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ANTIBACTERIAL AND HEALING POTENTIALS OF CHARMIL® AS A SURGICAL WOUND DRESSING AGENT IN GOATS.

Eze, C.A. and Nlebedum U.
Department of Veterinary Surgery, University of Nigeria.Nsukka, Nigeria.

ABSTRACT:
The antibacterial prophylaxis and wound healing potentials of Charmil® were assessed using rumenotomy procedure in twelve West African Dwarf Goats (WADG). Following their acclimatization, the animals were divided into groups of four animals each, with normal saline serving as negative control treatment whereas the (Tetrasol®) group served as positive control.

The incision wounds were swabbed for bacterial culture immediately after the closure of the surgical site on post surgical day (P.s.d)0, and P.sds 3,7,10, and 14. The daily clinical records which include the assessment of wound healing process as wetness, dryness, crust formation and scar tissue formation (S.T.F.) were obtained during the same period. The rate of wound healing in the three groups revealed that by the 14th day P.S. all the wounds treated with both Charmil® and oxytetracycline spray healed completely while those of the normal saline treated group healed at a much slower rate. Though Tetrasol® exhibited higher antimicrobial activity than Charmil®, there is no significant difference (P>0.05) between the two agents. In this study therefore Charmil® demonstrated very high antimicrobial and healing potentials in comparison to that of Oxytetracycline spray. The study shows that though Charmil® could be used as a sole prophylactic wound dressing agent, there is a strong need for the manufacturers of the product to make public the active constituents of the product to encourage its wider application in Veterinary practice.

KEY WORDS: Antibacterial, wound Healing, charmil®, Surgical Dressing Agent, Goat.

INTRODUCTION:
Small ruminants are a major component of the livestock sector in most parts of the world including Nigeria (Odo, 2003). They are primarily kept for meat (protein) (Payne 1990) and improvement of socio-economic status of communities (Devendra and Mclorey, 1987, Ajala, 1995). However, small ruminant production has some constraints and disease is one among them. This often is associated with high mortality and decline in productivity and especially reproductive performance and even public health concerns (Mbise et al, 1984; Nyange, 1984). Management system affects productivity of animals. The traditional free-range system often predispose sheep and goats to injuries like fractures, matchet cuts, and self-inflicting injuries like horn gores etc.
Further more, premature pregnancy with consequent dystocia is one of the major disadvantages of free-range system of management. In major Veterinary Hospitals and State clinics in Nigeria, surgical cases (arising from dystocia, castration, fractures, and sadism) ranks third among cases reported (Eze and Idowu, 2002). Sheep and goats are prone to surgical wounds resulting from caesarian operation, rumen fistulation and rumenotomy due to plastic disease (foreign body impaction). While cases of helminthosis, viral and bacterial infections are usually handled on herd basis, cases of injuries and surgical wounds are handled on individual basis. In Nigeria, small ruminant production is usually on small holder scheme by individual house-holds/families. This highlights the importance of individual treatments.

Despite improvements in surgical techniques and the use of antibacterial agents for prophylaxis (Lane and Copper, 1999), surgical site infections remain an important complication of surgery among these animals (Iwu, 1983, Amyces and Gemmel 1997, Robert, 1997). In fact, sepsis attributable to surgical site infection constitutes an important cause of post-surgical death (Vande Plasche, 1998).

Broad-spectrum systemic and topical antibiotics have been used to prevent/ control post operative sepsis. Penicillin and streptomycin have been widely used to combat post-operative infection in farm animals (Gyang, 1986, Irvin and Roger, 1994). Unfortunately many wound contaminating bacteria such as Stapylococcus spp. have developed resistance to commonly available antibiotics (Lowbury and Ayliffe, 1974, Gales et al, 2000). Many natural products are said to possess antibacterial properties (Chah and Nweze, 2000).

Problems of resistance, environmental degradation and pollution associated with irrational use of orthodox drugs have necessitated renewed interest in nature as source of effective and safer alternative in the management of animal and human infection. Thus, in recent years there has been a phenomenal rise in the interest of scientific community to explore the pharmacological activities of medicinal plants (Gales and Roland, 2002).

In spite of systemic antibiotic administration, topical agents are also instituted on the wound sites in other to achieve greater and quicker healing (Classen, 1992).

Charmil® is a scientific herbal oil reported to be possessing antibacterial, anti-fungal, anti inflammatory, anti – pruritic, and maggoticidal properties. This study therefore is aimed at confirming the claimed antibacterial prophylaxis and healing potential of this agent as a sole surgical wound-dressing agent. This agent is cheap and readily available and if found efficacious, would offer therapeutic alternative for use in the management of cutaneous wounds in clinical veterinary practice.

MATERIALS AND METHODS:

Twelve clinically healthy West African Dwarf (WAD) goats of both sexes aged between 6 – 9 months and weighing between 3.6 – 6.5 kg, were purchased at Afor-Opi market, Nsukka, Nigeria. They were acclimatized for three weeks. The animals were fed ad-lib with freshly cut forage and clean drinking water. They were dosed with Ivomec® at the rate of lml/50kg BW subcutaneously to control both ecto and endo parasites. Procaine penicillin (10,000 I.U/kg) and Streptomycin Sulphate (10mg/kg) were administered intramuscularly to obviate any bacterial infection. National Veterinary Research Institute produced Pestesdes petit
ruminant (PPR) vaccine was subcutaneously given at the fold of the neck against PPR infection.

The animals were then identified with tags and daily clinical parameters like the rectal temperature (RT), heart rate, (HR), and respiratory rate (RR) were monitored from the day of purchase till the end of the study. Following this, the goats were divided into groups of four animals each.

Surgery:

Rumenotomy was carried out on each of the animals in the three groups using standard procedure (Kneeth et al 1987). Surgical pre-anaesthetic medication as well as anaesthesia was achieved using Xylazine and 2% Lignocaine HCL (Hall and Clarke, 1991).

Prophylactic post surgical treatments using Procaine penicillin and Streptomycin Sulphate at dose rates of 10,000 I.U./kg and 10mg/kg I/M for five days were given. In addition, group A animals were topically dressed with Oxytetacycline spray (Tetrasol®), whereas groups B and C were dressed with Charmil® and normal saline respectively. This topical application lasted till day 14 post-surgery.

Bacteriological Evaluation:

The blood agar used in culturing the specimen was prepared using defibrinated sheep blood. Following surgery, the wounds were swabbed for culture at days 0, 3, 7, 10 and 14 post-surgery.

Parameters For Wound Healing:

The parameters used in monitoring wound healing include wetness, dryness, crust formation, and scar tissue formation (Archibald and Blackley, 1974, Oehme, and Prier, 1974).

Statistical analysis:

The data obtained from the study were analyzed using one way Analysis of variance (ANOVA).

RESULTS AND DISCUSSION:

Swab cultures obtained from the surgical wounds on post surgical day zero (P, sd.0) in all animals in the three experimental groups did not yield any bacterial growth. Between P.S. days 3-7, in spite of the daily topical dressing using the agents for the different experimental groups, bacterial yield was observed from swab cultures of the surgical wounds in all animal groups including the positive control group. However, from P. Sd. 10 up to the termination of the experiment (P.Sd.14), swab cultures in positive control group could not yield further growth (fig.1), while those of Charmil® and normal saline continued to yield bacterial growth. (fig.1).

In terms of bacterial yield, there was a significance difference (P<0.05) among the three groups on P.sd3 (Figure 1) as represented by the coded variations in the group means in bacterial growth on wounds dressed with these agents. Throughout the observation period while scanty (P.S.days3-7) or no (P.S.days10-14) bacterial growth was observed in positive control treatment wounds, those of Charmil® and normal saline (negative control) yielded scanty and extensive growth respectively.

However, yields got on P.S. day 7 on Charmil® treated group decreased to the level that there was no significance difference (P>0.05) between the group and positive control group. It appears from this result at this period, that while the bacterial growth in positive control group remained static the negative control group (normal saline treated) showed an increasing yield. By P. Sd 10 and 14, bacterial growth were absent in Tetrasol® treated group whereas Charmil® and normal saline treated groups continued to
demonstrate scanty and extensive growth respectively.

In terms of wound description, there was a significant difference (P<0.05) between the means in the rate of wound healing in the treatment groups by P.S.d3. Whereas crust formation was 100% in groups A and B, 50% of the animals in group C had wetness of the wound, the remaining, 50% had their wound dry (Table1). Similarly, by p.s. days 10-14, there was a significant (P<0.05) difference among the three treatment groups. The wounds in groups A and B were characterized by scar tissue formation, crust formation was common in-group C animals.

Surgical site contamination is not peculiar to this study. Study of surgical wounds in human and animals around Nsukka showed that a total of 134 bacterial isolates were obtained from 114 samples that yielded growth. *Staphylococcus aureus* (26.1%) was the most prevalent isolated from humans, followed by *E. coli* (17.4%), *Proteus Spp* (17.4%), *Enterococcus Spp* (13.0) and *Coagulase negative Staphylococcus [CNS] (10.9%). In animals, the most common agents isolated were *E. coli* (19.0%) and *Proteus Spp* (19.0%), *S. aureus* (14.3%) and CNS (9.5%) (Chah et al, 2005).

This study shows that in spite of hospital environment, the surgical sites in the animal groups were widely contaminated by day 3-post surgery. The contamination as shown could result from the hairs around the surgical wounds which trap a lot of bacteria within the wound (Achibald, 1974, Knecht, et al, 1987). In the field of surgery, aseptic surgical technique cannot prevent bacterial contamination, and despite its improvements and the use of antibacterial agents for prophylaxis, surgical site infections remain an important complication of surgery (Iwu, 1983, Amyces and Gemmel 1997). Sepsis attributable to surgical site infection constitutes an important cause of post-surgical death.

In the study, Oxytetracycline spray (Tetrasol®) was used for prophylactic purposes. Even though, this treatment was effective in controlling infection, it did not prevent the emergence of bacterial growth. This limitation of antibiotic therapy and antibiotic prophylaxis, has been widely recognized and emphasized in recent years (Iwu, 1983, Amyces and Gemmel 1997).

Since, 1953, tetracycline resistant bacteria have been found increasingly in humans, animals, food and environment. Today, many wound contaminating bacteria such as *Staphylococcus Spp.* have developed resistance to the commonly available antibiotics. The antimicrobials to which high rates of resistance are demonstrated are the ones that are commonly requested, inappropriately prescribed, and administered in Veterinary and human medical practice (Gyang, 1986, Chah, et al, 2003).

Nature has been a source of medical agents for thousands of years and an impressive number of modern drugs have been isolated from natural sources, mainly based on their use in traditional medicine. Natural products though effective are not readily accepted in the orthodox medical practices, because, they are not standardized and in cases of toxicity, their active constituents are not known (Akubue et al, 1983). The active constituents of Charmil® as currently packaged and marketed in Nigeria, is not known, neither is its safety guaranteed. This actually necessitated this study, at least to demonstrate its efficacy in prophylactic management of surgical wounds. Nevertheless, the agent, Charmil® has in this study demonstrated a high degree of antibacterial prophylaxis and wound healing activity, comparable to the Oxytetracycline spray.
Apart from the wound healing property of Charmil®, the preparation is said to be antipruritic, maggoticidal and miticidal. From the result of the study, Tetrasol® no doubt demonstrated greater antibacterial activity than Charmil®. However, their wound healing potentials showed no significance difference (P>0.05) throughout the experimental period. Its ability to compete with Tetrasol® in this regard could be attributed to the antipruritic property of this agent. From day 10p.s. whereas the same bacterial load (scanty growth) was observed for charmil® treatment, no bacterial growth was recorded for Tetrasol® spray. Bacterial infection is one of the factors that delay wound healing (Achibald, 1974, Chah, et al, 2003). Within the same period, day 10 (P.S.), both Charmil® and Tetrasol® (positive) treatments (wounds) showed 100% scar tissue formation whereas the normal saline (negative control) showed only crust formation.

**Conclusion:**
Charmil® has a high degree of antibacterial prophylactic and wound healing activity comparable to Tetrasol® spray as topical agent. However, it is suggested that the active constituents and possibly the structural formation be made known to encourage its use in veterinary practice.

**Table 1: Wound Healing descriptions during the post surgical days amongst the Treatment groups**

<table>
<thead>
<tr>
<th>Post-Surgical Observations</th>
<th>0</th>
<th>3</th>
<th>7</th>
<th>10</th>
<th>14</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Animal group A</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wet</td>
<td>4/4*</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
</tr>
<tr>
<td>Dry</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
</tr>
<tr>
<td>C.F</td>
<td>0/4</td>
<td>4/4</td>
<td>3/4</td>
<td>0/4</td>
<td>0/4</td>
</tr>
<tr>
<td>S.T.F</td>
<td>0/4</td>
<td>0/4</td>
<td>1/4</td>
<td>4/4</td>
<td>4/4</td>
</tr>
<tr>
<td><strong>Animal group B</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wet</td>
<td>4/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
</tr>
<tr>
<td>Dry</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
</tr>
<tr>
<td>C.F</td>
<td>0/4</td>
<td>4/4</td>
<td>2/4</td>
<td>0/4</td>
<td>0/4</td>
</tr>
<tr>
<td>S.T.F</td>
<td>0/4</td>
<td>0/4</td>
<td>2/4</td>
<td>4/4</td>
<td>4/4</td>
</tr>
<tr>
<td><strong>Animal group C</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wet</td>
<td>4/4</td>
<td>3/4</td>
<td>0/4</td>
<td>0/2</td>
<td>0/2</td>
</tr>
<tr>
<td>Dry</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/2</td>
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</tr>
<tr>
<td>C.F</td>
<td>0/4</td>
<td>1/4</td>
<td>3/3</td>
<td>2/2</td>
<td>1/2</td>
</tr>
<tr>
<td>S.T.F</td>
<td>0/4</td>
<td>0/4</td>
<td>0/4</td>
<td>0/2</td>
<td>1/2</td>
</tr>
</tbody>
</table>

*No of Animals per total number Animals in a group.
C.F. = Crust formation
S.T.F. = Scar tissue formation.
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