Rev. sci. tech. Off. int. Epiz., 2006, **25** (3), 1111-1123

Medico-legal considerations of canine leishmaniosis in Italy: an overview of an emerging disease with reference to the buying and selling of dogs

A. Passantino

Department of Veterinary Medical Science, Faculty of Veterinary Medicine, University of Messina, Polo Universitario Annunziata, 98168 Messina, Italy

Submitted for publication: 5 April 2005
Accepted for publication: 8 February 2006

Summary

Leishmaniosis is a common infection in the canine population. Domestic dogs are the main reservoir hosts for zoonotic human visceral leishmaniosis in both the Old and New Worlds. In fact, canine leishmaniosis is not just a veterinary problem.

In the light of some recent advances in the field of diagnosis, the author evaluated the medico-legal aspects of buying and selling dogs that are potentially affected by leishmaniosis.

This paper clarifies why it is impossible to attribute redhibitory vice (an essential defect in a product which renders it useless or so diminishes its usefulness or value that it must be presumed that, if he had been aware of it, the buyer would not have bought it, or would have paid a lesser price) to this disease and highlights the necessity to improve Sicilian regional law no. 15 of 3 July 2000 concerning the creation of a dog register by giving every animal a health book.

With this aim in mind, the author suggests that leishmaniosis be included in the list of notifiable diseases that appears in article 1 of the Italian Veterinary Police Regulations so as to reduce the possibilities of fraudulent trade in dogs known to be infected before purchase.

Keywords

Dog – Leishmaniosis – Public health problem – Purchase.

Introduction

Canine leishmaniosis is an infection caused by the protozoan parasite *Leishmania infantum*. Symptomatic animals may be divided into different symptomatologic classes on the basis of their clinical signs. This clinical disease does not occur in all infected animals, but after the transmission season a variable population of subjects develops a humoral response in endemic areas (for example in Italy).

Domestic dogs are the main reservoir hosts (92) for zoonotic human visceral leishmaniosis (VL) in both the Old and New Worlds. In fact, canine leishmaniosis is a disease of great veterinary importance and a serious public health problem (33) (for example, in southern Europe, leishmaniosis in human immunodeficiency virus positive patients [27, 28, 29, 30, 34, 120] is currently a cause of great public health concern).

Leishmaniosis is included within the World Organisation for Animal Health (OIE) list of notifiable diseases (see

chapter 2.1.1 of the OIE *Terrestrial Animal Health Code* for the criteria for listing a disease [123]).

Several mammals are implicated as host reservoirs of the protozoa in the *Leishmania* spp. cycle, including wild and domestic animals. Among domestic animals, dogs are incriminated in the domestic transmission to human beings, mainly in cases of VL caused by *L. infantum* (62). The role of the dog as a reservoir host of human VL has been recognised since Nobel Prizewinner Charles Nicole discovered the disease in dogs in Tunisia in 1908.

The fact that canine leishmaniosis can be significantly controlled with deltamethrin collars (22, 23, 47, 59, 74, 75) could mean that other domestic species (e.g. cats) are involved in the epidemiology of the disease in endemic foci (47, 89, 111). However, the cat is generally considered an unusual host for *Leishmania* infection and case reports of feline leishmaniosis (FL) are only sporadic, originating mostly from places where the organism and different species of sand fly vectors are endemic (Brazil, Venezuela, Argentina, Algeria, Egypt, Ile de la Réunion, Jordan, Iraq, Vietnam, Italy, France, Spain, Portugal, Switzerland) (2, 8, 10, 13, 14, 15, 16, 21, 25, 31, 43, 46, 53, 54, 58, 64, 67, 68, 69, 76, 81, 82, 87, 88, 90, 91, 108, 112).

Very few aspects of FL have been studied; little is know about its prevalence and clinical manifestations, and it is not known which protozoan species are involved or how the parasite is transmitted to the vector. In addition, there is little information about the actual susceptibility and importance of cats in the transmission of *Leishmania* spp. (113, 114). All of these factors may contribute to the idea that FL is uncommon. In addition, it is important to emphasise that previous studies have shown that some species of biting phlebotomine sandflies are more attracted to cats than to dogs (46) and use them more frequently as blood-meal sources (72).

In humans, *L. infantum* causes VL and cutaneous leishmaniosis (CL). Desjeux and Alvar have described the two epidemiological forms of the visceral disease (which is generally more severe than CL) as follows: 'In the zoonotic form, as found in the Mediterranean basin (119), the parasites infecting humans have generally come, via a sandlfy, from dogs. In the anthroponotic form *L. donovani* which occurs, sometimes in severe and deadly epidemics, in East Africa, Bangladesh, India and Nepal, there is human to human transmission via the sandfly vector. In immunocompetent people, infection by the *Leishmania* parasites that can cause VL is not always followed by disease – some remain completely asymptomatic, many others have oligosymptomatic infections that resolve on their own, and perhaps only one in every five to ten develops clinical VL. Patients who develop the clinical disease, after an incubation period varying from a few weeks to several months, usually have fever, severe loss of weight, hepato- and/or spleno-megaly (splenomegaly being more frequent) and pancytopenia. If clinically evident and untreated, VL can be fatal, especially in developing countries, where associated diseases often occur' (30).

Molina *et al.* (66) identify three main forms of VL:

a) the zoonotic form of VL, caused by *L. infantum*, with infection predominantly in wild and domestic canids and occasionally in humans. This form, which is mainly endemic, extends from the Mediterranean through Central Asia into the People's Republic of China and also exists in Latin America;

b) the anthropo-zoonotic form, caused by *L. donovani* or *L. infantum*. It occurs in East Africa and the south-west of the Arabian peninsula. It is normally endemic but can cause extensive epidemics in humans. Transmission of the parasites that cause this form of VL is predominantly human to human via non-synanthropic vectors such as *Phlebotomus martini* and *P. orientalis*, although there may also be other mammals acting as reservoir hosts;

c) the anthroponotic form, caused by *L. donovani*, occurs on the Indian sub-continent. It is normally endemic but severe epidemics can develop among humans.

Currently, VL is considered by the World Health Organization to be an emerging zoonosis in southern Europe (121).

In Italy, VL is an endemic disease (38, 48), with higher incidence rates in the central-southern Tyrrhenian regions of continental Italy and in the islands. Traditional endemic territories also include central-northern (Tuscany and part of Emilia Romagna) and northern regions (Liguria) (124). Since the early 1990s, VL cases have been increasing throughout the country, with about 200 cases recorded in 2001 (36).

The dog is considered the domestic reservoir of the infection and phlebotomine sandflies are the only proven vectors of leishmaniosis for dogs and humans. Canine leishmaniosis is widespread in the Mediterranean sub-region (37, 116). In Italy, canine leishmaniosis is highly endemic along the Mediterranean coasts and the islands (55, 121, 124); however, several autochthonous cases are also being reported in northern regions of the country, probably due to climatic changes (35).

The first case of leishmaniosis in dogs was discovered in Sicily by Basile (1910) almost a century ago (9). Since then, on the basis of some epidemiological studies (whose results have been repeated over time) and on the acquisition of recent epidemiological data, this parasite has proved to be 'endemically constant' in Sicily (19, 20, 70, 77, 84, 95). This suggests that it is opportune to consider

some legal aspects inherent to the purchase and sale of dogs coming from this territory and which are therefore potentially infected. In particular, in the case of absence of symptomatology, the author thinks it would be useful to evaluate when recent parasitological, serological or biological-molecular techniques should be employed to discover a latent disease, latency of a former infection or an asymptomatic carrier.

The paper concludes by considering the value attributable to the results of various diagnostic studies in the question of 'legal vice' with regard to this disease.

Diagnostic techniques

Accurate and rapid diagnosis of canine leishmaniosis is of great importance in order to start early treatment and to prevent transmission, but this remains problematic. The demonstration of *Leishmania* amastigotes in Giemsa-stained smears of lymph node or bone marrow aspirates or in blood, or the detection of promastigotes in cultures from these materials, are the traditional and simplest methods of diagnosis (86). Moreover, the detection by culture is the only undisputable test.

Detection of anti-*Leishmania* circulating antibodies (mainly IgG, especially IgG1, IgG3 and IgG4) (44, 98) using serodiagnostic techniques constitutes an essential tool in the diagnosis of canine leishmaniosis (26).

In general, the most satisfactory techniques are (122):

– immunofluorescent antibody test (IFAT) (which is considered the gold standard)

– enzyme-linked immunosorbent assay (ELISA) (5)

– direct agglutination test (DAT) (42).

Part 2, Section 2.2, Chapter 2.2.11 of the *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals* 2004, says: 'Serology is the preferred method for diagnosis of canine leishmaniosis and VL, even during the early stages of the disease. In subclinical forms, seropositive cases are confirmed by parasitological diagnosis or polymerase chain reaction (PCR). Serology is of less value for CL and MCL [mucocutaneous leishmaniosis]. Of the several serological techniques available, the indirect fluorescent antibody test and the ELISA are the most suitable. Serodiagnostic antigens need to be prepared in the laboratory, though some commercial products are now under evaluation' (123).

Although IFAT is the most widespread diagnostic method it does have limitations, the main ones being cross-reactions reported in individuals infected with *Trypanosoma cruzi* (122) and low sensitivity in detecting asymptomatic dogs. Cross-reactivity with other canine parasites and bacterial pathogens has also been reported for ELISA (57, 103).

Immunochromatographic dipstick tests for *Leishmania* diagnosis have recently been developed and are all based on recombinant K39 (rK39), a protein predominant in *L. infantum* and *L. donovani* tissue amastigotes (17). These dipstick tests have been shown to be quite sensitive (reported sensitivities, 67% to 100%) and very specific (reported specificities, 97% to 100%) when tested on Kala-azar patients (11, 24, 45, 117, 127), with results similar to those of rK39-ELISAs (1, 7, 17, 49, 80, 96, 115, 125, 126). Preliminary studies have shown that this test was also suitable for the serodiagnosis of canine leishmaniosis (63, 65, 79, 80, 104, 105, 125).

Several molecular biological techniques have been developed for the sensitive detection and identification of pathogens (6, 12, 37, 40, 41, 50, 51, 52, 60, 93, 94, 99, 118). The main approaches to nucleic-acid-based detection are hybridisation using DNA probes and amplification techniques, including reverse transcription (RT)-PCR for the detection of RNA and PCR for the detection of DNA (39). This technique is based on the amplification of a known, specific sequence using oligonucleotide primers that bind specifically to DNA flanking the region of interest. The target sequence is then amplified using a heat-stable DNA polymerase. The PCR products can be visualised with standard molecular biological methods, such as gel electrophoresis and Southern blotting. A number of different PCR assays have been developed for the detection of *Leishmania* DNA in clinical specimens, using several targets (61, 70, 71, 78, 101, 106, 107).

Illness qualifying as vice in veterinary legal medicine

In veterinary legal medicine, illness, although it is not specifically mentioned in the Italian Civil Code, must be considered a vice in those cases where it renders an animal unsuitable for its specified use or significantly reduces its value.

An animal which is unsuitable for its specified use must be affected with a behavioural 'character defect' or an 'illness'. A behavioural 'character defect' is a defect (as opposed to a virtue) which makes the animal not only less productive or of lesser value but also, sometimes, useless and dangerous A character defect involves senses, perceptions, affections, will, instincts and memory, while 'illness' causes a disturbance in normal organic functioning (which may be

localised or generalised) following an anatomical alteration which will inevitably cause an appreciable permanent or temporary impairment. To qualify as vice, however, illness must be 'serious' or 'chronic', 'pre-existing' and 'not easily recognised'.

Illness is 'serious' when the animal cannot be used immediately, or when it reduces the animal's value to a measurable extent. It is 'chronic' when it is not a temporary state, but rather its seriousness and non-curability persist over time. It is 'pre-existing' when it was present before the contract was made and continues to be so after sale. It is 'not easily recognised' if it cannot be observed by a diligent professional, that is, by the *bonus pater familias* (article 1176 of the Civil Code) (86).

Current law

In Italy, the purchase of animals is regulated by the Civil Code (article 1470 and following) and dates back to 1942. These articles regulate the purchase of all animals in general, and they are also adopted in the field of the purchase of dogs. In fact, in Italy, the Civil Code (article 812) considers animals as a *res*, nowadays, i.e. a thing (as property) as opposed to a person that is the subject of rights.

The guarantee against vices in the sale of animals is regulated by article 1496 of the Civil Code, which will take into consideration special laws concerning vices in different fields. However, as these laws have not as yet been drawn up the regulation of the matter is entrusted to 'local usages' (i.e. the ways in which one can justifiably expect a matter to be dealt with because that has been the practice in that community for a long time) and, if these are lacking, to article 1490 of the Civil Code and following. The latter reads as follows: 'the seller is obliged to guarantee that the object sold is immune from vices which make it unsuitable for the usage to which it is destined or that decrease the value of it in an appreciable way'.

The buyer, in his turn – only when the animal is affected by a serious or chronic vice that is pre-existing and not easily recognisable – will be able, with respect to the terms of expiry and to rules contained in article 1495 of the Civil Code, to exercise one of the two legal actions foreseen by the Code, namely, redhibitory action or estimatory action. 'Redhibitory action' (*actio redhibitoria*) means rescission of sale and 'estimatory action' (*actio estimatoria* or *quanti minoris*) means reduction of price.

Local usages in the matter of buying or selling dogs are set out in a text deposited at each Chamber of Commerce and it is to be noted that the 9th Chapter of article 935 of the Civil Code allows these texts to provide for the possibility of sale with the exclusion of guarantee. According to this article, if the dog is 'sold on the spot' or the seller declares 'I am selling it as it is', whenever, following the sale, the buyer observes the presence of a vice in the purchased animal, he will not be able to make any claim for the restitution of the amount paid (redhibitory action). In the same text, so as to limit pointless litigation, a list of vices has been drawn up which, because they are considered serious, chronic, pre-existing and not easily recognisable, permit the purchaser to legitimately exercise a redhibitory or estimatory action; leishmaniosis is not included in this list, except in the province of Turin.

Considerations

Leishmaniosis is a sword of Damocles, constantly present hanging over the purchase and sale of dogs in Sicily. Given the widespread awareness of the disease among people who work in this field it is the cause of justified fears in the purchaser and an object of necessarily partial reassurances on the part of the seller.

The author is inclined to share the prevailing opinion which would exclude leishmaniosis from the list of diseases indicated as redhibitory vices in the dog on the basis of the following considerations:

a) Even though there are numerous possible tests for the diagnosis of canine leishmaniosis, at the present time there is not a single diagnostic test available which reaches levels of 100% for both specificity and sensitivity, therefore, it appears impossible to exclude with absolute certainty that the animal could be host to the parasite. The test that comes closest to meeting these requirements is PCR carried out on the bone marrow.

All the diagnostic techniques, even the most sophisticated ones such as PCR or immunofluorescence assay (the gold standard), are of limited use for legal purposes: they can identify infection, but this is not recognised by law as reason enough to reduce the value of an animal. In the eyes of the law only clinical disease makes an animal unsuitable for use and thus less valuable.

In reality, for obvious reasons, this is most problematic in the case of buying and selling asymptomatic or oligosymptomatic animals which have given positive results to a test. In such a difficult situation, it will be necessary to carry out a complete clinical check which takes into account the variform aspect of the parasite and verifies all the information needed to identify the disease in its different evolutive stages, e.g. the velocity of the erythrosedimentation, serum electrophoresis, the main

parameters of hepatic and renal functionality. If one succeeds in doing this there should be no interpretative problem. However, quite often clinically and organically healthy animals will be encountered, as has been shown by numerous recent epidemiological studies (18, 56, 57, 83, 97, 100, 102, 104, 109, 110).

b) Cases are recorded in which experimentally or naturally infected dogs, even after serum conversion, have overcome the infection spontaneously, not developing symptoms or showing any paucisymptomatic self-resolving forms.

It has been experimentally shown that the incubation and prepatent periods for this are extremely variable, ranging from a few weeks to several months or years. It is clear that it is not possible to fix a certain lapse of time as a guarantee.

As regards serological studies, given that it can take months for serum conversion to occur, it would be impossible to show whether the infection has been contracted prior to or after the sale of the animal. Besides, there is no agreement among the literature as to the titre that must be considered positive for IFA. Therefore, one of the essential requirements for the recognition of legal vice would be missing, that is, the pre-existence of the vice. Furthermore, it must be kept in mind that there are animals that develop symptoms prior to the serum conversion or that do not produce any antibodies at all despite manifesting the disease.

As it is known that animals can overcome the infection spontaneously over different periods of time, the requirement of 'serious damage' would also be missing.

This controversial diagnostic situation, which originates from the interaction between the defence mechanisms of the parasite and the anti-*Leishmania* immunity reactions started by the host, demonstrates how complex the factors are that concur in the resistance to the disease and why all the multible possible evolutions of this infection are not rigorously described in the literature. In fact, it has already been shown both in rats and in man that the development of the disease or the spontaneous overcoming of the infection will depend most of all on the characteristics of the lymphocytic subpopulations that are activated by the encounter with the parasite (32, 73, 85), and in such an event it seems that individual genetic factors play an important role. It is reasonable to suppose that between these two extremes a great number of intermediary forms exist.

Table I
An example of a purchase contract

Between the undersigned Mr/Ms _____, the seller, and Mr/Ms _____, the buyer, the following is stipulated.

Mr/Ms _____ will sell to Mr/Ms _____ the following animal:

Canine species _____, breed _____, age _____, gender _____, coat _____, particular signs _____, microchip no. ____, warranting dispossession and the transferring of the ownership of the animal. In addition, the seller will give the buyer a registered certificate of origin as soon as possible.

The buyer has paid _____ as a deposit and he will take possession of the animal on __ __ __.

Signing this contract, the seller relinquishes ownership of the animal.

The seller guarantees that the animal is healthy and will provide a veterinary certificate. This certificate reports the state of the animal's health, as confirmed by Dr._____ on ___ ___ ___.

The seller guarantees that the sold animal is without any vices, malformations or diseases which would make it unsuitable for the use it is destined for or decrease its value in a considerable way. This guarantee is valid for one year from the date of delivery of the animal. When this term has passed and in the case of acts of ownership not explicitly agreed upon or authorized by the seller, he will be deemed free from the obligations underwritten in this document.

The buyer will pledge to maintain the animal as bonus pater familias with the obligation to inform the seller at once of any sign of illness in the animal.

Any controversy which should arise between the parties, resulting from the present contract and in particular as regards interpretation, execution, fulfilment, breach, rescission of the present contract, will be resolved by a group of Arbitrators consisting of three members, two nominated by each of the parties and the third by the first two nominees, in the case of disagreement, the case will be referred to the President of the relevant local Court.

The group of Arbitrators will take a decision in accordance with the procedures of informal [a] arbitration and its decision will be irrevocable.

Date _____

Mr/Ms _____ (the buyer) Mr/Ms _____ (the seller)

a) In Italy there are two types of arbitration: customary and informal. Customary arbitration gives the parties the opportunity to file the arbitrators' decision before a court so that it has the same effect as the decision of a judge. Informal arbitration is not subject to regulations; it is an agreement by which the parties agree to abide by the decision of an independent arbitrator

It should be remembered that the disease can be transmitted congenitally in dogs, this is rare, but it could nevertheless cause problems when puppies are sold.

Conclusions

On the basis of all that has been mentioned above, leishmaniosis, in the opinion of the author, cannot be considered a redhibitory vice.

However, the disease is characterised by extremely variable prepatent and/or incubation periods, and because of the limitations of the available tests it is difficult to exclude a diagnosis of leishmaniosis redhibitory vice with absolute certainty or to forecast how an individual case of infection will evolve; this means that asymptomatic animals can be bought and sold and this can cause civil quarrels if the animals later develop the disease.

In areas at risk from the parasite, the author suggests that in order to increase the probability of identifying asymptomatic and paucisymptomatic animals and to warn purchasers of the potential onset or reoccurrence of the disease, a neutral veterinarian should carry out the necessary investigations on the animal in question.

The author advises the parties involved to write a purchase contract (Table I).

The author advises the buyers to ask for the assistance of a trustworthy veterinarian who will evaluate the health status of the dog before the sale takes place. In this way, disagreements between the parties are avoided.

The author proposes that leishmaniosis be included in the list of diseases contained in article 1 of the Veterinary Police Regulations (3), which refers to the obligation to report cases of infectious diseases.

Sicilian regional law no. 15 of 3 July 2000 (4), which derives from national law 281/91, recommends the introduction of a special individual health book in which it would be obligatory for veterinarians to record all cases of the disease that they treat. This innovation, besides the doubtless health advantages of controlling the parasites present in the territory, would make the fraudulent trade of infected dogs more difficult, thus giving greater protection to the purchaser.

Acknowledgements

The author thanks Dr C. Keir for her kind correction of the English language of the manuscript. She is also grateful to Professor O. Catarsini and Professor M. Passantino for their invaluable assistance.

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Considérations médico-légales sur la leishmaniose canine en Italie : aperçu d'une maladie émergente liée à l'achat et à la vente de chiens

A. Passantino

Résumé

La leishmaniose est une maladie courante des chiens. Les chiens domestiques constituent le réservoir principal des leishmanioses viscérales zoonotiques affectant l'homme dans l'Ancien comme dans le Nouveau Monde. En réalité, la leishmaniose canine n'est pas seulement un problème vétérinaire.

À la lumière des dernières avancées dans le domaine du diagnostic, l'auteur évalue quelques aspects médico-légaux liés à l'achat et à la vente de chiens potentiellement atteints de leishmaniose.

L'article explique pourquoi cette maladie ne peut entrer dans la catégorie du « vice rédhibitoire » (défaut essentiel, rendant un produit inutilisable ou diminuant sa valeur d'usage ou vénale, et par rapport auquel on est en droit de considérer que si l'acheteur avait eu connaissance de son existence, il n'aurait

pas acheté le produit, ou l'aurait payé moins cher), tout en soulignant la nécessité d'améliorer la législation sicilienne (loi n° 15 du 3 juillet 2000 relative à la création du registre canin) en rendant obligatoire le carnet sanitaire individuel pour chaque chien.
Dans cette visée, l'auteur suggère d'inscrire la leishmaniose sur la liste des maladies à déclaration obligatoire, comme stipulé à l'article 1 du règlement italien de police vétérinaire, afin de limiter les possibilités de vendre illicitement les chiens reconnus infectés.

Mots-clés
Achat – Chien – Leishmaniose – Problème de santé publique.

Aspectos médico-legales de la leishmaniosis canina en Italia: panorámica de una enfermedad emergente, con referencia a la compra-venta de perros

A. Passantino

Resumen
La leishmaniosis es una infección común en la población canina. Los perros domésticos son el principal reservorio de la leishmaniosis visceral humana, afección zoonótica presente tanto en el Viejo como en el Nuevo Mundo. De hecho, la leishmaniosis canina es más que un mero problema veterinario.
Teniendo en cuenta una serie de recientes avances en el terreno del diagnóstico, el autor estudió los aspectos médico-legales que se plantean en la compra-venta de perros que pudieran estar infectados.
La autora aclara los motivos por los que es imposible achacar a esta enfermedad un vicio redhibitorio (defecto esencial que inutiliza un producto o le resta utilidad o valor hasta el punto de que el comprador no lo habría adquirido o habría pagado un precio inferior de haber conocido su existencia), e insiste en la necesidad de mejorar la ley regional siciliana nº 15 del 3 de julio de 2000, relativa a la creación de un registro de perros con la entrega de un carné sanitario a cada animal.
Con tal objetivo en mente, la autora propone que se inscriba la leishmaniosis en la lista de enfermedades de notificación obligatoria que figura en el primer artículo del Reglamento de Política Veterinaria italiano, lo que reduciría las posibilidades de comercio fraudulento con perros cuya condición de infectados fuera conocida en el momento de la transacción.

Palabras clave
Adquisición – Leishmaniosis – Perro – Problema de salud pública.

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