Original Article

Wounds Repairing Drugs

Effects of Lactobacillus (Acidophilus) on Cutaneous Wound Repair with Respect to Neutrophil Count in Comparison with Steroid

1. Hina Khan 2. Muhammad Asif Sheikh 3. Mazhar ul Haque 4. Aneela Qureshi 5. Amin Fahim 6. Nazia Qamar 7. Adnan Anwar

1. Asstt. Prof. of Anatomy, Al-Tibri Medical College, Karachi 2. Principal & Assoc. Prof. Isra Institute of Rehabilitation Sciences Karachi 3. Asstt. Prof. of Anatomy, 4. Prof. of Pathology, 5. Asstt. Prof. of Pathology, 6. Lecturer of Pathology, 7. Lecturer of Physiology, Al-Tibri Medical College, Karachi

ABSTRACT

Objective: To evaluate the effects of lactobacillus (acidophilus) in repairing of wound with respect to neutrophil count in comparison with steroid therapy.

Study Design: Experimental study

Place and Duration of Study: This study was conducted in the Department of Anatomy of Al-Tibri Medical College Isra University Karachi Campus from June 2014 to October 2014.

Materials and Methods: Randomly selected Wistar albino rats were taken and their weight was between 100 – 150 gms. The rats were divided in three groups on the basis of topical application. Group I was treated with topical normal saline, Group II with topical lactobacillus(acidophilus), and Group III topical Steroid were applied once daily).Each group is further divided into four sub groups according to the day of sampling like group Ia (day 03), group Ib (day 07), group Ic (day 14) and group Id (day 21). Samples were taken on day 3,7,14 & 21 from each group and tissues were processed and stained to observe the neutrophil count that showed rapid healing processes. Data was analyzed on SPSS version 21 by applying ANOVA and post hoc tukey's test.

Results: Results compared the neutrophil count of Lactobacillus group with steroid group and control group. Mean ± SD of numbers of Neutrophils / x400 in group IIa was20.500 ± 0.577, in group IIb11.250 ± 1.258, in group IIc5.7500 ±.0.5000, group IId3.7500 ± 0.9574and in groupIawas54.25 ± 2.872, group Ib41.250 ± 2.629, group Ic23.500 ± 3.000 and group Id18.750 ± 0.957. The significant value was in group IIa and group Ia(P<0.000), group IIb and group Ib(P<0.000), group IIc and group Ic (P<0.000) and group IId and group Id (P<0.000). Mean ± SD of numbers of Neutrophils / x400 in groups IIa was 20.500 ± 0.577, in group IIb 11.250 ± 1.258, in group IIc 5.7500 ±.0.5000, group IId 3.7500 ± 0.9574 and group IIIa was54.500 ± 1.290, group IIIb 42.000 ± 1.414, group IIIc32.000 ± 0.8165 and group IIId28.750 ± 1.258. The significant value was in group IIa and group IIIa (P<1.000), group IIb and group IIIb (P<0.002), group IIc and group IIIc (P<0.000) and group IId and group IIId (P<0.000).

Conclusion: Lactobacillus showed potent anti-inflammatory effect against both therapeutic groups.

Key Words: Wound, Lactobacillus, Cutaneous, Neutrophil

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INTRODUCTION

The primary role of the skin, it serves as a protective barrier from the environment[1]. The damage to the cell, tissues, organs and viscera including the skin is usually restored by a complex process termed as wound healing. Normal pattern of wound healing comprises a sequence of coordinated overlapping procedures or phases that engage acute and chronic inflammation and cell division[3]. Lactobacillus species and bifidobacterium species were reported as the most frequently used probiotic strains[4]. Lactobacillus species are referred to a group of lactic acid-producing organisms[5]. Lactobacillus acidophilus bacteria produce exopolysaccharides (EPS) that can be associated to a cell's surface or can discharge into the environment[6]. The main factor contributing to the process of healing is extracellular medium which is mainly composed of various structural and functional proteins[7].After the initial injury while there is formation of fibrin clot, there is extravasation of neutrophils at the site of injury. This is usually accomplished within 24hours at the margins of wound[8].Many researchers have focused the role of Lactic acid in wound repair as it is being consumed in cosmetic products such as moisturizers,

Correspondence: Dr. Hina Khan,
Assistant Professor of Anatomy, Al-Tibri Medical College, Karachi
Cell No.: 0331-3504341
E-mail: draminfahim@gmail.com

body lotions and exfoliants[9]. Anabolic hormones such as, glucocorticoides, cortisol, mineralocorticoides, prolactin are involved in the protein synthesis[10].The process of wound repair is also enhanced by Dehydroepiandosteron having potential in wound healing[11].

MATERIALS AND METHODS

Design of Study: Experimental study
Setting: Anatomy department of Al- Tibri Medical College Isra University Karachi campus. Conducted after the approval of ethical committee of Isra University from June 2014-October 2014.
Sampling Technique: Randomized
Sample size: A total of 60 Wister Albino rats were divided into three groups on the basis of treatment given. Group I was treated with topical normal saline, Group II with topical lactobacillus (acidophilus), and Group III topical Steroid were applied once daily. Samples were taken on day 3,7,14 & 21 from each group and tissues were processed and stained to observe the neutrophil count that showed rapid healing processes. Each group is further divided into four sub groups as follows; group Ia (day 03), group Ib (day 07), group Ic (day 14) and group Id (day 21). Data was analyzed on SPSS version 21 by applying ANOVA and post hoc tukey's test.
Isolation of Lactobacilli: Species of lactobacillus were obtained from yogurt and strains were identified by the Microbiology department of PCSIR, Karachi and grown on (MRS) agar medium and incubated at 35^0C for 48 hours. The solution contained 10^{10} to 10^{11} CFU / ml bacteria that were collected every day after 48 hours.
Wound Formation: "Wound" was created on the dorsal surface of rats which was open with full-thickness measuring about 1.5 × 1.5 cm^2. Tissue was processed and stained to observe the neutrophil count.
Data analysis procedure: Data was analyzed on SPSS version 21 by Applying ANOVA and post hoc tukey's test.

RESULTS

The results compare the neutrophil count among the three therapeutic groups. Basically compare the other with the group applied lactobacillus (B)**.** Results compared the neutrophil count of Lactobacillus group with steroid group and control group**.** Mean ± SD of numbers of neutrophils / x400 in group IIa was 20.500 ± 0.577, in group IIb 11.250 ± 1.258, in group IIc 5.7500 ±.0.5000, group IId 3.7500 ± 0.9574 and in group Ia was 54.25 ± 2.872, group Ib 41.250 ± 2.629, group Ic 23.500 ± 3.000 and group Id 18.750 ± 0.957. The significant value was in group IIa and group Ia (P<0.000), group IIb and group Ib (P<0.000), group IIc and group Ic (P<0.000) and group IId and group Id (P<0.000). Mean ± SD of numbers of neutrophils / x400 in groups IIa was 20.500 ± 0.577, in group IIb 11.250 ± 1.258, in group IIc 5.7500 ±.0.5000, group IId 3.7500 ± 0.9574 and group IIIa was 54.500 ± 1.290, group IIIb 42.000 ± 1.414, group IIIc 32.000 ± 0.8165 and group IIId 28.750 ± 1.258. The significant value was in group IIa and group IIIa (P<1.000), group IIb and group IIIb (P<0.002), group IIc and group IIIc (P<0.000) and group IId and group IIId (P<0.000) as shown in Figure 1a and figure number 1.1 and 1.3, that showed the decrease number of neutrophil in lactobacillus group as compared with steroid given group.

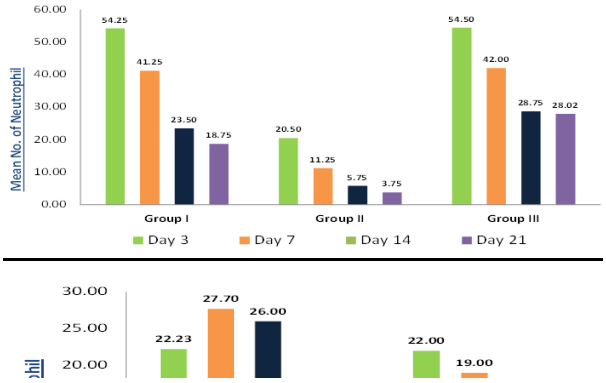
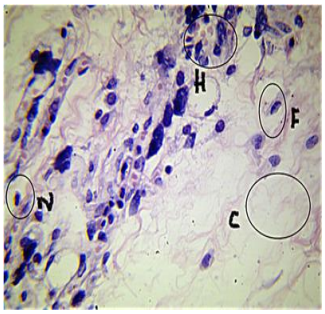
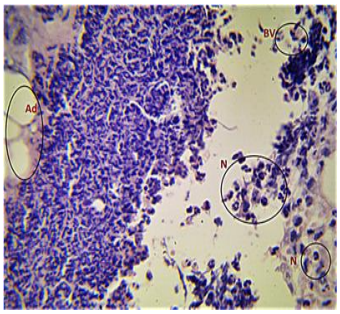


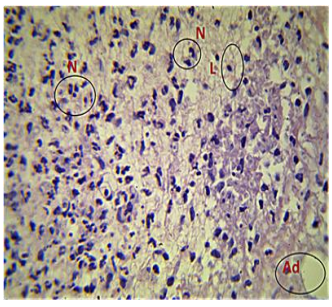
Figure No.1: Mean ± SD No. of Neutrophil /x 400



Photomicrograph 1.1 shows decreased neutrophil count/400X (H&E stain) of Group II. H (hemorrhage), F(Fibroblast), C (collagen fibers), N (



Photomicrograph 1.2 shows increased neutrophil count/400X (H&E stain) of Group I. Ad (Adipose tissue), BV(blood vessels) and N (neutrophil)



Photomicrograph 1.3 shows increased neutrophil count/400X (H&E stain) of Group III. Ad (Adiposetissue), L(Lymphocytes) and N (neutrophil)

DISCUSSION

Lactobacillus can easily be obtained by normal flora and from the food source of our daily life. It has got potential benefits if it is used for the accurate purposes. Few of the studies have also revealed the beneficial role of lactobacillus on the natural micro intestine[12]. The induction of live strains of Lactobacillus acidophilus activated the cytokines which in turn are involved in the re-epithelization by production of cell components[13]. Marked reduce count of neutrophil can be seemed in lactobacillus treated group in accordance with some of researcher studies[14]. In accordance with Halper J; our study showed significant reduction in neutrophil count in lactobacillus treated group, that's shows the potency of lactobacillus species as a strong anti inflammatory agent. On day 3-4 increase numbers of neutrophil shows inflammatory phase, but this study reports on day 3 decreased in count of neutrophil showed anti-inflammatory role of lactobacilli in comparison with others[15].

CONCLUSION

On the bases of this study it evaluated that Lactobacilli are potent anti-inflammatory agent, as they showed decrease count of neutrophil in inflammatory phase of healing in comparison with other therapeutic groups.

Conflict of Interest: The study has no conflict of interest to declare by any author.

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