

Declining Visceral Leishmaniasis in Malta

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Abstract

Aims: To study visceral leishmaniasis (VL) trends in Malta.

Methods: Analysis of epidemiological and clinical trends, a veterinary questionnaire and questioning a canine laboratory testing facility.

Results: A decline in VL in the past 25 years (1980-2005) was noted for both paediatric ($p < 0.001$) and adult ($p = 0.002$) populations. No seasonal variation in infection rates was found. Serological testing was reliable and a useful adjunct to bone marrow diagnosis. Treatment with sodium stibogluconate was highly effective and there were no permanent sequelae associated with disease or treatment. Vets are encountering less canine VL with no differences in localities

Discussion: The decline in VL was paralleled by a decline in the stray canine reservoir population and by better management of canine infection in both sanctuaries and in domestic settings.

Key words

Leishmaniasis, epidemiology, Malta

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Introduction

Visceral Leishmaniasis (VL) is a chronic parasitic infection caused primarily by *Leishmania donovani* in the Old World, and by *Leishmania amazonensis* in the New World.¹ It infects approximately 400,000 individuals annually worldwide, with a predilection towards early childhood.² It is a chronic illness characterized by irregular fever, hepatosplenomegaly, pancytopenia and progressive weakness and emaciation, which can result in death if left untreated. VL is particularly important in the setting of coexistent and rising rates of Human Immunodeficiency Virus (HIV) infection and for example, in southern Europe 25-70% of adult VL cases are associated with coexistent HIV, and 1.5-9.5% of individuals with HIV suffer from newly acquired or reactivated VL.³⁻⁵ Paediatric cases have also been reported in which both organisms have been isolated.⁶

The Maltese Archipelago comprises the islands of Malta, Gozo, and Comino, and is situated in the center of the Mediterranean Sea, 93 km south of Sicily and 288 km north of North Africa. VL is endemic in the circum Mediterranean region, including Malta.

As in most endemic areas, the vast majority of affected cases occur in the childhood population.⁶ Notification of human cases of leishmaniasis was made compulsory in Malta in 1946, making this country an ideal location for epidemiological studies of VL. Unfortunately, notification is not compulsory for canine infections.^{7,8} The diagnosis of VL was first made in Malta in 1911,⁹ and the cutaneous form of leishmaniasis is also found.¹⁰ The vector is the female sandfly *Phlebotomus perniciosus* and dogs continue to be the local reservoir for human Leishmania infection.¹¹ Specific treatment for leishmaniasis was first instituted by Caronia of Catania in 1914, using a mixture of sodium antimony tartarate.¹² Pentostam has been used for the treatment of leishmaniasis since the 1940s,¹³ and the current recommended dose of Pentostam is 20 mg/kg/day for 30 consecutive days. In an earlier study, we noted a declining trend in paediatric cases of VL.¹⁴ In this study, we reviewed Leishmaniasis trends in Malta for all ages, with special focus on the paediatric age group, and relate this decreasing trend to present prevalence rates of infection in dogs and current treatment practices among Maltese vets.

Table 1: Annual number of human leishmaniasis cases

| Year | Total cases | Paediatric cases |
|------|-------------|------------------|
| 1980 | 9 | 8 |
| 1981 | 18 | 11 |
| 1982 | 4 | 3 |
| 1983 | 4 | 3 |
| 1984 | 7 | 5 |
| 1985 | 4 | 1 |
| 1986 | 8 | 5 |
| 1987 | 6 | 4 |
| 1988 | 16 | 8 |
| 1989 | 14 | 7 |
| 1990 | 15 | 5 |
| 1991 | 13 | 6 |
| 1992 | 10 | 3 |
| 1993 | 2 | 2 |
| 1994 | 3 | 3 |
| 1995 | 2 | 0 |
| 1996 | 3 | 3 |
| 1997 | 3 | 2 |
| 1998 | 6 | 2 |
| 1999 | 8 | 4 |
| 2000 | 7 | 5 |
| 2001 | 5 | 3 |
| 2002 | 0 | 0 |
| 2003 | 3 | 2 |
| 2004 | 4 | 1 |
| 2005 | 4 | 1 |

Methods

Patients: All Maltese cases of VL notified between 1980 and 2005 were identified from Malta Health Division notification registers. Paediatric patients were defined as those aged 14 years or under. Case notes of paediatric cases were retrospectively examined, and a form detailing presentation, diagnosis, treatment, and complications thereof was completed for each patient. Data was then entered into an Excel spreadsheet for processing. At least one non-Maltese paediatric patient contracted VL during a brief visit to Malta, and 2 patients were not included (stay <2 weeks in all cases).

Vets were sent a questionnaire in order to outline number of cases of canine VL encountered, by locality, and treatment of such cases. Data on canine Indirect Fluorescent Antibody Test (IFAT) testing was obtained from a local private laboratory for the period 2002-2005.

Definitions

A case of VL was defined as a patient with classical clinical features of VL and a positive bone marrow or a positive IFAT for VL. IFAT for VL has been available in Malta since 1989, and this test became widely used in the paediatric age group from 1990. A positive bone marrow aspirate was defined as a bone marrow sample that showed the intracellular amastigote forms LDB. IFAT was performed using Bio-Merieux substrate slides and Binding Site FITC conjugate. A strongly positive test was taken as one positive at a dilution of at least 1/180 (St Luke's Pathology Laboratory definition in conjunction with test manufacturers). In an extremely small minority of patients who presented with a clinical picture of VL but had a negative bone marrow aspirate in the era prior to the availability of IFAT, treatment for VL was instituted empirically, and confirmation of VL was reinforced by response to treatment. Relapse was also defined as above. Cure was based on clinical criteria, and in more recent years, confirmed by a negative IFAT eight to 12 months after cessation of treatment.

Population

Population totals and paediatric population totals for 1980-2005 were obtained from official Maltese Government publications.¹⁵

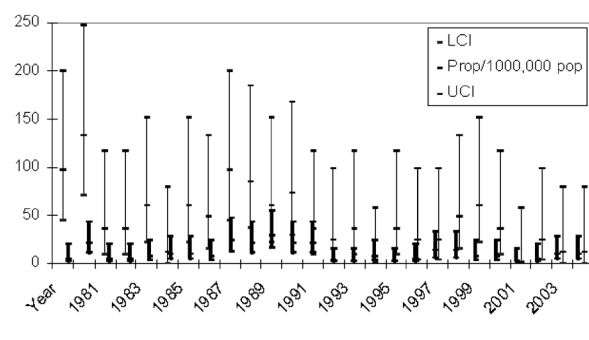
Veterinary study: A questionnaire was sent to a local veterinary representative who contacted all of the known practicing vets in Malta with a questionnaire. Questions asked included whether electronic records were kept by vets, the approximate number of dogs treated for leishmania every year, whether the severity of the disease has changed, and whether the disease is more prevalent in dog sanctuaries than in pets.

Lab study: Only one lab locally offered canine leishmaniasis testing up to 2006 (HSE) and only this lab was therefore included.

Statistics

Seasonal variation was analysed using Edward's method of cyclic variation.¹⁶ The quadratic equations of Fleiss were used to calculate 95% confidence intervals (CI) for sensitivity and

Figure 1: Rates of visceral leishmaniasis per 1,000,000 population (thin lines: adults, thick lines: paediatrics)



for population rates.¹⁷ Chi test for trends was used to analyse incidence over time. A *p* value of 0.05 or less was taken to represent a statistically significant result.

Results

Human VL

Epidemiology

The incidence of VL in Malta has decreased over the 25 year period studied (1980-2005: Table 1 and Figure 1). This was significant for both the paediatric ($p < 0.001$) and adult ($p = 0.002$) populations. No seasonal variation in infection rates was found ($p = 0.7$).

Investigations

Serology was positive in 21 cases and negative in one case of VL who had a positive bone marrow, an overall sensitivity of 95.1% (95% CI: 87.2 to 98.4%). Bone marrow aspirate failed to show LDBs in only four of the 81 cases, a sensitivity of 94.7% (95% CI: 86.2 to 98.3%). All four had repeated bone marrow aspirates, which were negative, but the clinical picture was highly suggestive of VL in all four. Treatment of VL was instituted in two cases (year of presentation 1996) that had positive IFAT. In two other cases, treatment for VL was instituted without a positive bone marrow owing to the unavailability of a serological test (year of presentation 1980). All four responded to treatment (see below). In four other cases, an initial bone marrow aspirate was negative, with subsequent positive bone marrow aspirates. There was no significant difference between the sensitivity of the two tests ($p = 1.0$). The sensitivity of combined bone marrow aspiration and IFAT was 97.5% (95% CI: 90.5 to 99.6%).

Treatment

In this study, treatment for VL in the paediatric age group with antimonials, a relatively inexpensive class of drugs, is safe and curative, with a low relapse rate and no permanent sequelae were associated with the disease or its treatment.

Lab study

Data on the number of IFAT tests performed in dogs from 2002 to 2005 show that positive tests for LV have not decreased

in the last five years (Dr. Alex Aquilina, HSE Laboratories, personal communication). These statistics are however difficult to interpret as repeat tests on the same dogs cannot be identified.

Vet study

Five vets answered the questionnaire. The general impression of vets practicing in Malta in response to a questionnaire was that they are encountering less canine VL than to five and ten years ago (Table 2). There were no significant differences in localities from where the infected dogs originated and vets who offered their services to dog sanctuaries did not report an increase in the incidence of the infection in dogs kept in sanctuaries. Dogs who presented with more than four symptoms were more likely to be put down as the cure rate in such dogs was poor.

Discussion

The incidence of VL in Malta has dramatically decreased over the 25-year period studied and indeed, in 1946, when human diseases became compulsorily notifiable on the islands, the leishmaniasis figures were 1264 visceral cases, 36 cutaneous cases and 5 unspecified.^{7,11}

This decline is attributed to the suppression of stray dogs due the establishment of several dog sanctuaries around the island, and the putting down or treatment of infected dogs with lifelong allopurinol.¹⁸

Although there are no official statistics regarding dog ownership in Malta, the general impression is that the Maltese are very keen dog owners and this is reflected by the ease with which dog owners sell litters of puppies and the frequent newspaper adverts related to sale of puppies in the local press.

The ultimate aim is prevention of VL, and the ideal way of achieving this is by the eventual development of an effective vaccine. This goal is particularly important in view of the emerging resistance to antimonial drugs in some parts of the world, and both of these should be the targets of future research efforts, along with compulsory veterinary notification of infected canines.

The main limitation of this study is the poor vet response

Table 2: Results of veterinary questionnaire

| | | |
|---|-----------------------|--|
| New cases seen annually | 20-30 cases ($n=4$) | 1-9 cases ($n=1$) |
| Compared to 5 years ago | 25-50% less ($n=4$) | No change ($n=1$) |
| Compared to 10 years ago | 50-75% less ($n=2$) | Unable to compare ($n=3$) |
| Is the disease milder | Yes ($n=4$) | No ($n=1$) |
| Any locality differences | No ($n=5$) | Yes ($n=0$) |
| Are more infected dogs seen in sanctuaries | No ($n=3$) | Do not practice in sanctuaries ($n=2$) |

rate, but this is unlikely to significantly bias our results as due to the small geographical nature of the island, individuals travel to all parts of the country in order to treat their pets.

Conclusions

In Malta, we have found a falling incidence of VL. This decline is attributed to better disease control in canines due to owner awareness of the condition, and its suppression/treatment, and widespread control measures for stray dogs. Current practice includes the putting down of dogs with more than 4 features of canine leishmaniasis, while 10 years ago, such dogs were consistently treated with very poor cure rates, thereby increasing the local leishmaniasis reservoir. Moreover, long-term allopurinol therapy is now routine, with low relapse rates.

Serological testing for VL is as sensitive as bone marrow aspiration. Treatment for VL in the paediatric age group with Pentostam, a relatively inexpensive drug, is safe and effective. To date, there appears to be very little resistance to this drug in Malta, and all patients were cured, with no disease or treatment related permanent morbidity or mortality. More expensive drugs, such as liposomal amphotericin B, should be reserved for cases which are resistant to treatment or which relapse after treatment.

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