Parasitic Infections in HIV Patients in Austria: 
First Results of a Long-term Study*

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Abstract
In the course of a long-term study of parasitic infections among HIV-infected persons in Austria during the period from November 1985 until May 1989, 618 persons infected with HIV (including 270 hospitalized patients, most of them with severe symptoms of AIDS) were examined. 58% of all persons had antibodies against Toxoplasma gondii. The incidence of clinically overt toxoplasmosis was about 20% in the 167 hospitalized persons infected with the parasite. In 29% of 68 patients with suspected pneumocystosis, the infection could be verified. In 9% of 219 patients, Cryptosporidium sp. was found. In two persons, an infection with Strongyloides stercoralis was diagnosed. Except these AIDS-associated opportunistic infections, the incidence of parasitic infections in the Austrian HIV-infected population was found to be low, and, except for Entamoeba histolytica, not significantly exceeding the frequency of parasitic infections in non-HIV-infected Austrians.

Zusammenfassung
Im Rahmen einer Langzeitstudie über parasitäre Infektionen bei HIV-Positiven in Österreich wurden in der Zeit vom November 1985 bis Mai 1989 618 HIV-Infizierte, darunter 270 hospitalisierte Patienten (zumeist mit klinisch manifestem AIDS), untersucht. 58% aller Personen hatten Antikörper gegen Toxoplasma gondii; eine klinisch manifeste Toxoplasmosis trat in 20% der 167 mit dem Erreger infizierten hospitalisierten Patienten auf. Unter den 68 Patienten mit Verdacht auf Pneumozystose konnte Pneumocystis carinii in 29% nachgewiesen werden. In 9% von 219 Patienten wurde Cryptosporidium sp. gefunden, in

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zwei Personen wurde Strongyloides stercoralis nachgewiesen. Abgesehen von diesen AIDS-
assozierten Infektionen sind parasitäre Infektionen unter HIV-infizierten Personen in Öster-
reich selten und — mit Ausnahme von Entamoeba histolytica — jedenfalls nicht signifikant
höher als in nicht HIV-infizierten Österreichern.

Beim Vergleich mit in anderen Ländern gewonnenen Daten zeigt sich, daß die Diagnose
einer Toxoplasmose in Österreich offenbar häufiger gestellt wird, ebenso die einer Infektion
mit Cryptosporidium sp.; hingegen wird eine Pneumocystose etwas seltener nachgewiesen.

Introduction

In November 1985 we started a project on the frequency and significance of parasitic
infections among HIV-infected persons with or without a clinical manifestation of AIDS. Most of
the persons were tested repeatedly over an extended period. HIV-positive persons
without clinical symptoms were included in a surveillance programme for parasitic diseases
on a voluntary basis.

The results reported in this paper allow a comparison with figures obtained in other
countries and offer prognostic figures for improvement and focal points of future
surveillance of HIV-infected persons in Austria.

Material and Methods

In the course of this study, altogether 618 HIV-infected persons could be examined until
May 1989. In our study, two groups of persons were distinguished and dealt with: Group 1
comprised all HIV-infected persons regardless of their clinical status; thus this group
included asymptomatic HIV-seropositive, ARC, and AIDS patients. For comparison: Until
January 1989 (latest figure available), altogether 2707 HIV-infected persons had been
recorded in Austria (19).

Group 2 comprised 270 persons of group 1 who had been hospitalized. These 270
persons were composed by 220 patients with clinical manifestation of AIDS according to the CDC
classification (7) (i.e. about 90% of all cases of AIDS so far notified in Austria (243 cases
known) (31)) on the one hand and by 50 patients with severe symptoms of opportunistic or
AIDS-related diseases, but not with AIDS according to the CDC classification on the other.

The average age of all persons examined was 35 years; 80% of them were males. The
individual risks of the persons examined were distributed as follows (18):

<table>
<thead>
<tr>
<th>Category</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>I. v. drug abusers</td>
<td>40.6%</td>
</tr>
<tr>
<td>Homo- or bisexuals</td>
<td>27.0%</td>
</tr>
<tr>
<td>Haemophiliacs</td>
<td>6.6%</td>
</tr>
<tr>
<td>Blood recipients</td>
<td>4.8%</td>
</tr>
<tr>
<td>Heterosexuals</td>
<td>9.6%</td>
</tr>
<tr>
<td>Children of seropositive mothers</td>
<td>0.7%</td>
</tr>
<tr>
<td>Unknown risk</td>
<td>10.7%</td>
</tr>
</tbody>
</table>

The number of specimens examined and the types of examinations are listed in Table 1.

Tests for toxoplasmosis

Serum samples were routinely tested in indirect fluorescent antibody tests (IFAT) for IgG
and IgM antibodies on the one hand and in an enzyme-linked immunosorbent assay (ELISA)
for circulating antigen, according to the test pattern described by Hassl and Aspöck, 1989
(14), on the other. Cerebrospinal fluid (CSF) samples were tested in the same manner,
moreover, the pellets were screened for Toxoplasma trophozoites after having been stained.
Table 1. Number of persons and number of specimens of HIV-positive persons examined and type of examinations

<table>
<thead>
<tr>
<th>Materials examined</th>
<th>Toxoplasma gondii</th>
<th>Intestinal parasites</th>
<th>Pneumocystis carinii</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum sample</td>
<td>564/1521</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CSF samples</td>
<td>57/174</td>
<td></td>
<td></td>
</tr>
<tr>
<td>BAL samples</td>
<td>63/69</td>
<td>45/135</td>
<td></td>
</tr>
<tr>
<td>Induced sputa</td>
<td>46/134</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stool samples</td>
<td>219/438</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>568/1764</td>
<td>219/438</td>
<td>68/269</td>
</tr>
</tbody>
</table>

in a direct immunofluorescent test (DFT) using a rabbit *anti-Toxoplasma* IgG and an FITC-conjugated anti-rabbit antibody (Cappel Lab., Cochranville, PA). Bronchoalveolar lavage (BAL) samples were stained in the DFT in the same way. Toxoplasmosis was diagnosed either if antigen or trophozoites could be detected in the specimens, or if seroconversion occurred, or if an intrathecal IgG production could be demonstrated, or if high IgG titres and/or IgM antibodies were detected (14, 15).

Specimens submitted for examination for *Pneumocystis carinii* (BAL samples or samples of induced sputum) were treated according to the recommendations of the Bundesgesundheitsamt, Berlin, (6) and Giemsa-stained. Moreover, each specimen was stained with a DFT according to the manufacturer's recommendation (Progen Biotechnik GmbH, Heidelberg/FRG).

For demonstration of parasites in stool samples the specimens were examined by the merthiolate-iodine-form-aldehyde method according to (6), and stained according to Heidenhain (26) on one hand and to Heine (6) for the demonstration of Cryptosporidium sp. on the other.

**Results**

*Toxoplasma* infections could be demonstrated serological and/or by detection of trophozoites in 330 subjects, i.e. 58%. The incidence of *Toxoplasma* infections and of overt toxoplasmosis in Austrian HIV-infected persons is listed in Table 2. *Toxoplasma* trophozoites could be demonstrated in the sediments of two samples of CSF of hospitalized patients with clinically important CNS toxoplasmosis (serum titres: first patient: IFAT: neg., IgM-IFAT: neg., no circulating antigens; second patient: IFAT: 1:256, IgM-IFAT: neg., circulating antigens detectable), and in one BAL sample (no concurrent serum titres available) of a patient with pneumonia suspected to be pneumocystosis.

The incidence of *Pneumocystis carinii* can be seen from Table 2.

The results of examinations for intestinal parasites of HIV-infected Austrians can be seen from Table 3.
Table 2. Number of persons examined / number of positive / incidence of pneumocystosis, *Toxoplasma* infections, and toxoplasmosis in HIV-infected Austrians and hospitalized patients

<table>
<thead>
<tr>
<th>Group:</th>
<th>Pneumocystosis</th>
<th><em>Toxoplasma</em> infections</th>
<th>Toxoplasmoma in <em>Toxoplasma</em> infected persons</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV-infected persons</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(group 1)</td>
<td>68/20/29%</td>
<td>568/330/58%</td>
<td>330/40/12%</td>
</tr>
<tr>
<td>Hospitalized patients</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(group 2)</td>
<td>59/20/34%</td>
<td>269/167/62%</td>
<td>167/33/20%</td>
</tr>
</tbody>
</table>

Table 3. Parasites and commensals of the intestine in Austrian HIV-infected persons

<table>
<thead>
<tr>
<th>Parasite species</th>
<th>Positive findings in HIV-infected persons (n = 219)</th>
<th>Parasite species</th>
<th>Positive findings in HIV-infected persons (n = 219)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Absolute</td>
<td>%</td>
<td>Absolute</td>
<td>%</td>
</tr>
<tr>
<td>Cryptosporidium sp.</td>
<td>20</td>
<td>9.1</td>
<td></td>
</tr>
<tr>
<td>Entamoeba histolytica</td>
<td>8</td>
<td>3.6</td>
<td></td>
</tr>
<tr>
<td>Entamoeba coli</td>
<td>13</td>
<td>6.0</td>
<td></td>
</tr>
<tr>
<td>Giardia lambila</td>
<td>6</td>
<td>2.7</td>
<td></td>
</tr>
<tr>
<td>Sarcocystis sp.</td>
<td>1</td>
<td>0.4</td>
<td></td>
</tr>
<tr>
<td>Blastocystis hominis</td>
<td>18</td>
<td>8.2</td>
<td></td>
</tr>
<tr>
<td>Chilomastix mesniili</td>
<td>2</td>
<td>0.9</td>
<td></td>
</tr>
<tr>
<td>Jodamoeba buetschlii</td>
<td>4</td>
<td>1.8</td>
<td></td>
</tr>
<tr>
<td>Dientamoeba fragilis</td>
<td>2</td>
<td>0.9</td>
<td></td>
</tr>
<tr>
<td>Enteromonas sp.</td>
<td>2</td>
<td>0.9</td>
<td></td>
</tr>
<tr>
<td>Strongyloides stercoralis</td>
<td>2</td>
<td>0.9</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>54</td>
<td>24.5</td>
<td></td>
</tr>
</tbody>
</table>

Discussion

Persons with an HIV-infection are especially endangered by several opportunistic parasites, the most important of them being undoubtedly *Toxoplasma gondii*, *Pneumocystis carinii*, and *Cryptosporidium sp.* (20, 23). The question arises whether these parasites distributed world-wide are as frequent in Austrian HIV-infected persons as in HIV-positive persons living in other parts of the world and whether other autochthonous parasites may become more frequent and important in AIDS patients than in immunocompetent Austrians.

Infections with *Toxoplasma gondii* are very frequent in the Austrian population; a large amount of data on infection rates of pregnant women is available demonstrating that about 45-50% of the Austrian women in this age group are infected with *Toxoplasma* (3), (unpublished data). The infection rate among Austrian HIV-positive per-
sons is, however, higher: about 58% were found to be infected with *Toxoplasma gondii*, in hospitalized patients, even 62% were found to be infected with this parasite. We have no conclusive and convincing explanation for this relatively high infection rate.

Diagnosis of toxoplasmosis is difficult in patients with an HIV infection due to their poor antibody response. The demonstration of antibodies has to be considered as a confirmation of infection (22, 25), but single antibody titration or even definition of antibody specificity by immunobLOTS does not allow a conclusive assessment of the toxoplasmosis status (16). We have, however, established a useful serodiagnostic system for the uncovering of toxoplasmosis in patients with HIV infections (14, 15). We could detect toxoplasmosis in 40 (= 12%) of the 330 HIV positives infected with the parasite (Table 2), and in 33 (= 12%) of the hospitalized patients. This is far less than expected by Pohle and Eichenlaub (25) for the Federal Republic of Germany, but corresponds well with data for the USA (3-12%) (9, 17, 29), and is even significantly higher than data reported by the Centers for Disease Control (27), which are, however, almost exclusively based on diagnosis by demonstration of trophozoites in brain biopsies. Although the detection of free trophozoites of *Toxoplasma gondii* in CSF is an important indication of CNS-toxoplasmosis (11), the method is of limited value in routine diagnosis as trials to detect the parasite itself are often unsuccessful even in patients with otherwise confirmed toxoplasmosis. Among the 40 patients with clinically important toxoplasmosis, we succeeded in detecting trophozoites in the CSF in only two patients, although from almost all of them several CSF samples had been taken.

*Pneumocystis carinii* has been less frequently found in HIV-infected persons in Austria than in other countries (5, 10, 27, 28). Among the 68 persons investigated, most of them with clinical manifestations of AIDS, this parasite was found in 29% only. This may at least in part be due to the excellent medical surveillance of HIV-infected persons in Austria (31) combined with a widely applied chemoprophylaxis, but possibly also to a lower number of examinations asked for by the clinicians. In part, induced sputum and BAL were examined in parallel. In such cases, we found a satisfying agreement of results obtained with both kinds of specimens. Thus, we believe that the examination of induced sputum is useful, although not definitely conclusive, in the diagnosis of pneumocystosis, especially if immunoStaining methods are used (4, 21).

Whereas the prevalence of cryptosporidiosis in Europe is between 1 and 2% among the general population (13), it is apparently higher in persons with an HIV infection, particularly in AIDS patients (world-wide about 3.6% (24), for our data see Table 3). Our data on the incidence of cryptosporidiosis among AIDS patients correspond well with data reported in other studies, particularly with infection rates found among AIDS patients in London (9.5%; (8) and 11%; (12) but are higher than found in Switzerland (5%; (5)). It is noteworthy that we could not find a clear correlation between diarrhoea and infection with this parasite. There seem to be many asymptomatic carriers of *Cryptosporidium sp.* among the HIV-infected persons (30), whereas on the other hand, diarrhoea is a normal and frequent symptom in AIDS.

Besides *Toxoplasma gondii, Pneumocystis carinii*, and *Cryptosporidium sp.* parasites are rarely found in Austrians, in HIV-infected persons as well as in immunocompetent persons. This can be demonstrated by comparison of the number of persons infected with intestinal helminths (about 3% in the normal Austrian population (2) vs. 1% in HIV-infected persons) and with intestinal protozoa (about 14% (2).
vs. 24%; in London, 30% were found to be infected with intestinal protozoa (12). We succeeded in detecting *Strongyloides stercoralis* infections in two AIDS patients; this parasite is, however, usually not found in the autochthonous human population in Austria (2). It is noteworthy that no other intestinal helminthoses could be diagnosed. *Entamoeba histolytica* was found in 3.6% of the AIDS patients which is significantly higher than the infection rate found in the normal population (1%) (2), but this parasite may be a commensal organism in homosexual men, usually with a low pathogenicity (1). Other intestinal protozoa were not found in significantly high rates.

Thus, there is no doubt that toxoplasmosis, pneumocystosis, and cryptosporidiosis are by far the most important infections with opportunistic parasites among Austrian HIV-infected persons and particularly among AIDS patients.

Due to the Austrian care system for HIV-infected persons, good follow-up data on the medical status are available. Therefore, longitudinal studies following cohorts of initially asymptomatic HIV-infected persons over several years will be more and more possible with the increasing number of AIDS victims. Then it will also be possible to determine the cumulative incidence of less common parasitic infections not directly related to AIDS.

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References


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