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Imported strongyloidiasis in Spain

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SUMMARY

Objectives: The objective of this study was to assess the epidemiological, laboratory, and clinical features of imported strongyloidiasis in a tropical medicine referral unit in Madrid, Spain.
Methods: This was a retrospective study based on a review of medical records. A patient was diagnosed with strongyloidiasis when the infection could be detected by conventional stool analysis and/or serology against *Strongyloides stercoralis*, regardless of the presence of symptoms.
Results: One hundred and seventy-eight cases of strongyloidiasis were included in the study. Stool tests were performed in all patients, and serology in 160 patients (89.9%). The diagnosis of strongyloidiasis was based on serology only in four patients; 21 patients only had positive stool tests. A third of the total strongyloidiasis cases in this study were travel-related, mainly associated with short trips (<2 months). Only 47.8% of total cases were symptomatic. We found no differences in clinical presentation between immigrants and travelers with strongyloidiasis.
Conclusions: Not only should strongyloidiasis be suspected in symptomatic travelers and immigrants, but it should also be ruled out when elevated IgE levels or eosinophilia are present. Strongyloidiasis can be asymptomatic in HIV patients, but it should be diagnosed and treated before a possible hyperinfection develops.

1. Introduction

Strongyloidiasis is caused by *Strongyloides stercoralis*, an intestinal nematode. It is usually acquired by walking barefoot on infested soil, and is an endemic infection in the tropics and subtropics. The worldwide prevalence is estimated at between 3 million and 100 million.[1] Autochthonous cases have been reported in Spain, mainly in the Mediterranean area,[2–4] but data from cases in immigrants and travelers are scarce.[5–7] Other foci in Europe have also been reported.[8–10]

Strongyloidiasis is one of the most difficult parasitic diseases to diagnose, because there is no gold standard for this purpose. It can be suspected in symptomatic patients with digestive, respiratory, or cutaneous complaints; however asymptomatic eosinophilia and even 'silent' infections have also been described. Traditional diagnostic methods are based on the visualization of *S. stercoralis* in stools and the demonstration of antibodies by serology, but the sensitivity and specificity can vary in different groups of patients.

Hyperinfection and disseminated infections can be fatal in immunosuppressed patients (transplant recipients and those on corticosteroid treatment). Focusing on HIV infection, the prevalence of this co-infection is variable;[11–13] the most frequently manifested symptoms are chronic diarrhea, fever, cough, and unintentional weight loss. *S. stercoralis* in persons infected with human T-cell lymphotropic virus type 1 (HTLV-1) is highly associated with parasite dissemination and the development of severe strongyloidiasis. These co-infected patients have a modified immunological responses against parasite antigens.[14,15]

Several questions remain to be answered related to imported strongyloidiasis. How common is strongyloidiasis linked to travel? What countries are the main sources of infection? Should travelers and immigrants be tested routinely for strongyloidiasis? The main

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objective of this study was to assess the epidemiological, laboratory, and clinical features of imported strongyloidiasis in a tropical medicine referral unit in Madrid. Other goals were to describe the diagnosis method and to evaluate the differences between two groups: immunocompetent and immunosuppressed patients.

2. Methods

Hospital Carlos III is a referral unit for tropical diseases in Madrid, Spain. Most patients come by themselves to the emergency unit or are referred from primary care or general hospitals in Madrid. A very small proportion of patients come from other regions.

A retrospective study based on a review of the medical records of adults who attended Hospital Carlos III between January 1, 2007 and December 31, 2011 was performed. Patients with positive parasite samples for *S. stercoralis* or positive serology against this parasite were identified through the databases of the Microbiology Department and the Tropical Diseases Unit.

Exclusion criteria were: (1) unspecified diagnosis methods, and (2) medical records with a lack of data (>25% items): epidemiological data (>5 items), clinical data (>5 items), and analytical data (>7 items).

A patient was diagnosed with strongyloidiasis when the infection could be detected by conventional stool analysis and/or serology against *S. stercoralis*, regardless of the presence of symptoms. Countries considered endemic for Strongyloides were those on the map published by Stanford University.[16]

Cases of strongyloidiasis were defined as: (1) autochthonous, when diagnosed in a person who had never travelled to a country endemic for Strongyloides; (2) traveler, when a person was diagnosed after travelling to a country endemic for Strongyloides; (3) native, when a person was born in a country endemic for Strongyloides.

For each case, demographic, clinical, and laboratory data were documented (see Table 1).

Countries were classified as follows: Africa includes the World Health Organization (WHO) African Region, Egypt, Libya, Morocco, Somalia, Sudan, and Tunisia. Asia includes the WHO South-East Asia Region, Western Pacific Region (excluding Australia), Afghanistan, and Pakistan. Central and South America includes the WHO Region of the Americas, excluding the British Virgin Islands, Canada, and the USA.

Serum samples were tested for the qualitative screening of IgG antibodies to *S. stercoralis* using an ELISA technique (DRG Strongyloides IgG ELISA). The microtest wells were coated with Strongyloides antigen. One hundred microliters of diluted serum (1/64) was dispensed into the wells and incubated for 10 min at room temperature. Next the wells were washed three times with the washing buffer provided, 100 μl of protein A–peroxidase conjugate was added, and the mixture was incubated for 5 min at room temperature. After washing and removing excess moisture, 100 μl of tetramethylbenzidine was then dispensed into each well. After incubation at room temperature, the reaction was stopped by the addition of 100 μl of 1 M phosphoric acid. A negative control and a positive control provided by the manufacturer were included in each assay. The reading of the plates was carried out at 450 nm/620 nm, subtracting the blank from all wells. In this study we used a cut-off value of 0.200. A test was considered positive if the index (ratio of the OD measure of the sample and OD measure of the cut-off) was >1.1.

Stool samples were tested by microscopic examination of the stool issued on three consecutive days and by blood–agar culture. Microscopic diagnosis was based on the observation of larvae in stool samples (samples were treated in a Mini Parasep SF Faecal Parasite Concentrator). For the blood–agar culture method, stool samples were placed on a blood–agar plate and incubated for 7 days. As the larvae crawl over the agar, they carry bacteria with them, creating visible tracks.

Data were analyzed using SPSS for Windows v. 17.0 (SPSS Inc., Chicago, IL, USA). For the univariate analysis of categorical variables, Pearson's Chi-square test was used (Fisher's test when needed). The Mann–Whitney *U*-test was used for quantitative variables. A p-value of <0.05 was considered significant.

3. Results

3.1. General features

One hundred and seventy-eight cases of strongyloidiasis were included in the study. Fifty-eight cases (32.6%) were classified as travelers and 120 (67.4%) as natives. There were no autochthonous cases. The main features of all the strongyloidiasis cases are shown in Table 2. Equatorial Guinea was the main country where the infection was acquired in natives (40.8%), followed by Bolivia (24.2%) and Ecuador (11.7%). The countries visited by the travelers group were very heterogeneous (Table 2). Reported countries visited in Africa were Algeria, Angola, Benin, Burkina Faso, Burundi, Cameroon, Chad, Central African Republic, Congo, DR Congo, Côte D'Ivoire, Equatorial Guinea, Gabon, Kenya, Malawi, Mali, Morocco, Mauritania, Namibia, Nigeria, Rwanda, Senegal, Tanzania, Togo, and Zimbabwe. Countries visited in Asia were Afghanistan, Bangladesh, Cambodia, Malaysia, Nepal, Thailand, and Vietnam. Countries visited in America were Argentina, Bolivia, Colombia, Cuba, Costa Rica, Dominican Republic, Ecuador, Guatemala, Haiti, Honduras, Jamaica, Mexico, Nicaragua, Peru, and Venezuela.

3.2. Symptoms, laboratory abnormalities, and comorbidities

Clinical features of strongyloidiasis are shown in Table 3, and laboratory abnormalities can be seen in Table 4 and Figure 1. An asymptomatic infection with eosinophilia $>700 \times 10^6$ cells/l was present in 28% of cases. Eosinophilia was higher (45.2%) in asymptomatic patients when the cut-off was lower (e.g. $>500 \times 10^6$ cells/l). An asymptomatic infection with normal IgE levels and with normal eosinophil counts was described in 27 cases (15.2%). An asymptomatic infection with normal IgE levels and

Table 1
Data collected[a]

Demographic data	Age; sex; place of birth; place of residence; last endemic zone for Strongyloides visited; dates of travel (traveler); date of arrival to non-endemic zone for Strongyloides (native)
Clinical data	Presence of symptoms (yes/no); date of onset of symptoms; presence of urticaria, larva currens, purpura, nausea, vomiting, reflux, dyspepsia, abdominal pain, constipation, diarrhea, cough, sputum, wheezing, comorbidity
Laboratory data	Hemoglobin (g/dl); white blood cells ($\times 10^9$/l); total eosinophil count ($\times 10^6$/l); percentage of eosinophils (%); platelet count ($\times 10^9$/l); serum level of immunoglobulin E (IU/ml); HIV, HBV, HCV, and HTLV[b] antibody testing

HIV, human immunodeficiency virus; HBV, hepatitis B virus; HCV, hepatitis C virus; HTLV, human T-cell lymphotropic virus.

[a] Clinical definitions: anemia: hemoglobin <13 g/dl (12 g/dl in females); leukopenia: white blood cells $<4 \times 10^9$/l; thrombocytopenia: platelets $<150 \times 10^9$/l; absolute eosinophilia: $>700 \times 10^6$ eosinophils/l; relative eosinophilia: >7% eosinophils.

[b] HTLV antibody testing is not routinely performed in clinical practice.