived from animals or plants, which usually consist of proteins or polysaccharides, mimic the fibrillar structure of a native extracellular matrix and have similar architectural similarities [Huang S., 2010; Maksimović Z, 2011; Merghoub N, 2011; Norouzi M., 2015.]. However, when the tissue does not have the inherent potential for regeneration, the supply of only protein wound cover to the defective area does not provide tissue repair. In this case, various biologically active substances (BAS) are used as an additional method, such as growth factors, antimicrobial, antioxidant and anti-inflammatory agents, which are introduced into the composition of protein wound cover to stimulate cell proliferation and tissue regeneration.

At present, a large number of various protein wound covers have been created and developed, but all of them have certain shortcomings and do not fully comply with all the requirements for protein wound cover s, and therefore research in this area is relevant and ongoing. In the present work, a new therapeutic composition of protein wound cover based on cattle collagen (cattle) with biologically active substances was created. A dry extract of polyphenolic compounds isolated from the plant Rumex confertus - ERc was used as a biologically active substance. In favor of the choice of this component to create new compositions of protein wound cover, literature data served. Thus, plants belonging to the genus Rumex are used worldwide in traditional medicine to treat various diseases caused by various microorganisms. A number of researchers have found antimicrobial [Orbán-Gyapai O, 2017], cytotoxic [Merghoub N, 2011; Wegiera M, 2012; Norouzi M., 2015.], antioxidant activity [Maksimović Z, 2011] and wound healing activity [Kustova T, 2014; Piesik D, 2011.] extracts of various species of plants of the Rumex family. In this regard, we obtained ERc, studied for cytotoxicity, toxicity, proliferative, antimicrobial activity and introduced it as a biologically active substance in the composition of protein wound cover based on collagen.

2. MATERIALS AND METHODS

Obtaining an extract of polyphenolic compounds ERc isolated from the plant Rumex confertus. The aerial part of the R.confértus plant was collected in July, and the roots and rhizomes in September in the Tashkent region. Thick rhizomes were cut longitudinally, and long roots, stems and leaves were cut across and dried in a drying cabinet ШС – 80-01SPU at a temperature of not more than 500 C. Dried raw materials were crushed to 2.5 5 cm. Tannins were extracted by maceration in laboratory percolators, with a capacity of 3 l at room temperature. Extraction was carried out as follows: an exact sample of the raw materials was placed in separate percolators. At the first contact of the phases, the extractant (ethyl alcohol 50%) was poured to a mirror layer and left for 8 hours, followed by draining of the obtained extract. In the next two extractions, the extraction duration was 4 hours and 6 hours, respectively. The extracts were further concentrated on a RE2000A laboratory rotary evaporator at a temperature of 40 ± 500 C until a thick extract was obtained. The amount of tannins was obtained by treating the thick extract with 96% ethanol, followed by filtration. The resulting product was dried in an oven at a temperature of 40 ± 500 C.

The content of the amount of tannins in raw materials and extracts was determined by the titrimetric method, according to GF XI, part 1 [State Pharmacopoeia of the USSR X1 edition. Vol. 1. Moscow, 1987. p. 286.]. The quantitative content of heavy metals and trace elements was also determined in the obtained samples. To do this, we used an Optima2400DV inductively coupled argon plasma optical emission spectrometer (USA).

Obtaining wound dressings based on collagen isolated from rat tendons. Cattle tendons (cattle) were thoroughly cleaned of muscles, ligaments and other things, crushed to pieces 1-2 mm in size, treated with 0.25% trypsin solution (trypsin crystal. Samson-Med LLC, St. Petersburg, 196158) at 37°C. After trypsinization, it was washed several times in distilled water to remove non-collagenic proteins and was hydrolyzed for 48-72 h in a 0.1: 1 solution of CH_3COOH. The resulting gel was centrifuged at 3000 rpm for 5-7 minutes. Then the collagen mass was filtered through a mesh material to remove large pieces, which were sent for re-hydrolysis.

The study of acute toxicity ERc. Conducted on sexually mature white male rats weighing 150-180 g. The experimental groups were formulated in 6 pieces each. The drug was administered to animals once orally in doses: 100-500-1000-5000 mg/kg. 3-4 hours after the introduction of the components, the animals were given natural and briquette feeds. Observation of animals was carried out for 14 days.

The study of chronic toxicity. The experiments were carried out on white outbred rats. Animals were divided into 4 groups of 6 rats per group. Experimental groups of animals were injected with aqueous solutions of ERc in doses of 50, 100 and 500 mg/kg for a month. A control group of animals was injected with water under similar conditions. All experimental and control animals were in the same conditions and on a normal diet, under daily supervision. After the last injection in all groups of animals from the tail vein, by partial resection (0.5-1.0 cm), blood aliquots were taken to determine the detailed indicators on a hematological analyzer BC-3000 (Mindray P.R., China). Then, under light ether anesthesia in animals, by simultaneous decapitation, blood was collected for biochemical studies. Biochemical parameters of blood serum were determined using standardized methods: aspartate aminotransferase (AST) and alanine aminotransferase (ALT) using the unified Wright-Frankel method; alkaline phosphatase (ALP) – a standardized method with nitrophenyl phosphate; total protein (TP) - colorimetric biuret method; gamma-glutamyltransferase (γ GT) - by the kinetic method; cholesterol (Chol) - by the enzymatic colorimetric method on a BA-88 A biochemical analyzer (Mindray, P.R., China) using complete sets of reagents (CYPRESS Diagnostics, Belgium).

Assessment of proliferative activity. Evaluation of the proliferative activity of ERc was carried out on cultures of normal fibroblast cells obtained by the explant method, according to the guidelines [Tseomashko N.Y. Azimova Sh.S., Urakov B.A. Evaluation of the cytotoxicity of drugs, medical devices, cosmetics, chemicals, pesticides and veterinary funds. Guidelines of Ministry of Health of the Republic of Uzbekistan, No. 8N-P/18. - 2016. - p. 46] from the skin of newborn rat pups. Cells were cultured in complete RPMI-1640 medium (Himedia, India) containing 10% FBS (Himedia, India) and a 1% solution of antibiotic-antimycotics (Himedia, India, where 10,000 units/ml of penicillin, 10,000 μg/ml streptomycin and 25 μg/ml amphotericin B) in a CO_2-incubator (ShelLab, USA) at +37°C, 5% CO_2. Cells after passage 5 were scattered in 96-well plates (Costar, USA) at 5 thousand cells per well. On the second day of incubation, the extract was digested with triplets at concentrations of 0.8-1.6-3.1-6.3-12.5-25-50-100-200 μg/ml. The proliferative activity of the cells was evaluated using the colorimetric method - neutral red test (Himedia, India), in which the vital dye neutral red through endocytosis enters the lysosomes of only living cells. As a comparison, we used intact cells into which only the culture medium was introduced [Tseomashko N. Y. Uzing cell cultures (normal and transformed malignantly) for screening biologically active substances and obtaining the monoclonal antibodies. Abstract for the degree of DSc in the specialty 02.00.10 - Bioorganic chemistry (biological sciences). - Tashkent. 2016. -p. 90].

Evaluation of antimicrobial activity by agar diffusion method. To determine the antibiotic potential of plant extracts, test bacteria from the collection of microorganism cultures of the Institute of Microbiology of the Academy of Sciences of the Republic of

Uzbekistan (Bacillus subtilis, Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Candida albicans) were used. A suspension of bacterial cells was prepared from the daily subculture of the corresponding strain, with 1×106 colonies in 1 ml. Sterile nutrient agar (Immunpräparate, Berlin, D, 25 g agar/L dis. Water) was inoculated with bacterial cells (200 µl of bacterial cells in 2 ml of 0.9% NaCl suspension and in 20 ml of medium) and added to Petri dishes to obtain a solid phase. The effect of substances on non-spore test cultures was determined in the exponential growth phase (after 36-42 hours), on spore-like ones at the stage of spore formation (after 48-72 hours). Evaluation of antagonistic activity was carried out on the 3rd day of incubation according to the diameter of the sterile zones in the bacterial lawn formed around the holes according to the Egorov method [Egorov, N.S., 1989. - 688 s.]. The experiments were carried out in three repetitions.

Creating a model of burn wounds IIIA degree. Models of burn wounds were modeled on outbred white rats weighing 180-200 g. The experiment was carried out on animals under ether anesthesia. On depilated areas of animal backs with a special electric device (a soldering iron at the end of which a copper plate with an area of 1.5x1.5 cm is fixed), heated to 1200 ° C, 2 wounds were applied to each animal. Each animal was kept in a separate cage. On the second day, 30% salicylic acid ointment was applied for 3 hours, after which scabs were removed from the wounds with gauze. Wounds were washed with sterile physical. solution (NaCl sodium chloride 0.9% isotonic solution) and immediately investigated wound coverings were applied. Monitoring the course of the wound process was carried out on the basis of: clinical observation data, such as the presence or absence of soft tissue edema in the wound area, hyperemia of the skin, healing time; as well as an objective criterion for assessing the course of the wound process were the histo-morphology data of the scarred area. Histo-morphology of the sites of skin injury after 22 days was carried out in the IPSUM PATHOLOGY Laboratory.

To assess the rate of wound healing, planimetric research methods were used. To do this, a sterile cellophane plate was placed on the wound, on which the contours of the wound were applied, and then its image was transferred to graph paper and the sizes of the contours were determined on days 3, 12 and 19.

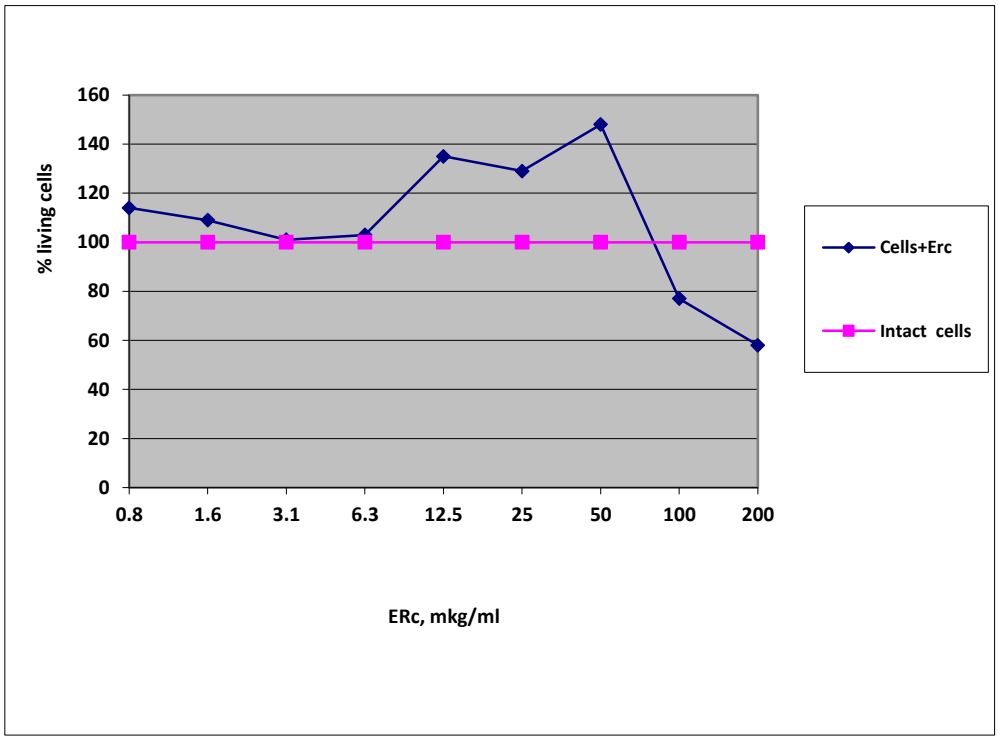
The experimental studies were carried out in compliance with the rules adopted by the European Convention for the Protection of Vertebrate Animals used for experiments or other scientific purposes (ETS N 123), Strasbourg, 03/18/1986. The results were subjected to statistical processing using the standard Statistika for Windows software package according to well-known methods of variation statistics with an assessment of the significance of indicators (M±m) and differences between the samples under study using the Student t-criterion. Differences in the compared groups were considered significant at a significance level of 95% ($p < 0.05$).

3. RESULTS

A new medicinal component was obtained - dry extract of tannic polyphenolic compounds - ERc, isolated from the ground and root parts of the plant Rumex confertus. The quantitative content of tannins in the extracts obtained by triple extraction showed that R. Confertus contains 8.3% in the roots and 5.1% in the aerial part. The yield of tanning polyphenolic compounds upon triple ethanol extraction was 91%. Assessed its toxicity, antimicrobial and proliferative activity, determined the active dose.

Assessment of cytotoxicity and proliferative activity was carried out on rat skin cell cultures, fibroblasts, using neutral red vital dye. The peak of skin cell proliferation was recorded at concentrations of 25–50 µg/ml, and an increase in concentration above 100 µg/ml led to a decrease in the lysosomal activity of cells and, accordingly, inhibition of their functional and proliferative activity (Fig. 1).

Fig. 1. The percentage of living fibroblast cells is normal and under the influence of different concentrations



As can be seen from Figure 1, with a decrease in the ERc concentration below 3 μg/ml, proliferative activity of fibroblasts is also observed. The most effective concentration of ERc for regenerative purposes can be considered 50 μg/ml.

When studying acute toxicity, an aqueous extract of ERc in doses of 100-500-1000-5000 mg/kg was administered orally and once. Symptoms of intoxication in animals were not observed in any of the groups. Animals did not die. In this regard, the LD50 of the extract isolated from the ground and root parts of the plant Rumex confertus, at this stage of research could not be established.

Subchronic toxicity of ERc was studied. The drug was injected into the stomachs of rats daily at doses of 50-100-500 mg/kg for one month. Each dose was tested in six rats. After the last injection of ERc, blood and organs were removed from all groups of animals for research. The results of the studies showed that long-term oral administration of aqueous ERc solutions in doses of 50-500 mg/kg is well tolerated by experimental animals. Hematological and biochemical blood parameters of rats that took the study drug in doses of 50-100-500 mg/kg are within acceptable generally accepted norms [Makarov V.G., 2013] and indices of the intact group of animals (Table 1, 2).

External morphological indices of internal organs also did not go beyond the norm, which confirms the picture of histological sections of the internal organs of experimental animals, i.e. internal organs - spleen, kidney, liver, stomach, small and large intestines without morphostructural changes (Fig. 2).

Table 1. Blood counts of rats treated with ERc for 30 days at three concentrations

Dose groups	White blood cells 10^9/l	The absolute content of lymphocytes, 10^9/l	The absolute content of a mixture of monocytes, basophils and eosonophils 10^9/l	Granulocyte count, 10^9/l	Hemoglobin, g/l	RBC	HCT	The average concentration of hemoglobin in the red blood cell g/l	Platelets in absolute numbers, 10^9/l	Thrombocrit %
Control	14,45 ±1,09	6,43±0,73	2,65±0,28	5,15±0,45	138.5±5,52	6,22 ±0,44	36,90 ±1,49	365.83 ±4,98	602,17 ±52,32	0.540 ±0.06
50 mg/kg	14.18 ±1,09	6,40±0,66	2,55±0,34	5,40±0,35	134,50 ±4,59	6,16 ±0,35	36,98 ±0,92	366,17 ±4,63	600.67 ±42,01	0.570 ±0.05
100 mg/kg	14,12 ±1,07	6,40±0,67	2,47±0,33	5,65±0,46	133.67 ±5,02	6,32 ±0,31	35,47 ±1,15	367,67 ±5	600,33 ±49,43	0.550 ±0.04
500 mg/kg	14,45 ±1,09	6,43±0,73	2,65±0,28	5,15±0,45	138.5±5,52	6,22 ±0,44	36,90 ±1,49	365.83 ±4,98	602,17 ±52,32	0.540 ±0.06

Note: P≥0.05 compared to control

Table 2. Blood biochemical parameters of rats treated with ERc for 30 days at three concentrations

Groups	Alanine Aminotransferase Activity, ALT	Activity of Aspartate Aminotransferase, AST	Alkaline phosphatase activity, ALP	Activity gamma-glutamyltransferase, γ GT	Cholesterol, Chol	Total protein, TP
	U/L (at 370C)				Mmol/l	g/dl
Control	64,80±3,44	231,67±12,67	593,83±31	4,67±0,75	81,77±3,	92,83±10

(intact)			,08		80	,54
50 mg/kg	66,43±2,58	247,17±11,71	549,02±20,64	4,50±0,76	76,90±3,39	99,60±4,54
100 mg/kg	67,03±3,20	242,33±11,33	603,67±34,42	4,50±0,82	74,92±4,63	94,48±4,14
500 mg/kg	70,13±4,63	249,17±11,21	582,03±27,72	5,17±0,76	73,92±3,13	94,28±3,24

Note: P≥0.05 compared to control

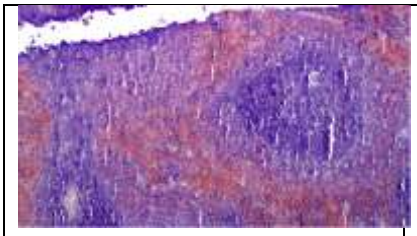
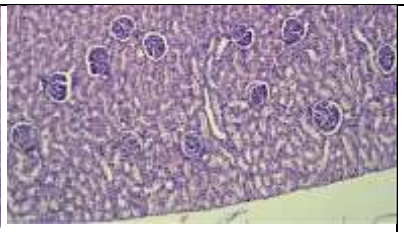
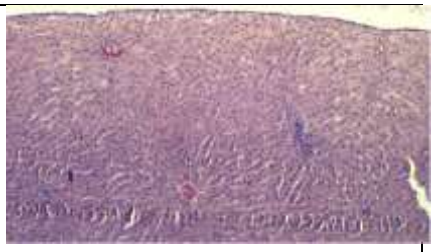
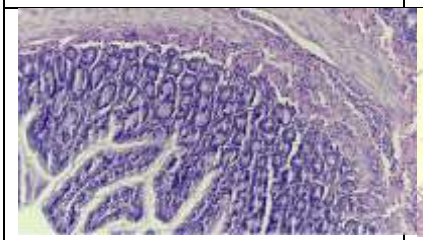
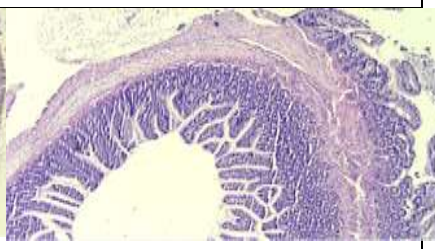
		
A: A capsule of the spleen is noted, from which the trabeculae depart. In the white pulp, clearly visible central arteries are visible. Trabecular veins are also noted. Coloring GE. SW 20x10.	B: Stroma is expressed by loose connective tissue. In the parenchyma, renal tubules are noted. Collector tubes are not modified. Coloring GE. SW 10x10.	C: The liver capsule consists of loose connective tissue, there are scantily distinguishable lobules in the parenchyma, a central vein is located in the center of the lobule, from it there are radial cords formed by two rows of hepatocytes. Coloring GE. SW 10x10.
		
D: Crypts of various sizes, Brunner's glands are noted. Coloring GE. SW 10x10.	E: Large intestine, with short villi and moderate lymphocytic infiltration in the stroma. Coloring GE. SW 10x10.	F: The mucous membrane of the stomach is lined with monolayer prismatic glandular epithelium. The mucous membrane has simple unbranched glands, the muscle

		plate consists of three layers of the inner and outer circular and middle longitudinal. The submucous membrane has a loose fibrous connective tissue. Coloring GE. SW 10x10.

Fig. 2. Histological sections of organs of rats taking ERc for a month at a dose of 500 mg/kg, where A is the spleen; In - a kidney; C is the liver; D - small intestine; E - large intestine; F is the stomach**.**

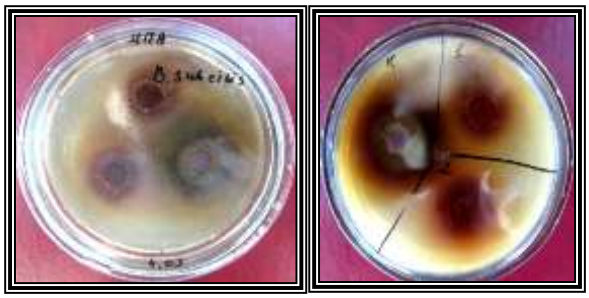
As can be seen from tables 1, 2 and Figure 2, hematological, biochemical and histomorphological parameters of tissues and fluids of animals treated with ERc are within normal limits, which indicates the absence of toxicity of this extract containing polyphenolic compounds. To determine the antibiotic potential of ERc, the test bacteria Bacillus subtilis, Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, and Candida albicans were used. As a control, the drug "Rutan" was used, obtained from the plant Rhuscoriaria at the Institute of Bioorganic Chemistry. The active ingredients of Rutan are polyphenolic compounds (tannins), as in ERc. A study of the antibiotic activity of these plant substances in relation to test cultures showed definite antagonistic and bacteriostatic activity (Table 3 and Fig. 3).

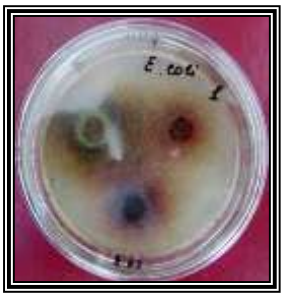
Table 3. Antimicrobial effect of substances (3rd day) (M ± m, n = 3, P <0.05)

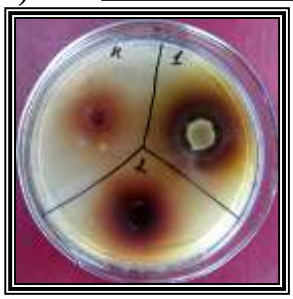
№	Antagonistic activity d (growth suppression zone), mm				
	Bacillus subtilis	*Escherichia coli*	*Pseudomonas aeruginosa*	*Staphylococcus aureus*	*Candida albicans*
Rutan 50 mcg/ml	**28,0±0,31**	**25,0±0,2**	**28,0±0,15**	**30,0±0,12**	**11,0±0,05**
Erc 50 mcg/ml	**23,0±0,17**	0	**18,0±0,09**	**12,0±0,07**	0
ERc 100 mcg/ml	**25,0±0,22**	**25,0±0,19**	**27,0±0,05**	**18,0±0,04**	0

Note: "0" - not detected

Fig. 3. Antimicrobial activity of ERc: 1 - 50 μg/ml ERc, 2 - ERc 100 μg/ml, K - Rutan, and - Bacillus subtilis; b - Escherichia coli; c - Pseudomonas aeruginosa; Mr. Staphylococcus aureus

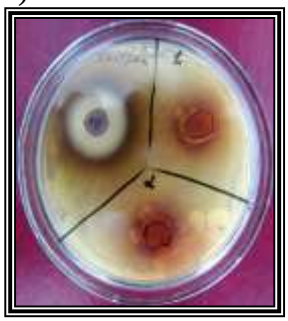
a) 

b) 



c) 

d) 



As a result of the studies, it was found that ERc at a concentration of 50-100 mg/ml is able to significantly suppress the growth and development of the studied conditionally pathogenic cultures, except for fungi of the genus Candida, while the most effective antibacterial concentration is 100 mg/ml.

Thus, it was found that concentrations of 50 mg/ml ERc are the most effective and acceptable for introducing wound dressing in the composition, since it stimulates the proliferation of mammalian dermal cells and partially inhibits the growth of Bacillus subtilis, Pseudomonas aeruginosa and Staphylococcus aureus.

In this regard, finely dispersed ERc powder at a rate of 50 mg/ml was introduced into a gel-like form of collagen isolated from bovine tail tendons, where 1% glycerol was also added to increase elasticity. The thoroughly mixed gel was poured into Petri dishes under sterile conditions, treated with UV rays for 20 minutes and dried in ammonia vapor to a film state of wound coverage, which was designated as CC-Rc.

Assessment of the specific activity of the obtained CC-Rc was carried out on models of thermal burns of the IIIA degree in 10 rats (2 wounds per animal). As a comparison, the commercial drug NeuSkin-F (Eucare Pharmaceuticals (P) Limited, India), a collagen-based

wound dressing, was used. In the process of applying wound coverage to wounds, it was revealed that the commercial NeuSkin-F preparation has very poor adhesion and congruency with the wound surface, while our CC-Rc immediately adhered to the surface of the wound beds. It was found that CC-Rc lead to wound healing of the indicated area in 18.4-19.4 days, while NeuSkin-F - for 22.1-22.9 days, and wounds without treatment for 25.3- 26.3 days. In addition, CC-Rc led to the formation of the correct architectonics of the pericardial and cicatricial repair zones, which is confirmed by the histomorphological data (Fig. 4, Table 4).

Table 4. The effect of wound dressing on the change in the area of wounds and the healing time (M±m, n=10, P <0.05)

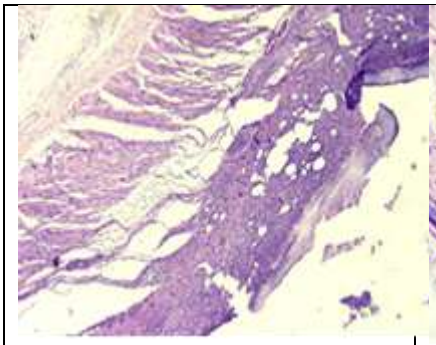
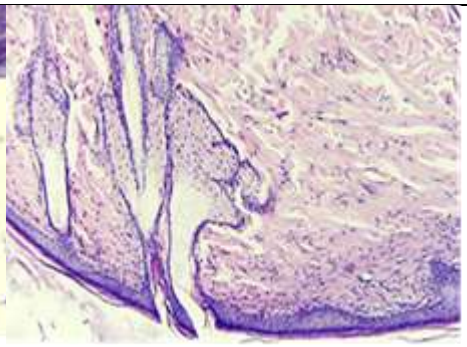
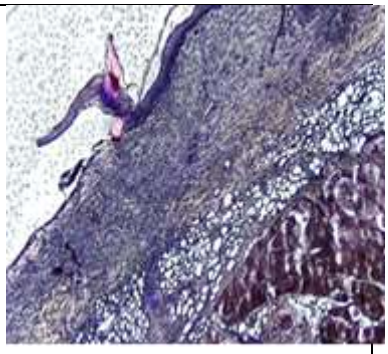
Experiment Conditions	**The average area of the wound, cm2**				**Wound Regeneration Dates**
	3rd day	**9th day**	**15th day**	**18th day**	**Day**
CC-Rc	2,5±0,07	1,8±0,01	0,8±0,01	0,02±0,01	18,9±0,5
NeuSkin-F	2,9±0,12	2,2±0,03	1,9±0,06	0,9±0,09	22,5±0,4
Control	2,9±0,11	2,3±0,09	1,8±0,07	1,1±0,07	25,8±0,5

Note: the healing time is indicated until the wounds are completely epithelized.

As can be seen from table 4, for complete epithelization of experimental burn wounds without human intervention, rats took 25.3 - 26.3 days, while wound therapy CC-Rc reduced the time for complete epithelization of wounds to 18.4 - 19.4 days, speeding up the process compared to control, about 1.5 times. At the same time, the commercial NeuSkin-F comparison drug (Eucare Pharmaceuticals (P) Limited, India) consisting of collagen without drug additives showed very low wound healing properties (complete healing only for 22.5 ± 0.4 days) in this experiment, but we believe that this is due to poor adhesion to the wound bed of this drug.

It is known that when applying NeuSkin-F to the wound bed in the clinic, it is fixed with additional dressings, which was difficult to perform in an animal experiment and was not required for CC-Rc. In this regard, our wound cover, which has significantly better congruence and adhesion to the wound, shows an excellent result and does not require additional fixation.

Slaughter of some animals for morphology was performed on the 20th day. A morphological study of the skin was evaluated using a 3-point system (developed by Ipsum Pathology (Tashkent, Uzbekistan), where histomorphological studies were conducted), to simplify the assessment of lesions: 1 point - slight changes in the form of thinning of the epidermis, minor erosion, not a thick inflammatory infiltration in the dermis, proliferation of connective tissue elements, which characterizes the regenerative activity of the tissue; 2

		
Control: The integumentary epithelium is replaced by granulation tissue and is represented by a ulcerative defect. 3 points. Coloring GE. SW 10x10.	CC-Rc: The epidermis is a stratified squamous epithelium. The papillary and reticular layers of the dermis are clearly distinguishable. Signs of compensation are noted in the form of proliferation of collagen fibers. 1 point Coloring GE. SW 10x10.	"NeuSkin-F" - comparison drug: Dermal exfoliation and peptic ulcer. 3 points. Coloring GE. SW 10x10.

points - erosive changes, moderate granulation and inflammatory infiltration with slight connective tissue replacement; 3 points - ulcerative defects of the skin, thick lymphocytic infiltration, severe granulomatosis (Fig. 4

Fig. 4. Histo-morphology of the cicatricial part of the skin of experimental animals.

As can be seen from Figure 4, the best effect is obtained by CC-Rc wound dressings, in which epithelization, at the time of removal, is at the last final stage, while in the remaining groups there is a delay in wound healing.

4. DISCUSSION

As a result of the studies, a dry extract of polyphenolic compounds was obtained, isolated from the ground and root parts of the plant Rumex confertus. It was found that ERc during intragastric administration does not cause toxic effects in experimental animals and their death, and can be assigned to hazard class 5 - practically non-toxic substances, although when studying the proliferative activity of this extract on skin cell cultures - fibroblasts, an increase in concentration above 100 μg/ml caused a suppression of the activity of fibroblasts and the predominance of cytotoxic effects, which is due to the direct effect of the extract on the cells, and with oral administration of ERc in the stomach and intestines, it may undergo metabolic changes, which reduces its toxicity. Wegiera M et al. Showed that ethanol extracts of plants of the species Rumex L., including Rumex confertus, exhibit high cytotoxic activity

in malignant transformed cell cultures in vitro, where the IC50 ranges from 0.22 to 1.91 mg/ml, respectively, which correlates with our data on fibroblast cells [Wegiera M, 2012.]. The cytotoxic effect of ERc at a concentration of 100 μg/ml and partially at a concentration of 50 μg/ml was also established on cultures of opportunistic strains of the microorganisms Bacillus subtilis, Pseudomonas aeruginosa and Staphylococcus aureus. Kustova T. et al. Identified extracts of various plants, among which was an extract from Rumex confertus. The authors showed that the extract from R. confertus is active against Staphylococcus aureus (IC50=10.80 μg/ml), Methicillin-resistant S. aureus (IC50=16.20 μg/ml), and also has antioxidant activity and offer it along with other plant extracts for the treatment of long-standing trophic ulcers [Kustova T, 2014]. Such a high antimicrobial activity of the extract of Rumex confertus obtained by the Kustova T. team and the difference with the antimicrobial activity of our ERc is due to the difference in the methods of isolation, use of different parts of the plant for extraction, places and dates of collection of the plant and, accordingly, the composition of the extracts, since it is known that anthraquinones The tannins of the pyrocatechol group, flavonoids, vitamin K, organic acids, resins, essential oils, iron, ascorbic acid, rutin, carotene and many other substances are contained in different quantities, compositions in different parts of the plant.

5. CONCLUSION

The resulting ERc containing polyphenolic tannins from Rumex confertus is assigned to hazard class 5 - practically non-toxic substances, since with a single oral dose of 5000 mg/kg it did not cause the death of experimental animals. But at a concentration above 100 μg/ml, it showed a cytotoxic effect both on normal skin cells and on the microorganisms Bacillus subtilis, Pseudomonas aeruginosa and Staphylococcus aureu. At the same time, a concentration of 50 μg/ml ERc significantly stimulates proliferative processes in skin cells, which allowed us to choose this concentration as effective and introduce it into the wound coating on a collagen basis. As a result, the resulting wound dressing under the name CC-Rc accelerates the time of complete epithelialization of wounds by 1.5 times. At the same time, the commercial NeuSkin-F comparison drug (Eucare Pharmaceuticals (P) Limited, India), consisting of collagen without drug additives, showed very low wound healing properties in our studies, which, in our opinion, is due to poor adhesion to the wound bed of this drug. In this regard, our wound coverings, which have significantly better congruence and adhesion to the wound, show excellent results and do not require additional fixation.

CONFLICT OF INTERESTS AND CONTRIBUTION OF AUTHORS

The authors declare the absence of obvious and potential conflicts of interest related to the publication of this article and report on the contribution of each author.

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