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Assessing the potentials of two local topical ointments as affordable treatment against tungiasis infestation: A self-experimentation in Igbokoda, Nigeria

Victor N. Enwemiwe [a,b,*], Cynthia C. Ojianwuna [a], Okorie O. Anyaele [b]

[a] Department of Animal and Environmental Biology, Delta State University, Abraka, Nigeria
[b] Department of Zoology, University of Ibadan, Ibadan, Nigeria

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ABSTRACT

Infestations caused by penetration of *Tunga penetrans* (Siphonaptera: Hectopsyllidae) in cutaneous sub-layers present morbidities and resulting mortalities if surgical treatment remains the obtainable. Considering the neglected nature of this infestation and the absence of marketable product, we report an observation on the use of grounded naphthalene in kerosene ointment, and powder of *Piper guineense* in coconut oil ointment as affordable treatment of embedded tungiasis flea in Igbokoda. A total of 80 individuals partitioned into two groups of 40 individuals each were assigned the locally made topical ointment irrespective of stages of tungiasis lesions. The mean of stage I, II and III tungiasis lesions treated with grounded naphthalene in kerosene ointment respectively decreased from 1.83, 3.42 and 3.89 to 0 after 5–6 days ($p < .05$). Also, the mean of stage I, II and III tungiasis lesions treated with grounded *P. guineense* ointment respectively decreased from 1.52, 3.10 and 5.00 to 0 after 6 to 7 days of treatment exposure ($p < .05$). Very high significant difference between stages and exposure days of participants assigned the two topical ointment was recorded $p < .0001$ and $p = .0005$ respectively. Naphthalene ointment and to a lesser extent *P. guineense* ointment is best to control and hinder development cycle of embedded fleas irrespective of infested part and stage of infection.

1. Introduction

Tungiasis infestation results from the penetration of the cutaneous layer of an individual by *Tunga penetrans* (Pampiglione et al., 2009). In Igbokoda, sand fleas is known as *takpere* but variously known in the world as pecker, chigo, chigoe, chica, tungiasis, pique, pico and chique (Macías and Sashida, 2000; Veraldi and Valsecchi, 2007; Hakeem et al., 2010; Smith and Procop, 2002; Feldmeier et al., 2004; Winter et al., 2009). The infestation has generally been considered an endemic disease associated with the rural poor (Harvey et al., 2017). *T. penetrans* occur mainly in areas with poor hygiene. Bitam et al. (2010) ascribed the growth of *T. penetrans* larvae in endemic areas to the organic matter present in improperly disposed sewage and animal nest associated with human surroundings. Sand fleas as the name implies inhabit white dry loose sandy or dusty soil of coastlines, beaches, foot paths and unpaved areas. Animals that bask and bath on loosed sands during warm periods are mostly

* Corresponding author at: Department of Animal and Environmental Biology, Delta State University, Abraka, Nigeria.
E-mail address: enwemiwevictor@gmail.com. (V.N. Enwemiwe).

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at risk of sand flea penetrations. This has led to the resultant increase in animal reservoir host of the infestation in chickens and pigs especially (Ugbomoiko et al., 2008; Mutebi et al., 2015).

In some parts of the world such as in rural Madagascar where treatment approaches were investigated, tungiasis infestation is highly prevalent and its prevalence is reportedly between the ranges of 13 to 100% (Thielecke et al., 2014). Various treatment methods have been adopted against *T. penetrans* infestation. They include the surgical treatment of viable lesions through sterile extraction, the use of Zanzarin® - a plant-based repellent, the use of various mixtures into lotion such as 0.5% ivermectin, 0.2% metrifonate, 5% thiabendazole lotion, and disinfectants such as 0.05% potassium permanganate, hydrogen peroxide, Liasol ™, dimeticones mixture of low viscosity, Dettol and Izal, tincture of iodine, and sodium hypochlorite (Heukelbach et al., 2003; Ugbomoiko et al., 2007; Buckendahl et al., 2010; Morkve, 2013; Thielecke et al., 2014). In addition, treatment targeted on animal reservoir host of infestation using the combination of 10% imidacloprid and 50% permethrin (Advantix®) has been investigated in dogs with positive results (Klimpel et al., 2005). The potentials of Naphthalene and kerosene, and *Piper guineense* as insecticidal agents against tungiasis lesions has not been demonstrated. However, the use of *P. guineense* as larvicidal agent has been proven by a few studies. Anyaele et al. (2002) has recorded positive results with the use of hexanolic extract of *P. guineense* seed oil against *Aedes aegypti*. Akunne et al. (2014) has also used *Piper guineense* fruit powder for short term protection of dry fish against *Dermestes maculatus* (Degeer) and recorded highest mortality in both adult and larvae respectively.

Various other practices including the use of soap to bath, personal protection such as protective clothings, closed shoes, and socks, and control measures such as the use of insecticide to spray house, wetting sandy or dusty floors, paving the unpaved floors and sweeping floors have been reported to reduce the attack by *T. penetrans* (Macías and Sashida, 2000; Feldmeier et al., 2004; Ugbomoiko et al., 2007; Veraldi and Valsecchi, 2007; Pampiglione et al., 2009; Hakeem et al., 2010). The recommended insecticide include ICON or hyper-cypermethrin spray, carbaryl insecticidal dust, propoxur insecticidal dust, DEET topical repellant, Malathion and many other repellents (Heukelbach et al., 2003; Feldmeier et al., 2004; Schwalfenberg et al., 2004). The use of friendly chemical spray on the environment has been reported to ward off fleas and play the critical function in the decrease of flea infestation burden whenever other personal protection measures are lacking (houses Schwalfeereerg et al., 2004; Karunamoorthi, 2013). Heavily infested households in Kenya have their houses fumigated with recommended fumigant by the Ministry of Public Health and efforts are been made to increase the number of target households. But in Nigeria this kind of intervention is unheard of.

Ugbomoiko et al. (2007) noted that an integrated approach involving clinical treatment of flea lesions, spraying of environment by public health specialists, studies on animal reservoirs by veterinarians, and correct identification of flea by zoologists would engineer the complete eradication of sand flea infestation. The implementation of this integrated approach comes with financial constraint. The rural poor households cannot afford this approach unless government or non-governmental organizations intervene. It is equally difficult for individuals with no formal education to acquire the technical knowledge on the use of insecticide fumigation, topical application of lotion on the skin, or the use of protective clothing.

All reported cases of tungiasis have shown that it is a neglected infestation associated with the rural poor and all the treatments tried so far are either unavailable in the market or unaffordable where they exist. The complete eradication of tungiasis-associated morbidities would be impossible where control measure with positive results are unaffordable. Failure to outline the production process of a treatment, use raw materials that are country-based, and state directives for use would make the treatment process difficult. There are no published studies on the potentials of Naphthalene and kerosene mixture, and ground *P. guineense* on tungiasis lesions. However, the information of this study is important for the development of a marketable prophylaxis for tungiasis lesions or as affordable treatment in any rural poor community infested with tungiasis fleas. The use of the raw materials in this study is imperative because they are common in all areas of the world and are affordable. The successful adoption of these locally available and affordable raw materials topical treatment would minimize infestation by tungiasis flea in the affected areas. Therefore, we report an observation on the use of grounded naphthalene in kerosene ointment, and grounded *P. guineense* in coconut oil ointment as affordable treatment of embedded tungiasis flea in Igbokoda.

2. Materials & methods

2.1. Study area

The study was performed in Zion Igbokoda, a delineated area of Ilaje Local Government, an agglomeration of Ondo State. Zion is a village along the coastlines. According to the observations made by the community health officers, individuals in Zion Igbokoda are at a high-risk of tungiasis flea penetrations.

2.2. Experimental population

Participants for the study included a total of 80 randomly selected infested individuals in Zion Igbokoda. The study population was made up male and females of ages 5–60 years. The prevalence of tungiasis in Zion is reportedly 43.9% (Unpublished study of Enwemiwe and Anyaele). Their tungiasis lesions were partitioned using classification by Eisele et al. (2003) and recorded. Lesions were further sectioned into the following: Stage I represented a penetrating flea burrowing into the epidermis of the skin, Stage II represented an itching reddish brown spot with a diameter of 1 to 3 mm, and Stage III circular lesions presenting as a whitish patch with a diameter of 4 to 10 mm with a central black dot. Exposure of the participants to further tungiasis infection was not maintained to allow for effective assessment of the topical ointments.

2.3. Ointment preparation

Considering the self-experimental nature of the study, household was randomly selected and blindly grouped into two with 40 individuals in each. Group A purchased 100 g of synthesized naphthalene and a litre of kerosene from the local market. Naphthalene balls were grounded and mixed with kerosene (100 g/l). Group B equally purchased *Piper guineense* seed (100 g) and a litre of freshly milled coconut oil from a local dealer in the community (100 g/l). The seeds were dried, blended into powder and mixed with coconut oil. Of the 80 infested individuals, forty was assigned each to Naphthalene and kerosene mixture, and ground *P. guineense* in coconut oil topical ointment irrespective of the stages of lesions. Infested individuals applied the Naphthalene in kerosene ointment by dipping infected feet or hands in a bowl containing the mixture while the ground *P. guineense* in coconut oil was applied only on the affected part by rubbing. All individual included in this study had at least one tungiasis lesion on hands and feet, cooperated with the self-experiment by applying treatment twice daily for 2 weeks, and was present during the four weeks treatment period. Infested individuals were examined daily during the topical application of Naphthalene in kerosene (100 g/l) and ground *P. guineense* in coconut oil (100 g/l).

2.4. Ethical considerations

This study involved human subjects who voluntarily decided to participate following a written informed consent translated to Igbokoda (Ilaje) language for better understanding. Thus this study was conducted according to the "Nigeria National Code for Health Research Ethics (NNCHRE)". The ethical review committee of the College of Medicine, University of Ibadan (reference no.: UI/EC/17/0404) approved the written protocol for the study.

2.5. Ointment assessment and analysis

The effectiveness of the two topical treatment was assessed during a follow up of fourteen days using morphological features of lesions, and no pain upon pressure from viable lesions. All data were entered into a Microsoft Excel database and analyzed using XLSTAT, 2019 Version. Two way ANOVA test was used to determine the differences of significance within days of treatment and infection stages.

3. Results

3.1. Treatment with naphthalene in kerosene ointment

The summary of mean of different stages of tungiasis lesions treated with the Naphthalene and kerosene ointment from day 0 over a two weeks post treatment period is presented in Table 1. Before topical application of Naphthalene and kerosene ointment on infested individuals, the mean of stage I, II and III tungiasis lesions were 1.83, 3.42 and 3.89 respectively. Two days after topical application, 62.3% of stage I lesions, 8.9% of stage II, 15.3% of the stage III lesions presented no pain. On the third day of topical application, 94.3% of the stage I lesions, 61.8% of stage II lesions and 79.9% of the stage III lesions presented no pain and drying up. On the fourth day of topical application, a mean of 0.24 of stage I lesions were remaining on affected body part and 96.2% had dried up while 10.6% of the stage II lesions had dried up and 77.8% of the stage III lesions presented no pain, and dried up. On the fifth day, 100% of the stage I lesions, 47.8% of the stage II lesions and 75% of the stage III lesions had dried up. On the sixth day onwards, 100% of the stage II and III lesions were dried up. The 40 infested individuals that had 53, 123 and 144 stage I, II and III respectively before topical application of ointment recorded no lesions on day 5 for stage I and scares of dried lesions on affected body part on day 6 for stage II and III lesions. No adverse reactions or health-related condition was observed during and after 2 weeks post treatment.

3.2. Treatment with P. guineense in coconut oil ointment

The summary of mean of different stages of tungiasis lesions treated with the ground *P. guineense* mixed with coconut oil ointment from day 0 over a two week post treatment period is presented in Table 2. The mean of stage I, II and III tungiasis lesions on infested individuals before topical application of grounded *P. guineense* in coconut oil ointment were 1.52, 3.10 and 5.00 respectively. From the day 0 to 2nd day after topical application, no change was observed in stage I and II but 21% of lesions presented no pain. However on the 3rd day, 13.7% of stage I lesions, 1.5% of stage II lesions, 74% of the stage III lesions showed no pain respectively. On the 4th day, 56.3%, 95.4% and 74% of stage I, II and III lesions respectively presented no pain and dried up. On the 5th day, a mean of 0.78 and 2.95 stage I and II lesions were respectively recorded on the affected body part of the participants. Although, 85.7% of stage I lesions presented no pain and dried up while 1.5% of stage II lesions showed no pain only. But 55% of the stage III lesions continuously dried up. On the 6th day, 100% of the stage I lesions were dried up while stage II and III lesions have reduced to a mean of 2.43 and 1.23 respectively with 11% and 98% continuously dried up. On the 7th day up till the 14th day, 100% of the stage II and III lesions dried up and no visible lesions were observed. The 40 infested individuals that had 51, 130 and 195 stage I, II and III respectively before topical application of ointment recorded scares of dried lesions after treatment exposures and showed no adverse reactions or health-related conditions.

Table 1
Summary of mean of different stages of tungiasis lesions treated with the Naphthalene in kerosene ointment (day 0) over a two weeks post treatment periods.

Exposure (days)	Stage I Lesions	Mortality (%)	Stage II lesions	Mortality (%)	Stage III lesions	Mortality (%)
0	1.83 ± 0.2	0.0	3.42 ± 0.4	0.0	3.89 ± 0.4	0.0
1	1.83 ± 0.2	0.0	3.42 ± 0.4	0.0	3.89 ± 0.4	0.0
2	1.83 ± 0.2	62.3[a]	3.42 ± 0.4	8.9[a]	3.89 ± 0.4	15.3[a]
3	1.66 ± 0.2	94.3[ad]	3.19 ± 0.4	61.8[ad]	3.49 ± 0.4	79.9[ad]
4	0.24 ± 0.2	96.2[ad]	3.11 ± 0.4	10.6[d]	2.30 ± 0.3	77.8[ad]
5	0.00 ± 0.0	100.0[d]	1.56 ± 0.4	47.8[d]	1.00 ± 0.3	75.0[d]
6	0.00 ± 0.0	0.0	0.00 ± 0.0	100.0[d]	0.00 ± 0.0	100.0[d]
7	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
8	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
9	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
10	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
11	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
12	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
13	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
14	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0

Analysis of variance ($\alpha = 0.05$): $F_{(Days)} = 10.98$, $P < .05$; $F_{(Stage)} = 6.02$, $P < .05$.
[a] Lesions presenting no pain upon pressure.
[d] Lesions dried up.

Table 2
Summary of mean of different stages of tungiasis lesions treated with the grounded *P. guineense* in coconut oil ointment (day 0) over a two weeks post treatment periods.

Exposure Days	Stage I lesions	Mortality (%)	Stage II lesions	Mortality (%)	Stage III Lesions	Mortality (%)
0	1.52 ± 0.1	0.0	3.10 ± 0.4	0.0	5.00 ± 0.9	0.0
1	1.52 ± 0.1	0.0	3.10 ± 0.4	0.0	5.00 ± 0.9	0.0
2	1.52 ± 0.1	0.0	3.10 ± 0.4	0.0	5.00 ± 0.9	21.0[a]
3	1.52 ± 0.1	13.7[a]	3.10 ± 0.4	1.5[a]	3.72 ± 0.5	73.9[a]
4	1.42 ± 0.2	56.3[ad]	2.95 ± 0.3	95.4[ad]	3.64 ± 0.6	74.4[ad]
5	0.78 ± 0.2	85.7 [ad]	2.95 ± 0.3	1.5[a]	1.23 ± 0.3	55.3[d]
6	0.00 ± 0.0	100.0[d]	2.43 ± 0.3	10.8[d]	0.87 ± 0.2	98.4[d]
7	0.00 ± 0.0	0.0	0.00 ± 0.0	100.0[d]	0.00 ± 0.0	100.0[d]
8	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
9	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
10	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
11	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
12	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
13	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0
14	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0	0.00 ± 0.0	0.0

Analysis of variance ($\alpha = 0.05$): $F_{(Days)} = 8.26$, $P < .05$; $F_{(Stage)} = 15.03$, $P < .05$.
[a] Lesions presenting no pain upon pressure.
[d] Lesions dried up.

4. Discussion

The control of tungiasis through on-host intervention measure have so far been successful. The use of different substances including plant extracts, compounds, and ointments on tungiasis lesions have been studied (Heukelbach et al., 2003; Ugbomoiko et al., 2007; Buckendahl et al., 2010; Morkve, 2013; Thielecke et al., 2014). The continuous use of compounds in positive reports is sometimes not possible because they are not affordable treatments for any rural poor household. The deficiency in continuous treatment may be due to that raw materials of successful interventions are not affordable, production process are lacking, difficult in production due to lack of formal education, and that government have neglected the sponsorship or follow up of successful interventions. These reasons have possibly contributed to the status of the infestation as neglected disease. We report the use of grounded *P. guineense* in coconut oil ointment and naphthalene in kerosene ointment. Although, naphthalene and kerosene have been reported to be highly toxic substances (European Chemicals Agency (ECHA), 2020) but their combination have been in use in this community for some years with viable results to ameliorate the infestation without any side effect or any death recorded. This prompted its design into measured ointment as well as the grounded *P. guineense* in coconut oil ointment for testing to evaluate their efficiency and resulting effects during and after applications. The topical ointments were efficient ameliorating tungiasis lesions with no side effects and death occurring since the ointments were not administered through oral routes but used on the body part where tungiasis lesions threatened comfort. This comes as a surprise, as the potentials of these local ointments against tungiasis had been unknown so far in Nigeria and have not been tried in endemic areas of the world. Although,

the raw materials for the production of these local ointment are readily available as affordable substance around the world. The use of grounded naphthalene in kerosene showed no pain upon pressure in all stages of the embedded tungiasis fleas on the second day of local ointment applications. This representing 62%, 9% and 15% for stage I, II and III lesions respectively. From the third day of topical application, 94% of stage I lesion reduced pain upon pressure and dried up, and 100% dried up on the fifth day while 62% and 80% of stage II and III lesions reduced pain upon pressure and dried up, and 100% dried up on the sixth day. It is possible that tungiasis fleas were suffocated to death after resulting pain was not recorded in the various stages. All stages of tungiasis infestation on individuals were approximately zero on the sixth day for stage I and seventh day for stage II and III of Naphthalene in kerosene ointment applications respectively. The differences in days of treatment exposure and stage of tungiasis lesions was significant at $p < .05$ respectively. Evolution from stage I to II, stage II to III or eggs of ovipositioning flea was not observed during and two weeks after treatment. This shows that this ointment interrupted reproductive activities of embedded fleas.

However, the use of grounded *P. guineense* in coconut oil ointment reduced pain upon pressure in all stages of embedded tungiasis fleas on the second day for stage III lesions and third day for stage I and II respectively. On the fourth day of topical ointment applications, 56%, 95% and 74% of stage I, II, III lesions were dried up and showed no pain upon pressure. From the fifth day to sixth day, 100% of stage I lesion dried up while 100% of stage II and III lesions dried up on the seventh day. Buckendahl et al. (2010) had earlier indicated that the intermittent application of Zanzarin, twice daily for one week every second week was effective in interrupting *T. penetrans* transmissions and preventing severe morbidities. The result of this study is in accordance with their findings; as the topical ointments were applied twice daily and for two weeks exposure period. The residual effect observed after the application of Zanzarin for one week corroborate the result of Naphthalene and kerosene ointment. It was observed that there was a diminished residual effect in grounded *P. guineense* in coconut oil ointment. According to Feldmeier et al. (2004), this residual effect can persist for more than a week. Although, a break in the application of the repellent for about four weeks was observed as too long to prevent the invasion of tungiasis fleas into the skin (Buckendahl et al., 2010). The frequent application of the topical ointments in this study warded off tungiasis fleas and penetrations even though infestations were at peak season. Buckendahl et al. (2010) also noted that repellent can be prepared from local coconuts by individuals in the affected villages themselves with minimal input from the health sector. Their observation triggered the use of coconut oil in ointment formulation. It is equally not surprising because the coconut oil used as diluent of the grounded *P. guineense* was freshly milled from the village where infestations were observed high. The topical application of the local ointment caused drying up of 56% of stage I, 95% of stage II, and 73.9% of stage III lesions on the fourth day. Death of lesions occurred from the third day up to the seventh day as the reproductive activities of embedded fleas were equally interrupted by treatment. Evolution from stage I to II, stage II to III or eggs of ovipositioning flea was not also observed. The mechanism responsible for the death of embedded fleas were poorly understood and thus ascribed to the active components of the topical ointment. There was very high significant difference between stages and exposure days of participants assigned the two topical ointment $p < .0001$ and $p = .0005$ respectively.

Other treatment routes has been investigated. Oral dose with 50 mg/kg/day for 15–18 days as well as 0.8% ivermectin, 0.2% metrifonate, and 5% thiabendazole lotion were hypothetically effective (Cardoso, 1981; Heukelbach et al., 2003). The treatment with a combination of 10% imidacloprid and 50% permethrin (Advantix®) was effective against tungiasis lesions on dogs (Klimpel et al., 2005). The continuous use of these treatments may not be feasible since they are not available in local grocery stores close to endemic areas. Even though they are available, the question is whether the rural poor households can afford the cost of purchasing them and if the uneducated can stick to direction of use? As reported by Heukelbach et al. (2002), health education on the use of primary and secondary prevention would be of importance. Educating the head of any household with infestations on production method for these topical ointment would go a long way to disrupt developmental cycle in endemic settings. The disruption of developmental cycle of tungiasis fleas through sustained use of affordable treatment and the use of protective wears for a very long time would push tungiasis fleas into extinction if animal reservoir of infestation is equally tackled. Frequent self-extraction of tungiasis through surgical means could be risky. Thus, the assistance of a health facilitator is required.

Grounded naphthalene in kerosene ointment was best and effective against tungiasis lesions compared to the grounded *P. guineense* in coconut oil ointment. Although, there was no side effect in the physiology of participants that topically applied the ointments but cutaneous ossification was observed in individuals that used Naphthalene and kerosene ointment. It is probable that the ossification of feet and hands where the ointment was applied discouraged the further penetration of fleas. However, this was reversed within two weeks of post treatment exposures. Tungiasis morbidities were observably reduced in the Naphthalene and kerosene ointment group. Cutaneous ossification of affected part was not observed in individuals that used the grounded *P. guineense* in coconut oil ointment during and after topical applications on tungiasis lesions. In both ointment exposure groups, no adverse health-related reactions was observed in the participants but in a way to measure effectiveness of the topical ointments, the developmental cycle of embedded fleas was hindered in the observed reduced time. Thus, it was considered a good treatment approach. It is also probable that Naphthalene and kerosene ointment killed larvae of sand fleas embedded on affected hands and feet more than the grounded *P. guineense* oil in coconut oil ointment. Thus, with a promising ability of interrupting the reproductive cycle for re-infestation and evolutions.

5. Conclusion

The Naphthalene and kerosene ointment ameliorated tungiasis lesions rapidly with no re-infestation after weeks of topical applications. Associated morbidities especially pain upon pressure during the use of the topical ointments was observably low. The cutaneous ossification observed in individuals that used Naphthalene and kerosene ointment and not in the ground *P. guineense* in

coconut oil ointment predicts the barrier to penetrations by tungiasis fleas. Studies to justify and sustain these affordable treatments are required for future references.

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