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ALTERNATIVE WOUND HEALING EFFECTS OF COCONUT OIL EXTRACT USING ADULT WISTAR RATS

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ABSTRACT

Introduction

The occurrence of wound is a common phenomenon as long as the body is exposed to environmental forces, and the problems of treating them are as old as mankind. **Objectives:** This study aimed at developing an applicable and cost effective wound treatment agent, using indigenous material such as coconut oil, to enhance the wound healing processes via animal model. **Materials and methods:** Coconut oil was obtained from coconut milk extract of twenty (20) fresh coconuts, each weighing between 200-230 g. Fifteen (15) male wistar rats weighing approximately 192g - 226g were used. They were grouped into a control (C), standard (S) and experimental (E) groups, with each comprising of five (5) rats. A wound size of 2cm by 2cm was inflicted on the dorsolateral aspect of the thorax of the rats. The animals in the control group were dressed with distilled water, those in the standard group were dressed with hydrogen peroxide (methylated spirit) and those in the experimental group were dressed with coconut oil. Wound size measurement and dressing were done every three (3) days, until their wounds were completely healed. **Results:** The percentage (%) mean wound contraction on day 9 of the C group (39.3 ± 5.2) against the E group (69.8 ± 3.8) was found to be statistically significant ($P < 0.05$). The mean healing day of C group (28.8 ± 1.6) against the E group (18.6 ± 1.3) was found to be statistically significant ($P < 0.05$). The percentage mean wound contraction on day 9 of the S group (59.0 ± 5.5) against the C and E groups respectively, were found to be statistically significant ($P < 0.05$). The mean healing day of the S group (22.8 ± 1.6) against the C and E groups, were found to be statistically significant. **Conclusion:** This study has shown that rats treated with coconut oil extract had fewer days of wound healing and this was evident considering the wound contracture.

KEYWORDS

Contracture, Healing, Methylated spirit and Coconut.

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INTRODUCTION

Wound healing is an intricate process where the skin or other body tissue repairs itself after injury. In normal skin, the epidermis (surface layer) and dermis (deeper layer) form a protective barrier against the external environment. When the barrier is broken, an orchestrated cascade of biochemical events is quickly set into motion to repair the damage[1,2]. Wound healing, as a normal biological process in the human body, is achieved through four precisely and highly programmed phases: hemostasis, inflammation, proliferation, and remodeling. For a wound to heal successfully, all four phases must occur in the proper sequence and time frame[3]. The events of each phase must happen in a precise and regulated manner. Interruptions, aberrancies, or prolongation in the process can lead to delayed wound healing or a non-healing chronic wound[4]. Wound contracture is a process that occurs throughout the healing process, commencing in the fibroblastic stage[5].

Wound care in long-term care continues to be a difficult issue. The utilization of proper wound care techniques is essential in promoting healing. Product cost, nursing time, patient comfort, and infection control issues need to be considered[6]. The treatment of acute and chronic wounds is an ancient area of specialization in medical practice, with a long and eventful clinical history that traces its origins ancient Egypt and Greece. The Ebers Papyrus, circa 1500 BC, details the use of lint, animal grease, and honey as topical treatments for wounds[7]. Dressings may play an important adjunctive role in concert with overall efforts to manage the underlying causes of chronic non-healing wounds. Wound dressings have undergone an evolutionary process from natural materials that simply covered and concealed the wound, to materials that focused on moisture management and more recently, to materials that either deliver active ingredients or interact directly with cells or specific chemicals in the local wound environment[8]. Even with an extensive array of research available to discount their place in wound care, wet-to-dry dressings continue to be used. Moisture dressings have shown promise in reducing pain and infection and promoting healing[6].

Wound management is a significant clinical and economic problem[9]. There is a paucity of information on the actual cost of wound management in Nigeria. Studies have calculated the cost of wounds to the NHS in United Kingdom to about £1 billion a year[10]. Studies also revealed that 31.55% of the total hospital patients at a Nigeria Teaching Hospital were being managed for wounds. The cost to the health system can be very significant; this particularly is so in a developing country like Nigeria[11]. Therefore in this study, we research to ascertain the effect of coconut oil on wound healing processes with respect to wound contraction. This may be a cheaper and good alternative for wound healing since coconut, an indigenous plant is readily available.

MATERIAL AND METHODS

Plant Materials

Coconuts (*Cocosnucifera* L.) weighing 200-230 g were obtained from Umuokeh Community, in Obowo L.G.A., Imo State of Nigeria. It was authenticated and identified by Iwueze Francis O., of the department of Forestry and Wildlife, School of Agricultural Technology, Federal University of Technology, Owerri; as a dwarf (autogamous) Coconut (*Cocosnucifera* L. Arecaceae). The fresh coconut oil was obtained from the coconuts.

Experimental Animal

A total of 15 adult male Wistar rats with body weights of192g-226g obtained from the breeder in the animal house of Department of Human Physiology, University of Port Harcourt, Choba, Nigeria, were used in the study. The animals were housed in standard environmental conditions where they were fed with standard rodent diet and given water ad libitum. The animals were acclimatized in the laboratory conditions for 2 week before the commencement of the study. The experimental procedures adopted in this study were in strict compliance with the United States National Institutes of Health Guidelines for Care and Use of Laboratory Animals in Biomedical Research (1985, no. 85-23)[12].

Methods

Extraction of Coconut oil

Twenty coconuts (*Cocosnucifera* L) were obtained; the exocarp and mesocarp were removed. After that, the endocarp (meat) was blended using a blender. The coconut water which obtained from the coconuts was added to the blended coconut and mixed. After which, the blended coconut particles (chaffs) were sieved and squeezed out of the mixture, leaving the liquid remnant which is a mixture of the coconut milk and coconut water. The liquid remnant is heated to evaporate the water present thereby leaving the oil which comes settle on top of the coconut cake that is being formed. The coconut oil was then carefully filtered in a clean sterile bottle and stored in glass case maintained at room temperature.

Experimental Groups and Protocol

The rats were divided randomly into 3 groups comprising 5 rats in each group. They were all fed with the same diet throughout the experimental period. The experimental design is described as follows:

Anaesthesia

On expiration of the acclimatization, the rats from the respective groups were an anesthetized one at a time. This was done by clapping and shaving the dorsal aspect of each animal, and then the area cleaned with 70% ethanol. This was followed by (intramuscular) I/M injection of 10mg/kg body weight ketamine hydrochloride and 5mg/kg body weight zylazine by injecting the animals with 0.3ml of equal mixture of Diazepam and ketamin intra-muscularly.

Infliction of Wound

When animal attained a subconscious state, at the dorsolateral aspect of the thorax using a sharp razor with the help of a forcep, a 2cm by 2cm square full thickness circular excisional wound including all layers of skin; after determination the area of each wound with a marker wound was inflicted on the skin of the shaved region.

Mode of Dressing

This was done by covering the wound with gauze soaked in coconut oil and firmly secured with a plaster in the experiment group, also a gauze soaked in hydrogen peroxide (methylated spirit) and firmly secured with a plaster in the standard group, while the control group were dressed as well with the wound covered with gauze soaked with distilled water and secured with plaster. The wound was re-dressed after every seventy two (72) hours, that is every three (3) days.

Assessment of Wound Contraction

This was done by placing a white transparent sheet of about 5cm square on the wound after the plaster and gauze had been removed after each three days interval, and then marking the sized of the wound on the white transparent sheet using a felt pen. The marked part was calculated by counting the number of square blocks of graph sheet that falls within the circumference when placed on the graph sheet.

Statistical Analysis

Data were analyzed using SPSS. One-way Analysis of Variance (ANOVA) was applied and the significant means were separated using the Duncan multiple range test. The significance of differences was calculated by using student t-test. $P<0.05$ was considered statistically significant.

RESULTS

A better healing pattern was observed in Wister rats treated with methylated spirit (Standard group) and coconut oil and (Experimental group), with complete wound closure observed within 24 days and 18 days respectively; while it took about 30 days in wistar rats treated with water (Control group) as shown (Figure No.1). There was a significant reduction in wound area from day three onwards in treated Wistar rats and also on later days the closure rate was much faster than when compared with control wistar rats. However, in some of the control wistar rats we observed that the wound size increased on day 24, with subsequent death of the animals on day 30.

DISCUSSION

Excision wound model are often used for wound healing bioassays because they represent a true wound that could be reproducibly analyzed in a non-subjective highly controlled manner[13]. Wound healing involves a complex and superbly orchestrated interaction of cells, extracellular matrix and cytokines[14]. Granulation, collagen maturation and scar formation are some of the cascade of wound healing which run concurrently, but independent of each other[15]. Multiple factors can lead to impaired wound healing. In general terms, the factors that influence repair can be categorized into local and systemic. Local factors are those that directly influence the characteristics of the wound itself, while systemic factors are the overall health or disease state of the individual that affect his or her ability to heal. Many of these factors are related, and the systemic factors act through the local effects affecting wound healing[3]. In modern biomedical area, development for the management of wound healing is an expensive program for the people of developed countries, several drugs obtained from natural sources[16-25] are known to increase the healing and repair processes of different types of infected wounds[26].

Wound contraction is considered an important factor in the evaluation of healing process in large open wounds[27]. In this study, a close examination of wound healing of animals in the control, standard and experimental groups respectively showed that on day 9 (Table No.2), wounds treated with distilled water (control) had a percentage mean wound contraction of (39.34 ± 6.3), while those treated with hydrogen peroxide methylated spirit (standard) had percentage mean wound contraction of (59.00 ± 2.3), and those treated with coconut oil (experiment) had a mean wound contraction of (69.84 ± 4.5). The results depicts a significant increase ($p<0.05$) in the wound contraction of the experimental group (Wister rats treated with coconut oil) compared to the experimental group (Wister rats treated with methylated spirit) and control group (Wister rats treated with distilled water). However, *Aloe vera*acet one extract on full thickness excisional wounds results in more accelerated contraction of treated wounds with 100% contraction rate at 11th day post-wounding[28], while *A. indica* and *T. cordifolia* treated wounds which showed 95.65% and 77.02% mean wound contraction on the 21st day respectively; which also showed a significant increase compared to the control group[29].

A drug to be used for effective wound healing should be able to clear the wound by the 19th day after infliction. A hundred percent (100 %) healing was achieved in animals treated with ointments containing 100 mg/g of NI in cationic emulsifying agent and in the ointment containing Cicatrin powder, by the 19th day after treatment, as the wound sizes reduced to zero[30]. Also, this study showed (Table No.3) that a complete wound healing was achieved in coconut oil treated wounds on the 18th day. The wound healing day obtained in the methylated spirit and distilled water treated Wister rats were 24 days and 30days, respectively. It was also observed that two out of the five rats that were treated with distilled water died before the 30th day. The delayed wound healing and death can be due to contamination of the wound[31]. Base on this study, coconut oil showed a more efficient wound healing potential when compared to the complete wound healing achieved with *A. indica* (21 days) and *T. cordifolia* (21 days) treated wound[29]. The wound contraction and healing effect of coconut oil can be attributed to its antioxidant[32] and antibacterial property, as a result of high monolaur in content[33-36]. Thus, coconut oil achieved its therapeutic effect on wound healing by its ability to overcome some factors that militate against faster wound healing.

Recommendation

The inclusion of coconut oil into the list of therapeutic agents, and its applications in cutaneous wound management, would be a step-in the right direction. Also, more investigations into the molecular interactions of coconut oil and other living tissues would be necessary.

Group	Protocol
Control group (C)	Rats were fed with pellets and water, wound was inflicted, and dressed with distilled water and gauze.
Standard group (S)	Rats were fed with pellets and water, wound was inflicted, and dressed with methylated spirit and gauze.
Experimental group (E)	Rats were fed with pellets and water, wound was inflicted, and dressed with coconut oil and gauze.

Table No.1: Percentage Wound contraction WC for control group (C), standard group (S) and experimental group (E) at day 9

S.No	Sample	%WC for C Group (X_1)	% WC for S Group (X_2)	%WC for E Group (X_3)
1	1	32.61	56.40	68.51
2	2	31.14	56.29	68.68
3	3	37.67	53.87	59.93
4	4	45.98	57.12	71.53
5	5	42.19	60.21	70.00
Mean		**37.92**	**56.78**	**67.73**

% = Percentage, WC = Wound contraction, X_1 = C group, X_2 = S group, X_3 = E group

Table No.2: Paired Samples Test for Percentage Wound Healing at Day 9

		Paired Differences							
		Mean	**Std. Deviation**	**Std. Error Mean**	**95% Confidence Interval of the Difference**		**T**	**Df**	**Sig. (2-tailed)**
					Lower	**Upper**			
Pair 1	CONTROL - STANDARD	-18.86000*	5.72819	2.56172	-25.97249	-11.74751	-7.362	4	0.002
Pair 2	CONTROL - EXPERIMENTAL	-29.81200*	6.63308	2.96640	-38.04806	-21.57594	-10.050	4	0.001
Pair 3	STANDARD - EXPERIMENTAL	-10.95200*	3.18781	1.42563	-14.91019	-6.99381	-7.682	4	0.002

*. The mean difference is significant at P < 0.05.

Table No.3: Paired Samples Test for Mean Wound Size at day 18

		Paired Differences							
		Mean	**Std. Deviation**	**Std. Error Mean**	**95% Confidence Interval of the Difference**		**t**	**df**	**Sig. (2-tailed)**
					Lower	**Upper**			
Pair 1	CONTROL - STANDARD	0.55800*	0.26117	0.11680	0.23371	0.88229	4.777	4	0.009
Pair 2	CONTROL – EXPERIMENTAL	1.19429*	0.11692	0.05229	1.04911	1.33946	22.841	4	0.000
Pair 3	STANDARD - EXPERIMENTAL	0.63629*	0.19261	0.08614	0.39713	0.87544	7.387	4	0.002

*. The mean difference is significant at P < 0.05.

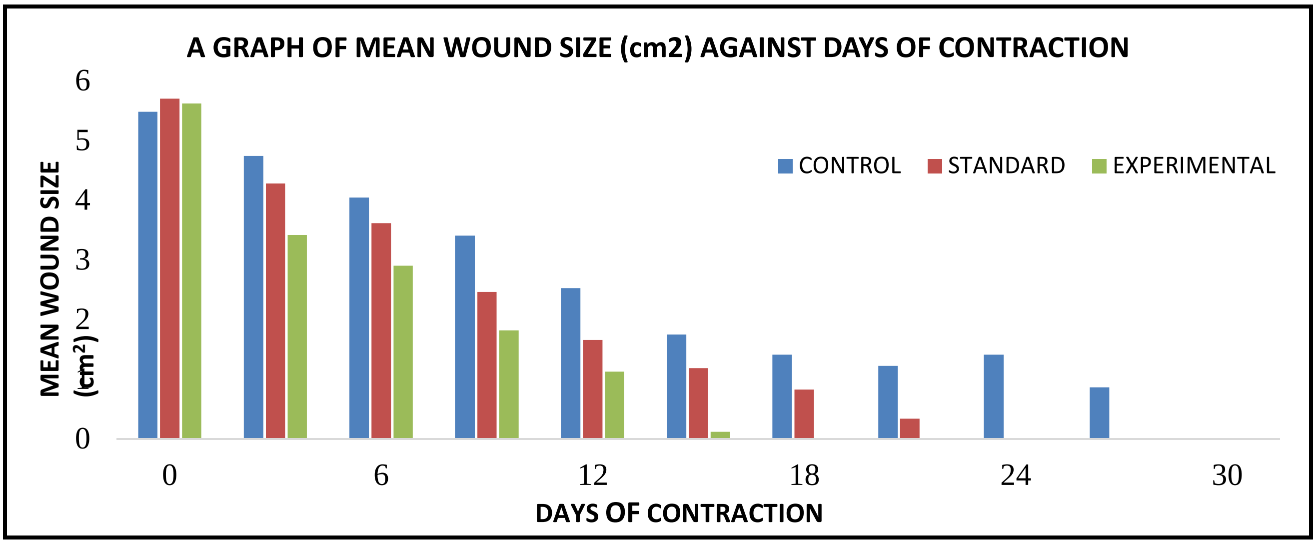

Figure No.1: Differential wound healing in Wister rats treated with water (control), methylated spirit (standard) and coconut oil (experimental)

CONCLUSION

The profound wound contraction and healing effect of coconut oil on Wister rats as documented in this study, is a clear indication of its ability to enhance healing through the prevention of infection and inhibition of prolonged inflammation. These were achieved as result of the antibacterial and antioxidant activities of coconut oil in the model animals.

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CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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