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REGENERATION EFFECT OF AN AQUEOUS EXTRACT OF CYMBOPOGON PROXIMUS ON INFECTED WOUNDS IN A RODENT MODEL OF STEROID HYPERGLYCEMIA

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Vera Inchina, Mutwakel Hussein, Tatyana Tarasova✉, Ziaul Haq

National Research Ogarev Mordovia State University, Saransk, Russia

✉ *9023060@mail.ru*

ABSTRACT — We analyzed the processes of regeneration of an infected skin wound with Streptococcus epidermidis (Streptococcus) in 30 laboratory rats where a model of hyperglycemia was created by administration of dexamethasone. Further, these animals were divided into 3 equal groups: in the 1st group the wound was treated with an ointment containing Cymbopogon proximus; in the 2nd group an ointment containing sodium fusidate 2% was applied to the wound; in the 3rd group (control) — Vaseline was applied. The use of Cymbopogon proximus leads to an increase in the total number of white blood cells, an increase in the activity of lymphocytes (+383%) (p= 0.03), which leads to improved regeneration of the infected wound. Also, we noticed that the glucose level was restored to the initial values, in 5 days of oral intake of an aqueous extract of Cymbopogon proximus, in contrast to the other groups (p=0.001). We found that the regeneration activity of an infected wound in rats with steroid hyperglycemia does not significantly differ in case of antimicrobial ointment and Cymbopogon proximus treatment.

KEYWORDS — Diabetic foot, infected wound, medicinal plants research, Cymbopogon proximus, regeneration, steroid hyperglycemia.

INTRODUCTION

There are many diseases that are accompanied by chronic pain syndrome [1, 2]. Corticosteroids are often used to relieve persistent pain [3]. Long-lasting use of corticosteroids is the main cause of hyperglycemia [4]. This makes some trouble, as the number of patients suffering from diabetes continues to increase. Chronic hyperglycemia in diabetes leads to the development of severe complications, such as atherosclerosis, heart and kidney failure, neuropathy, and immunodeficiency [5, 6, 7]. The most serious complication of diabetes is the development of a diabetic foot [8]. The dynamics of the frequency of detection of new cases of diabetic foot in adult patients in the Russian Federation tends to increase annually [8]. It is known that diabetic foot often forms trophic ulcers, which are 50% infected [9]. Therefore, it is important to search for safe drugs with both hypoglycemic, antimicrobial and anti-inflammatory effects [5, 10].

Aim

To explore the regeneration processes of infected skin wounds in rats on the background of steroid hyperglycemia using Cymbopogon proximus (lemongrass) growing in Sudan.

METHODS

This was an experimental research that we carried out on 30 white rats weighing 200–250 g, which were obtained in the animal nursery laboratory *Andreevka* (Moscow region). All manipulations with animals were performed in accordance with the guidelines for the maintenance and use of laboratory animals [11]. First, we created a model of hyperglycemia in animals by intramuscular administration of dexamethasone 4% — 0.2 ml for 4 days. This rhythm of dexamethasone injections leads to the development of hyperglycemia, the glucose level in animals increases 6 times — from 5.08±1.18 to 30.025±16 mmol/l (p<0.001). Next, we created a model of an infected skin wound in rats. To do this, we made an incision of 2,0 x 0,5 cm on the skin of the animal in sterile conditions, under local anesthesia. Then, we performed a double transfer of the exudate infused with Streptococcus epidermidis (which we took from a patient suffering from streptodermia) to the wound area. The next step was to divide all experimental animals in a randomized manner into 3 groups:

— Group 1 — treatment of wounds with an ointment based on vaseline and powder from plant Cymbopogon proximus in 1:2 dilution. In addition, these rats received daily 2 ml of water extract of Cymbopogon proximus orally for 5 days (n=10).

— Group 2 — treatment with an ointment containing sodium fusidate 2% for 5 days (n=10).

— Group 3 (control) — treatment of the wound with vaseline for 5 days (n=10).

After 5 days of therapy, we visually assessed the wound condition on a five-point scale, where 5 points

were the maximum signs of wound inflammation, and 0 points were the absence of signs of inflammation. We also studied changes in the peripheral blood of animals on the hematological analyzer PCE-90Vet (USA). Statistical processing of the material was carried out using the program "STATISTICA 7.0". The reliability of differences between quantitative indicators was assessed using the Mann–Whitney test. The differences were considered significant at $p < 0.05$.

RESULTS

After stopping the administration of dexamethasone and before the end of the experiment, the glucose values in the second and third groups of the study were within the range of 10.4±2.85 mmol/l. in animals of the first group, when using an aqueous extract of Cymbopogon proximus, the glucose level decreased to 5.4 ± 0.79 mmol/l ($p=0.001$). Table 1 shows the analysis of infected wound regeneration in all groups of rats. We did not get significant differences between the effectiveness of antimicrobial ointment based on sodium fusidate 2% and Cymbopogon proximus ($p>0.05$). At the same time, we observed marked wound regeneration on the background of Cymbopogon proximus compared to the control group ($p< 0.05$).

Accelerated regeneration of a wound, infected with Streptococcus epidermidis, when treated with Cymbopogon proximus extract, is associated with the effect of this herb on the cells of the immune system (Table 2).

The drug form based on Cymbopogon proximus prevents the development of leukopenia, leads to the growth of lymphocytes (+383%), which has a positive effect on the rate of wound regeneration. However, we recorded a decrease in the level of neutrophils in the first group of rats compared to other groups of animals. Perhaps this fact was associated with a faster exudation phase, which is characterized by the migration of neutrophils from the vascular lumen to the tissues for bacterial phagocytosis.

DISCUSSION

Currently, an active research is underway for effective and safe drugs that stimulate wound regeneration. Widely known are herbal preparations based on Aloe vera, Calendula officinalis, Hypericum perforatum, etc., which have antimicrobial, antiviral, anti-inflammatory, immunomodulatory properties due to the inhibition of cytokines and macrophages [10]. Selim SA. evaluated the antimicrobial and antioxidant properties of Cymbopogon proximus (oil and methanol extract) in vitro [12]. The results of the antimicrobial test showed that the methanol extract of Cymbopogon proximus has moderate antibacterial activity, and the oil extract strongly suppresses the growth of the test bacteria, with the exception of yeast [12]. Our research shows that cymbopogon proximus extract has a positive effect on most components of the wound process: it reduces the severity of hyperemia, swelling, compaction, soreness and increases the intensity of regeneration. We did not find a significant difference in the rate of wound regeneration when treating with an ointment based on cymbopogon proximus extract and an ointment containing sodium fusidate ($p>0.05$).

CONCLUSIONS

Therapy with an aqueous extract of Cymbopogon proximus has a reliable hypoglycemic effect, which allows using this herb for both prevention and correction of hyperglycemia. The effect of Cymbopogon proximus on the regeneration of an infected wound in conditions of hyperglycemia caused by dexamethasone did not differ from the effectiveness of antimicrobial ointment.

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Table 1. *Regeneration indicators of an infected wound in white rats with steroid hyperglycemia during the treatment with various groups of drugs*

The wound state	1st group(therapy Cymbopogon proximus)	2nd group (therapy sodium fusidate 2%)	3rd group (control)
Hyperemia	+++	++	+++++
Edema	+	+	+++++
Painfulness	+	++	++++
Consolidation of the wound edges	+	++	++++
Regeneration activity	+++++	++++	+

Table 2.*The level of immune blood cells in rats with an infected skin wound on the background of steroid hyperglycemia in the treatment of various groups of drugs*

Blood / Indicators / Patient Groups	Leukocytes (*109/л)	Neutrophils (%)	Lymphocytes (%)
1st group (therapy Cymbopogon proximus)	2,9+0,2	16,46±5,82	87,05±2,8
2nd group (therapy Sodium fusidate 2%)	0,92±0,15	39,07±6,66	31,25±12,25
3rd group (Control)	2,42±0,26	24,43±8,06	74,65±11,19
p	< 0,05	< 0,05	= 0,03

** comparing the first group of animals with the second and third*

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