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Epidemiology and Risk Factors for the Development of Leishmaniasis in Western Visitors to Honduras

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Introduction

Leishmaniasis is a major world health problem, which is increasing in incidence.[1] The leishmaniases mostly affect the world's poorest populations in the tropics and the subtropics with an estimated two million cases a year.[2] The major clinical syndromes are visceral, cutaneous and mucosal leishmaniasis, resulting from infection of macrophages in the reticuloendothelial system, skin, and nasal and oral mucosae, respectively.[3] In this research we will primarily be looking at cutaneous leishmaniasis which is a vector-borne protozoan disease caused primarily by the *L. braziliensis* and *L. mexicana* complexes in Central America.[4] The vector is the infected female phlebotomine sand-fly of the genus *Lutzomyia.*[5]

Cutaneous leishmaniasis skin lesions evolve over weeks to months starting as papules, which go on to form nodules and eventually ulcers with raised indurated borders which eventually heal with scarring. [6] Until recently cutaneous leishmaniasis has largely been neglected by researchers and funding agencies.

Although more work has been done in recent years the epidemiology of cutaneous leishmaniasis in civilian Western populations is still largely unknown. Cutaneous leishmaniasis in the Americas and in particular Honduras tends to affect persons working in lowland forest environments, particularly those conducting field studies[iii7,8] and increasingly tourists. In this study we set out to examine the epidemiology and risk factors involved in contracting the cutaneous form of leishmaniasis in Western visitors to Honduras.

Method

Subjects

One hundred and fifteen healthy Western volunteers who were conducting research with Operation Wallacea were the study population. The volunteers ranged in age from 16 to 30, gave written and verbal informed consent before the study was undertaken and were studied over eight weeks in five separate forest sites and two different beach sites. It was believed, through discussion with the ethics department of the University of Edinburgh and the College of Medicine, that ethical approval would not be required. This was due to the perceived nature of the study in that it was taking the form of an audit and we were not aiming to alter the volunteers' behavior. Alongside the information given by the researcher in the consent process recommendations were also provided to the participants by Operation Wallacea before the study on recommended vaccinations, malaria prophylaxis and rabies.

Questionnaires

A questionnaire was given to all the volunteers at week one to determine their prior knowledge about leishmaniasis. They were then split into separate groups with each of these groups being observed at their respective sites and examined for insect bites and non healing ulcers on days three and six of the study week. During this week a second questionnaire was given, to examine the various risk factors involved for the development of insect bites and cutaneous leishmaniasis.

Follow up and diagnosis of cutaneous leishmaniasis
The primary end point of the study was the diagnosis of cutaneous leishmaniasis by a dermatologist, GP or other specialist in the UK. Written information was given to the volunteers on measures to be taken if they developed a non-healing ulcer or a non-healing insect bite.

The secondary endpoint was a persistent, non-healing skin ulcer six weeks after returning to the UK. Follow up took place six weeks after the return of the volunteers to the UK and this was carried out by email, with requests for further information on risk factors in Honduras if cutaneous leishmaniasis was suspected or identified. All those who had a nonhealing ulcer at follow up sought medical advice.

Analysis and Publication
Comparisons between the risk factors for leishmaniasis especially insect bites were analysed alongside preventative methods undertaken by volunteers. These results have been subject to statistical analysis using the t-test. Results were issued to Operation Wallacea for development of better prevention and diagnostic procedures.

Results
Knowledge base on cutanous leishmaniasis
The results from the first questionnaire are seen below. Figure 1 shows the proportion of incorrect and correct responses to the multiple choice questions whilst the subjects' interpretation of their knowledge base on cutaneous leishmaniasis is shown in figure 3.

Figure 1

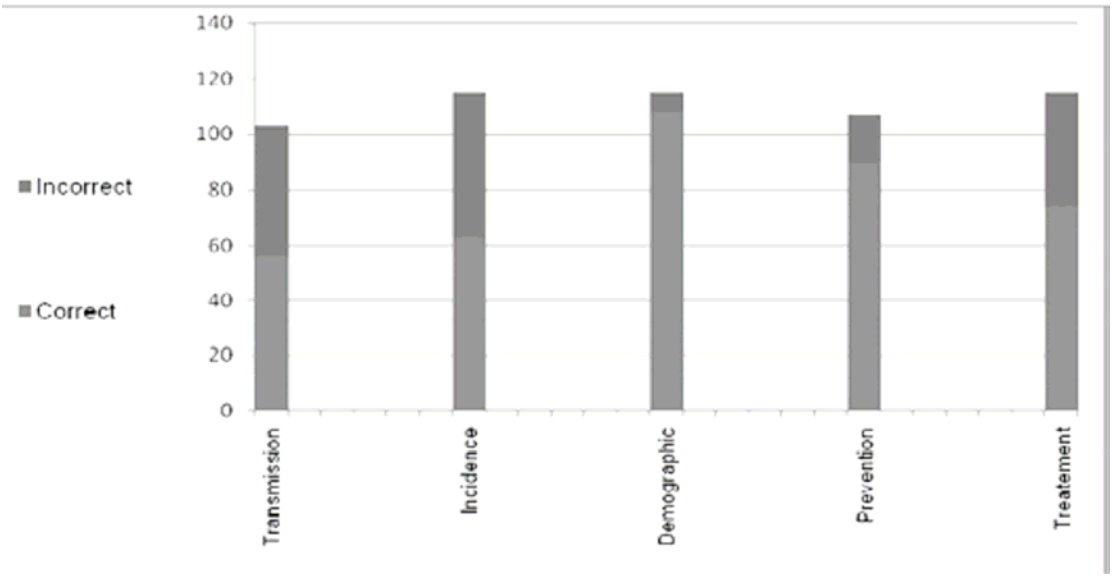

Figure 1. Results of questionnaire 1 showing the incorrect and correct responses to questions about the transmission, incidence, demographic, prevention and treatment of cutaneous leishmaniasis. n= 115

Incidence of Cutaneous leishmaniasis in study population
Eighty seven subjects were successfully followed up 6 weeks after return to the UK. Of these 87 subjects, six still had residual bites with five of those subjects fulfilling the secondary endpoint of a persistent, non-healing skin ulcer six weeks after return to the UK. There have been no episodes of cutaneous leishmaniasis amongst the study population at the time of write up.

Figure 2

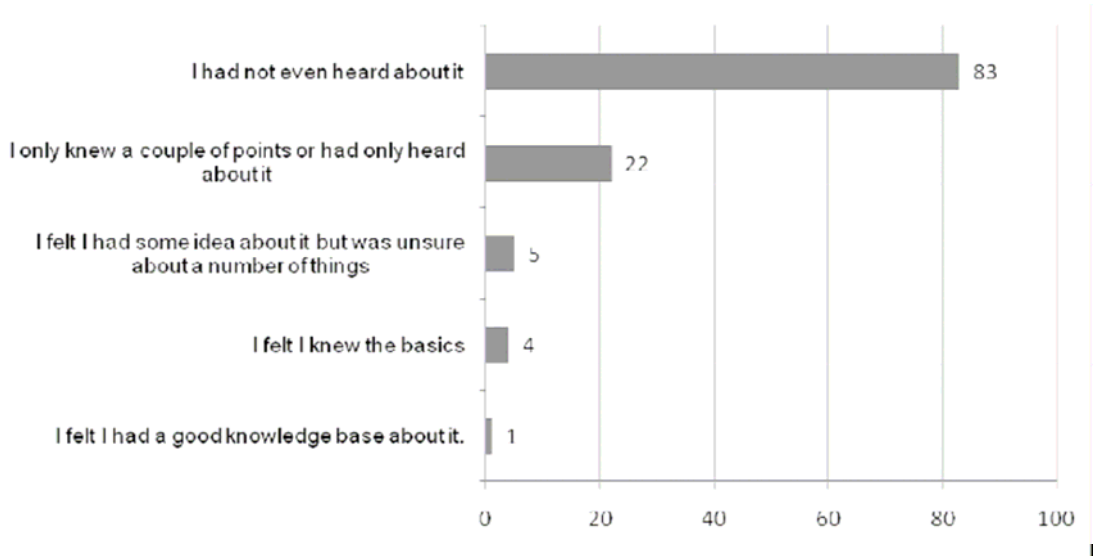

Figure 2. Subjects opinion of their knowledge base of cutaneous leishmaniasis before they commenced the study. n= 115.

Figure 3

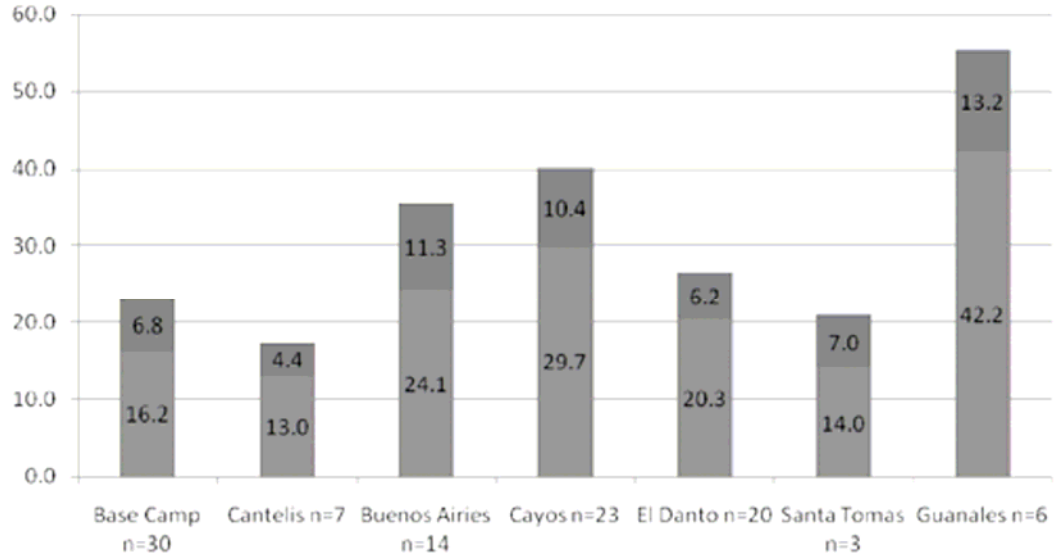

Figure 3. Inter site variability of insect bites. The number of lower limb bites (the bottom number in blue) and the number of upper limb bites (the top number in red) can be compared between the sites.

Risk factors

One hypothesised risk factor for cutaneous leishmaniasis was the number of lower limb insect bites. In recording the numbers of insect bites it was notable that there was site variability in the mean numbers of insect bites along with predominance for a greater number of lower limb insect bites compared with upper limb bites. Figure 3 demonstrates this. One of the preventative methods to prevent sand fly bites and therefore potentially

Figure 4

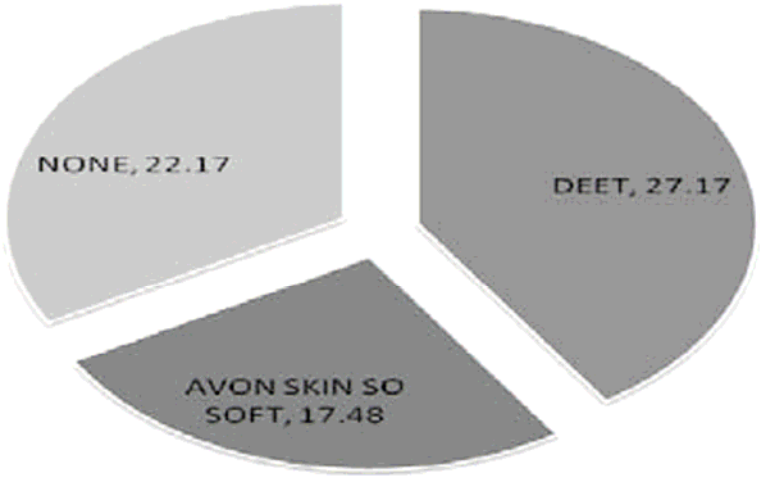

Figure 4. Mean number of insect bites in relation to insect repellant. The insect repellant DEET (N,N-diethyl-mtoluamide), the moisturiser Avon Skin-so-soft and the control of none (no insect repellant used) are shown.

cutaneous leishmaniasis is insect repellent. To investigate whether insect repellent has an effect on the number of insect bites the mean number of bites for each insect repellent within each site was calculated and the total outcome is shown below in figure 4.
This seemed to show that DEET (*N,N*-diethyl-*m*toluamide) and Avon Skin-so-soft did not incur any benefit in reducing the number of insect bites. This presumption was tested by using the t-test comparing the mean number of bites at each site, for subjects using the different methods of insect repellent. This produced p values for each of the preventative methods which are shown in the table below.

T test	*P value*
DEET vs. no insect repellent	0.331
Avon Skin-so-soft vs. no insect repellent	0.334
Avon Skin-so-soft vs. DEET	0.137

Discussion
Despite its relatively high incidence in the tropics and subtropics[ii] it is apparent that the subjects in this study did not feel that they knew much about cutaneous leishmaniasis. This can be seen from figure 2 with 72% stating that they had never heard of it previously. This figure may seem high when this is compared to the number of correct responses in the multiple choice questions. However it is likely that the high rate of correct answers to the multiple choice questions was due to knowledge acquired through the consent approach rather than a firm knowledge base. In addition the subject population was well educated and well versed in answering multiple choice questions potentially raising the correct answer rate. The primary end point was the diagnosis of cutaneous leishmaniasis by a dermatologist, GP or other specialist in the UK. After 6 weeks of follow up, no subject fulfilled these criteria. This could be explained by the following two factors.

1. Although there were a high mean number of bites, especially on the lower limbs (typical of sand flies see figure 3), there was no effective way in this study to correctly identify whether the sand flies were carrying *L. braziliensis* or *L. Mexicana* protozoa.
2. As we alluded to in the introduction, typically cutaneous leishmaniasis skin lesions evolve over weeks to months following exposure to the protozoan but longer periods of incubation have also been known.[6,10] Subjects were contacted a median of two months following their return to the UK but it may be the case that some cases are diagnosed subsequently.

The second endpoint was a persistent, non-healing skin ulcer six weeks after returning to the UK and five subjects met these criteria and are being followed up. The problems resulting from a short follow up programme would ideally be addressed in future studies in an attempt to fully quantify the risk. Alternatively case based follow up via tropical disease centres and travel clinics could provide better evidence on risk factors and localised incidence. Despite the fact that no subjects fulfilled the primary end point of the study there were a number of risk factors for cutaneous leishmaniasis categorised by the large number of bites on subjects' legs which is well demonstrated in figure 3.

Interestingly in this study, the insect repellents used by the study population have not shown to be of any benefit. Although it was hard to quantify how much and how often insect repellent was being used, the intention of every subject who applied the insect repellent was to prevent any insect bites, therefore demonstrating the intention to treat. The effectiveness of DEET has been widely tested especially by armed forces operating in the tropics and subtropics. A study in Panama comparing the skin applications of five selected repellents including DEET demonstrated a mean coefficient of protection (CP) of 99.2% against the attack of at least three sandfly species.[11] A further lab and field based study of a repellant soap containing DEET and permethrin against the phlebotomine sand-fly species in Columbia demonstrated that the soap gave up to 100% protection immediately after application but after a total of 12 hours the coefficient protection value had decreased to a median value of only 66.3% for females.[12] This highlights the potential problem with the application of DEET by the subjects in this study, as the majority despite having the intention to treat, only applied insect repellant when they remembered, resulting in a median period before reapplication of 12 hours.

– Conclusion –

Despite recent advances it seems that difficulty still remains in quantifying, controlling and treating Leishmaniasis. Awareness in the public and health care profession seems scarce and further advances in this field may depend on greater education and concentration of services. We believe further epidemiological studies may play an essential role and case based follow up studies may help us to gain a greater understanding of the risk for Western travellers. In the future we intend to carry out a large randomised controlled trial in this field looking at risk factors for insect bites and the relation of these to the incidence of Cutaneous Leishmaniasis.

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References:

1 Hepburn NC. Cutaneous leishmaniasis. *Clin Exp Dermatol*. 2000 Jul;25(5):363-70.
2 Clive R Davies, Paul Kaye, Simon L Croft et al. Leishmaniasis: new approaches to disease control. *BMJ.* 2003; 326(7385): 377–382.

3 Barbara L. Herwaldt; Susan L. Stokes; and Dennis D. Juranek . American Cutaneous Leishmaniasis in U.S. Travelers. *Annals of Internal Medicine.* 1993; 118; 779-784.
4 Grimaldi G Jr, Tesh RB, McMahon-Pratt D. A review of the geographic distribution and epidemiology of leishmaniasis in the New World. *Am J Trop Med Hyg.* 1989; 41:687-725.
5 World Health Organization. Control of the Leishmaniases. Geneva: World Health Organization; 1990. *World Health Organization Technical Report Series 79.*
6 Gerwaldt BL, Arana BA, Navin TR. The natural history of cutaneous leishmaniasis in Guatemala. *J Infect Dis.* 1992; 165:518-27.
7 Lainson R, Strangways-Dixon J.Leishmania mexicana: the epidemiology of dermal leishmaniasis in British Honduras. *Trans R Soc Trop Med Hyg.* 1963; 57:242-65.
8 Garnham PC, Lewis DJ. Parasites of British Honduras with special reference to leishmaniasis. *Trans R Soc Trop Med Hyg.* 1959; 53: 12-40.
9 Maguire JH, Gantz NM, Moschella S, Pan SC. Leishmanial infections: a consideration in travellers returning from abroad. *Am J Med Sci.* 1983; 285:32-40.
10 Makhoul K, Cutaneous leishmaniasis: recognition and treatment. *Am Fam Physician.* 2005; 71(4):663.
11 C. E. Schrec, D. L. Kline, B. N. Chaniotis et al, Evaluation of Personal Protection Methods against Phlebotomine Sand Flies including Vectors of Leishmaniasis in Panama, *Am. J. Trop. Med. Hyg.,* 1982; 31(5): 1046-1053.
12 Claudia F. Golenda, Victoria B. et al; Gender-related efficacy difference to an extended duration formulation of topical *N,N*-diethyl-*m*-toluamide (DEET). *Am. J. Trop. Med. Hyg.,* 1999; 60(4): 654-657.

Sand-Fly Vector