Acute toxoplasmoses in immunocompetent patients hospitalized in an intensive care unit in French Guiana

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Abstract

Atypical *Toxoplasma gondii* strains, unrelated to archetypal clonal lineages (I, II, III), have been reported more frequently over the last decade in areas other than Europe and North America. A newly described form of toxoplasmosis, 'Amazonian toxoplasmosis' (AT), has been reported since 2002 in French Guiana. It is characterized by severe cases and atypical strains linked to a neotropical forest-based cycle. We report on the cases of AT that required intensive care management. We performed a prospective observational study on hospitalized adults in the Intensive Care Unit (ICU) from 2002 to 2008. Clinical and laboratory data, microbiological findings and outcomes were recorded. Data, including the ICU simplified acute physiology score and the pneumonia severity index, were calculated. Epidemiological risk factors for AT were assessed through questionnaires. Eleven non-immunodeficient patients were admitted to the ICU in Cayenne for life-threatening pneumonia associated with disseminated toxoplasmosis. Mechanical ventilation was necessary in seven patients, four of whom required immediate orotracheal intubation. Cardiac and ophthalmological abnormalities were found in five and four patients, respectively. One patient died from multiple organ failure. The genetic characterization of *Toxoplasma* DNA using six microsatellite markers revealed unique and atypical genotypes in eight patients. All patients presented epidemiological risk factors for AT. In French Guiana, significant *T. gondii*-related infectious syndrome associated with the lungs, a high level of LDH activity and the reported risk factors for AT was strongly suggestive of disseminated toxoplasmosis with a possible trend toward life-threatening pneumonia.

Keywords: Amazonian toxoplasmosis, atypical *Toxoplasma gondii*, disseminated infection, French Guiana, life-threatening pneumonia
Original Submission: 8 February 2011; **Revised Submission:** 6 August 2011; **Accepted:** 6 August 2011
Editor: S. Cutler
Article published online: 17 August 2011
Clin Microbiol Infect 2012; **18:** E221–E231
10.1111/j.1469-0691.2011.03648.x

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Introduction

Some reports from South America suggest a higher human pathogenicity [1–4] of newly discovered genotypes of *T. gondii*. Particular epidemiological and bioclinical features have been reported during individual infections or community outbreaks in immunocompetent adults and children in French Guiana and Suriname since 1998 [5–7]. All the strains isolated had atypical genotypes and appeared to be linked to a neotropical forest-based cycle involving wild felids and their prey [8–10]. These case reports helped to define a new form of toxoplasmosis, "Amazonian toxoplasmosis" (AT). Through 11 case reports, we describe the most severe form of AT, a disseminated toxoplasmosis with life-threatening pneumonia, occurring in French Guianese adults from 2002 to 2008. Each patient required intensive care management, as they displayed at least one form of organ failure.

Materials and Methods

Study site

This study was performed in the ten-bed medical–surgical Intensive Care Unit (ICU) of Cayenne Hospital in French Guiana, which is a French overseas department of 220 000 inhabitants neighbouring Brazil and Suriname in the Amazonian region.

Patient selection

A prospective analysis was carried out on all consecutive adult non-immunodeficient patients that were admitted to the Cayenne Hospital ICU and diagnosed with acute toxoplasmosis from 2002 to 2008.

Data

Data were collected from patient medical records: (i) general demographic data; (ii) laboratory analysis, microbiology, radiology variables and additional explorations such as ophthalmological examinations (for nine patients) and/or cardiac ultrasonography (for ten patients) depending on the signs; and (iii) the reasons for admission to the ICU, the hospital and ICU length of stay (LOS), the ICU scoring system at admission, and the life-support treatment and drugs prescribed in the ICU. Epidemiological risk factors for AT (biotope of residence, meat-borne risk factors and contact with forestry elements) were obtained from individual patients through interviews when possible or from the patient's family. All patients and/or relatives gave informed consents.

Toxoplasmosis diagnosis

Acute toxoplasmosis was defined as serological evidence of recent *Toxoplasma* infection corresponding to the presence of *Toxoplasma*-specific IgM with seroconversion or a significant increase in IgG anti-Toxoplasma antibody titres. Patient serum samples were analysed using an enzyme immunoassay (EIA) kit to detect *Toxoplasma*-specific IgG and IgM (Abbott Diagnostics, Abbott Park, IL, USA). Patient specimens (12 blood samples, five pulmonary fluids, one pleural fluid and one muscular biopsy) were sent for DNA analysis to the National Reference Center for Toxoplasmosis (Limoges, France) 3–14 days after collection. A Qiagen DNA Mini Kit (Hilden, Germany) was used to extract DNA from these samples and a PCR-based assay was performed to detect the *T. gondii* B1 gene [11]. *Toxoplasma* DNA for genotyping was extracted directly from bronchoalveolar lavage (BAL) or bronchial aspiration (BA) samples (cases no. 4, 5 and 10) and indirectly from infected mouse ascitic fluid after the inoculation of blood samples (cases no. 1, 2, 6, 8 and 11). DNA was genotyped by comparing length polymorphisms in six microsatellite markers (*TUB2*, *W35*, *TgM-A*, *B18*, *B17* and *M33*), as described elsewhere [12].

Medical and ICU scoring systems

We calculated the ICU Simplified Acute Physiology Score (SAPS II) during the first 24 h of admission [13]. This point scale, ranging from zero to 163, corresponds to a predicted mortality between 0% and 100%. The pneumonia severity index (PSI) is used to classify the severity of a patient's community-acquired pneumonia (CAP) before ICU admission as described by Fine *et al.* [14], and make appropriate site-of-care decisions. Patients can be stratified into five risk categories (risk classes I-V) that are used to predict 30-day survival.

Retroviral investigation

All patients were tested for HIV serological status using two HIV tests by EIA (Abbott, Biorad, Hercules, CA, USA or Biomérieux, Marcy l'Etoile, France). The Murex HTLV-1/2 EIA test (Abbott) was used to screen for human T lymphotropic virus status.

Microbiology differential diagnosis

Additional tests, including serological tests, direct examination, media culture or PCR-based tests were performed on patients' samples (blood, cerebrospinal fluid, BAL, sputum and gastric aspirations) for differential diagnosis: malaria, tuberculosis, Q fever, leptospirosis, infection with *Chlamydia sp*, *Mycoplasma pneumoniae*, *Legionella pneumophila*, *Rickettsia sp, Pneumocystis sp, Histoplasma capsulatum* or other exotic fungi, Epstein-Bar virus, cytomegalovirus, respiratory syncytial virus and arbovirus. Bronchoscopic samples were obtained based on clinical features and CAP management guidelines. Cytology was performed on BAL from seven patients.

Results

From January 2002 to December 2008, 11 patients were admitted to the Cayenne Hospital ICU for life-threatening pneumonia with disseminated toxoplasmosis. Two of the 11 patients had been previously described by Carme *et al.* [6].

Epidemiological description of the study population

The median age was 28 years (range, 18–41) and there was a predominance of men (nine patients) and Maroon people (seven patients) (Table 1 and Fig. 1). Eight patients were permanently living in the forest, but all presented risk factors for AT in the 2 weeks before the onset of symptoms. Dietary risk factors were reported for eight patients, and the animal eaten was clearly identified in four patients. Meat

TABLE 1. Epidemiological characteristics of the patients

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6	Case 7	Case 8	Case 9	Case 10	Case 11	Summary
Epidemiology												
Sex	Male	Male	Male	Male	Male	Male	Male	Female	Female	Male	Male	9 M/2F
Age	22	41	36	39	19	30	38	28	22	24	18	—[b]
Ethnic group	Creole	Maroon	Maroon	Caucasian	Maroon	Caucasian	Caucasian	Maroon	Maroon	Maroon	Maroon	–
Biotope of residence	Littoral	Primary forest	Primary forest	Semi-natural forest	Primary forest	Littoral	Semi-natural forest	Semi-natural forest	Semi-natural f orest	Littoral	Primary forest	–
Risk factors for AT												
Environmental conditions	No	Yes	Yes	Yes	Yes	No	Yes	Yes	Yes	No	Yes	8 Yes
Permanent (P)/Occasional (O)		P	P	O	P	–	O	O	O	–	P	4 P
Presence of wild felids		Yes	Yes	No	No	–	Yes	Yes	No	–	Yes	5 Yes
Contact with surface/river water		Yes	Yes	Yes	Yes	–	No	Yes	Yes	–	Yes	7 Yes
Use of forestry plants		Yes	No	No	Yes	–	No	Yes	Yes	–	Yes	5 Yes
Practice of forestry activities		Yes	Yes	Yes	Yes	–	Yes	Yes	Yes	–	Yes	8 Yes
Alimentary habits												
Game meat consumption	Yes	No	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	10 Yes
Type of meat	Various	–	Various	Various	Various	Various	Various	Various	Various	Various	Various	10 various
Frequence	Two times per week	–	Two times per week	Two times per month	One time per 2 months	One time per 6 months	One time per 6 months	Three times per week	Two times per week	One time per 6 months	Three times per week	–
Cooked	Cooked	–	Various	Various	Various	Various	Cooked	Various	Cooked	Cooked	Various	–
Surface/river water	No	Yes	Yes	No	Yes	No	No	Yes	Yes	No	Yes	6 Yes
Forest plants/vegetables	No	Yes	Yes	No	Yes	No	No	Yes	Yes	No	Yes	6 Yes
Recent exposure to AT[a]												
Environmental	No	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes	Yes	Yes	9Yes
Place	–	Kourou	Stoolman	–	Grand Santi	St Elie	SLM	Sparouine	SLM	Sinamary	Nasson	–
Activities in forest	–	Plantation	Goldpanning	–	Games	Military	Aeromodelling	Plantation	Plantation	Games	Goldpanning	–
Alimentary	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes	10 Yes
Surface/river water	No	Yes	Yes	No	Yes	Yes	No	Yes	Yes	No	Yes	7 Yes
Game meat	Yes	No	Yes	Yes	Yes	Yes	No	No	Yes	Yes	Yes	8 Yes
Type of meat	Various	–	Various	*H. Hy*	*A. paca*	Various	–	–	*T. pecari*	*T. terrestris*	Various	–
Under/Not cooked	Various	–	Various	Carpaccio	Under	Various	–	–	Cooked	UD	UD	–

AT, Amazonian toxoplasmosis; SLM, Saint-Laurent-du-Maroni; *H. hy*, *Hydrochaeris hydrochaeris* nominated 'Cabiai'; *A. paca*, *Agouti paca* nominated 'agouti'; *T. pecari*, *Tayassu pecari* nominated 'Pakira'; *T. terrestris*, *Tapirus terrestris* nominated 'Tapir' or 'Maipouri'; UD, undetermined.

[a]Recent exposure to AT means exposure in the 2 weeks before the onset of the symptoms.

[b]Age (years): mean = 28.8; median = 28; 95% CI, 23.8–33.8; interquartile range (Q1–Q3), 22–37.

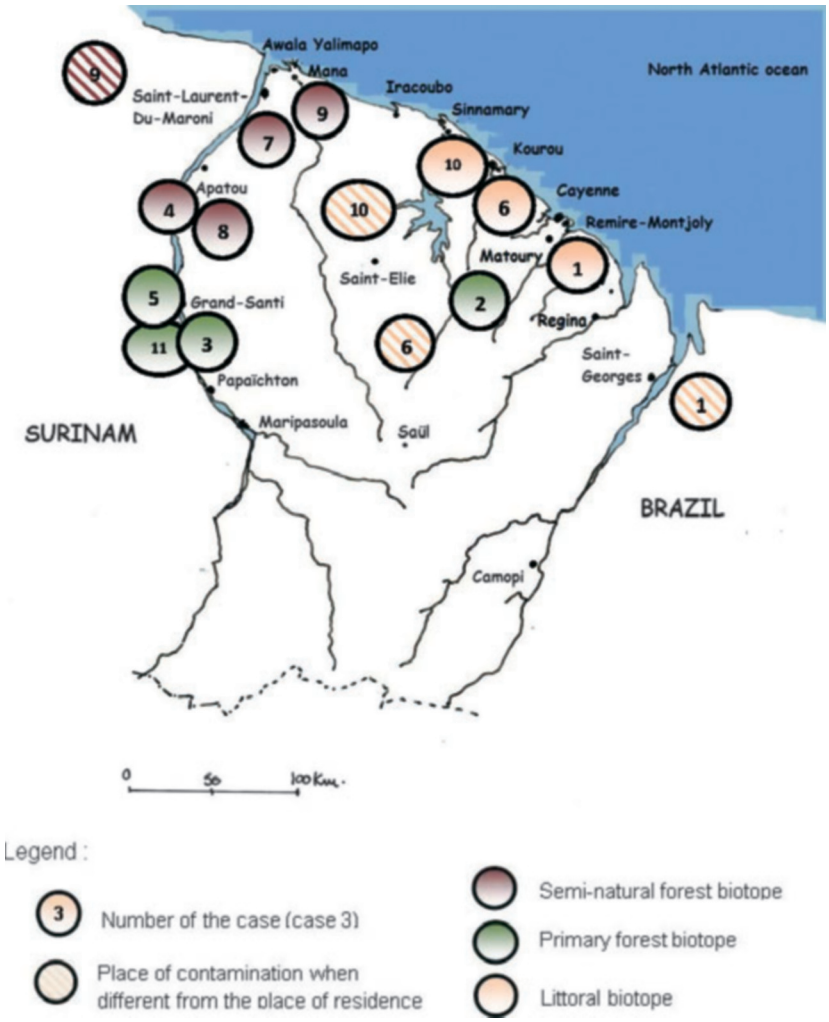

FIG. 1. Map of French Guiana indicating for each patient the place and the biotope of their residence and the place of contamination. The number indicates the case's number (1 = case no. 1), the colour of the circle indicates the type of biotope (green = primary forest, red = semi-natural forest, yellow = littoral biotope) and the hatched colour indicates the place of contamination when it is different from the place of living. The map of French Guiana is available on http://www.carnetderoutes.net/html/guyane/guyane.html. (Please note that it is the responsibility of the author(s) to ensure that all URLs given in this article are correct and useable.)

consumption was regular in cases 1, 3 and 6, making it difficult to reliably identify which type of meat was responsible for the infection. Environmental risk factors for patient 2 were clearly identified, as he was a vegetarian and a healer, and thus was used to handling plants from the forest.

Medical characteristics of patients

None of the other patients presented relevant medical history, except one patient who had homozygous sickle cell disease (case 1) (Tables 2 and 3). The main clinical presentation consisted of a sudden, significant non-specific infectious syndrome with (i) a history of high (median value of 40°C), prolonged (lasting more than 15 days) and continuous fever, (ii) a slow major general decrease in health with progressive weight loss, anorexia and weakness that started with the onset of symptoms, and (iii) lung involvement. Hepatomegaly, splenomegaly or lymph node enlargement was found, and cardiac or ophthalmological abnormalities were described in five and four patients, respectively. One patient had anasarca with hemodynamic instability. The patient's initial symptoms quickly developed into a severe sepsis-like infection, mainly associated with acute respiratory distress, requiring referral to the ICU in Cayenne. Haemodynamic instability with septic shock, renal insufficiency or hepatic insufficiency occurred in three, one and one patients, respectively. Five patients had an ICU SAPS II score that was >30 and a predicted mortality that was >12%; this was especially apparent in the patient that died (predicted mortality, 43.8%).

TABLE 2. Clinical characteristics of the 11 patients

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6	Case 7	Case 8	Case 9	Case 10	Case 11	Summary
Medical history	Sickle-cell disease	None	None	None	None	None	None	None	None	None	None	–
Period of hospitalization	January 2002	March 2002	May 2002	February 2003	January 2004	February 2004	April 2004	December 2004	December 2004	June 2005	July 2006	2002–2004 (9 cases) 2005–2008 (2 cases)
Time to 1st hospitalization after symptom onset (days)	12	9	7	11	6	19	14	21	10	14	30	——[a]
Time to hospitalization in the ICU after 1st hospital visit (h)	504	24	26.7	213	150	48	30	22	18	73	38	——[b]
Time to diagnosis since symptoms (days)	14	11	15	17	8	21	15	24	11	17	32	——[c]
Time to adequate treatment (days)	15	12	16	19	11	20	15	27	11	18	31	——[d]
Number of consultations before hospitalization	1	1	1	1	2	1	2	1	1	1	2	–
Length of stay (LOS) (days)												
In hospital	70	20	19	49	39	15	11	27	14	20	6	——[e]
In ICU (percentage of LOS hospital)	10 (14)	4 (20)	4 (21)	28 (57)	13 (33)	6 (40)	4 (36)	15 (55)	6 (43)	8 (40)	4 (66)	——[f]
Highest temperature (°C)	41.4	38.8	41	41.9	40.8	40.4	40	39.4	40.2	40.2	40.4	——[g]
Painful syndrome (headache, aches, myalgia)	No	No	Marked	No	Yes	Marked	Yes	Yes	Yes	Marked	Marked	8 Yes
Major general impairment	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	11 Yes
Splenomegaly	Yes	No	No	Yes	No	No	No	No	No	No	Yes	3 Yes
Hepatomegaly	No	No	No	No	No	Yes	No	Yes	No	No	Yes	3 Yes
Adenopathy	Yes	Yes	Yes	Yes	No	Yes	No	No	No	No	Yes	6 Yes
Digestive disorders	No	Yes	No	No	Yes	Yes	No	No	No	No	Yes	4 Yes
Respiratory disorders	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	11 Yes
Heart disorders	Pericarditis	No	No	No	No	Heart rhythm disturbance	No	Pericarditis, Hypokinesy, Low Ef, 26%	Pericarditis	No	Pericarditis, Low Ef, PH	5 Yes
(Cardiac ultrasonography)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(No)	(Yes)	(11 Yes)
Other disorders	No	No	Chorioretinitis	No	Meningismus	Conjunctivitis	No	Ascites +++ Turgescence Chorioretinitis	Sore throat	Conjunctivitis Meningismus	No	6 Yes
(Ophtalmological examination)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(Yes)	(No)	(No)	(9 Yes)

Ef, ejection fraction; PH, pulmonary hypertension; SLM, Saint-Laurent du Maroni; ICU, intensive care unit. Digestive disorders: diarrhoea, vomiting or abdominal pain.
[a]Time to 1st hospitalization (days): mean = 13.9; median = 12; 95% CI, 11.2–16.6; interquartile range (Q1–Q3), 9.5–16.5.
[b]Time to hospital ICU (h): mean = 104.1; median = 38; 95% CI, 17.6–190.5; Q1–Q3, 25–111.5.
[c]Time to diagnosis (day): mean = 16.8; median = 15; 95% CI, 13.9–19.6; Q1–Q3, 12.5–19.
[d]Time to treatment (day): mean = 17.7; median = 16; 95% CI, 14.9–20.53; Q1–Q3, 13.5–19.5.
[e]LOS in hospital (days): mean = 26.3; median = 20; 95% CI, 15.1–37.6; Q1–Q3, 14.4–33.
[f]LOS in ICU (%): mean = 38.7; median = 40; 95% CI, 27–49; Q1–Q3, 22.3–48.3.
[g]Highest temperature (°C): mean = 40.4; median = 40.4; 95% CI, 39.9–40.9; Q1–Q3, 40.1–40.9.

TABLE 3. Characteristics of respiratory insufficiency in the 11 patients

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6	Case 7	Case 8	Case 9	Case 10	Case 11	Summary
Reasons for admission to ICU												
Numbers of insufficiency	1	1	1	2	3	1	1	2	1	1	3	____[a]
Type of insufficiency	R	R	R	R,H	R,H, He	R	R	R,H	R	R	R,H, Re	11 R, 4 H, 1 Re
Respiratory insufficiency variables												
Before ICU												
Oxygenation status	10 L of O_2	Room air	15 L of O_2	Room air	10 L of O_2	Room air	12 L of O_2	Room air	Room air	Room air	6 L of O_2	
Respiratory rate	54	48	54	57	30	40	36	53	44	45	45	____[b]
Arterial PH	7.5	7.43	7.42	7.32	7.47	7.43	7.46	7.49	7.46	7.44	7.01	____[c]
Serum bicarbonate (mmol/l)	23	20.1	19	27.3	13.9	22	24.5	21.7	23.3	19.6	19	____[d]
Arterial partial O_2 pressure ($Pa0_2$)	46	51	67.5	47	54	53	70	39.7	40.6	57	95	____[e]
Oxygen saturation (SaO_2)	87	87	93.1	87	91	88.3	95	80.2	79	90.9	90	____[f]
PSI (score/class)	97/IV	121/IV	101/IV	154/V	154/V	95/IV	113/IV	113/IV	77/III	89/III	153/V	____[g]
ICU scoring system at admission												
SAPS II (predicted mortality) (%)	29 (9.7)	21(4.2)	32 (12.8)	33 (14)	36 (18.1)	20 (3.7)	32 (12.8	18 (2.9)	22 (4.7)	13 (1.5)	40 (43.8)	____[h]
$Fi02/PaO_2$	277.5	88	67.2	65	67	260.2	103	158.6	142.4	152.2	93	____[i]
Thoracic imaging during 1st hospital admission	Bilateral interstitial syndrome	Bilateral interstitial syndrome	Bilateral alveolar interstitial syndrome	Normal	Normal	Bilateral interstitial syndrome	Bilateral alveolar interstitial syndrome	Bilateral a lveolar interstitial syndrome	Bilateral interstitial syndrome Left PE	Bilateral interstitial syndrome	Normal	3 normal 5 interstitial syndrome 3 alveoloar interstitial syndrome
Thoracic imaging at ICU admission												
Alveolar abnormalities	Bilateral, R > L	Bilateral, R > L	Bilateral	Bilateral, inf	White lung	Bilateral, R > L	Bilateral	Bilateral,basal	Basal, R > L	Bilateral	Bilateral	11 yes
Interstitial abnormalities	Bilateral	No	Bilateral	Bilateral		Bilateral	Bilateral, L > R	No	Bilateral	Mild, bilateral	Mild	9 Yes
Pleural effusion	R	No	L	No		No	L	Bilateral, L > R	L	No	Mild	6 Yes
Other abnormalities	Cardiomegaly	No	No	No		No	No	Cardiomegaly	No	No	Cardiomegaly	3 Yes

Insufficiency: R, respiratory; H, haemodynamic; He, hepatic; Re, renal. AA, ambient air; $Fi0_2$, fraction of inspiratory 0_2; PE, pleural effusion; ICU, intensive care unit; PSI, pneumonia severity index; SAPS, Simplified Acute Physiology Score; Inf, inferior; ND, not done.

[a]Number of insufficiency: 1 for seven patients; 2 for three patients; 3 for one patient.

[b]Respiratory rate (breaths per min): mean = 46; median = 45; 95% CI, 41–50.94; interquartile range (Q1–Q3), 42–53.5.

[c]Arterial PH: mean = 7.4; median = 7.44; 95% CI, 7.32–7.48; Q1–Q3, 7.42–7.46.

[d]Serum bicarbonate (mmol/L): mean = 21.8; median = 22; 95% CI, 18.8–24.9; Q1–Q3, 19.5–23.9.

[e]PaO_2 (mmHg): mean = 57.6; median = 53; 95% CI, 45.9–69.3; Q1–Q3, 46.5–68.

[f]Oxygen saturation (%): mean = 88.3; median = 88.3; 95% CI, 84.9–92; Q1–Q3, 87–92.

[g]PSI (score): mean = 114.2; median = 113; 95% CI, 94.2–132.1; Q1–Q3, 96–136.5.

[h]SAPS II (score): mean = 26.9; median = 29; 95% CI, 21.6–32.2; Q1–Q3, 20.5–32.5.

[i]$Fi0_2/PaO_2$ (mmHg): mean = 128.7; median = 93; 95% CI, 83.7–173.7; Q1–Q3, 75.7–155.4.

Pulmonary aspects of toxoplasmosis

For all patients, lung involvement started progressively with a dry cough and chest pain during the first 2 weeks of disease (Table 3, Photos 1 and 2). Clinical examinations revealed unilateral or bilateral crackles, unilateral bronchial breathing or areas of dullness. It quickly developed into respiratory insufficiency with acute dyspnoea, and significant abnormality in the blood gas values. Nine patients presented a PSI score that corresponded to risk class IV or above; thus, patients had between a 9.3% and 27% risk of death within 30 days. According to the North American–European Consensus Committee (NAECC) [15], patients 1 and 6 suffered from acute lung injury (ALI), whereas the other patients had acute respiratory distress syndrome (ARDS) on admission to the ICU.

Biological aspects

All patients tested negative for HIV and HTLV1-2 infection and, except for patients 8 and 11, tests for other infectious diseases remained negative. There were no other particular abnormalities on admission, except for mild inflammatory syndrome (median protein C reactive value, 67 mg/L; range, 19.4–323), hyponatraemia (median value, 130 mmol/L; range, 118–139) and elevated LDH (median value, 1432 UI/L; range, 304–1720). Hepatic enzyme activities were also elevated, with a stronger increase in aspartate aminotransferase (median value, 3.9-fold; range, 3–46.11) than in alanine aminotransferase (median value, 2.8-fold; range, 1.18–11.5). Some biological variables worsened after admission to the ICU, such as anaemia (median hemoglobin value, 13.3 g/dL vs. 11.5 g/dL, p 0.006) and the serum prothrombin time (median value, 83.5% vs. 61.5%, p 0.014). BAL analysis in seven patients revealed inflammation with a median leucocyte value of 200/mm^3, a predominance of lymphocytes (median value, 72%), and the presence of macrophages. No parasites were detected on direct examination.

Immunological, molecular diagnosis and genotype analysis of *Toxoplasma*

All patients presented serological evidence of recent *Toxoplasma* infection (Table 4). Although patient 4 displayed seroconversion with the appearance of *Toxoplasma*-specific IgM and IgG in a second sample, the other patients presented significant changes to *Toxoplasma*-specific IgG and IgM values over a short period of time. Humoral response was characterized by high IgM index values in all patients and hypergammaglobulinaemia in five patients (not shown). Molecular diagnosis confirmed *T. gondii* infection in eight patients (Table 4). The muscle biopsy (case 5) was positive even though the patient had been receiving treatment for 22 days. Genotype analysis of *Toxoplasma* DNA samples from eight patients revealed atypical (unusual alleles at loci, shuffled combination of classical and/or unusual alleles) and unique multilocus genotypes.

Treatment and outcome

All patients received antibiotics immediately, which included beta-lactams, fluoroquinolones, macrolides, and more or less aminoglycosides due to the severity of symptoms (Table 4). All patients received oral sulfadiazine and pyrimethamine for a mean period of 42.4 days, except patient 11 who died on day 6 after the start of this treatment. This patient had been treated for longer than a month after the onset of symptoms, unlike the other patients, who received treatment for only 2–3 weeks following the onset of symptoms. All patients except patient 11, who died with a multiple organ dysfunction syndrome, recovered after the initiation of sulfadiazine and pyrimethamine therapy. Side-effects due to sulfadiazine were reported in two patients (cases 4 and 7), requiring replacement with clindamycin. Three patients whilst under treatment developed signs consistent with toxoplasmosis infection: (i) cerebral oedema without focal lesions on day 5 of sulfadiazine/pyrimethamine treatment (patient 4), (ii) intense polymyositis on day 16 of treatment requiring oral corticoids (patient 5), and (iii) unusual hyperprolactinaemia and galactorrhea with no other hormonal or cerebral abnormalities (patient 9). The appearance of long-lasting apyrexia was delayed due to nosocomial infections (patients 1 and 4) and complications such as phlebitis and pulmonary emboly (patient 1). In addition to sulfadiazine/pyrimethamine treatment, patients received ICU treatment based on the type of organ dysfunction. Four patients required a high-concentration oxygen mask, and mechanical ventilation was required by seven patients, four of whom received tracheal intubation. Patients received these non-specific treatments over short periods, lasting no more than 4 days, except for patient 4, who required 16 days of mechanical ventilation (tracheal intubation and non-invasive ventilation). Long-term follow-up was possible in eight patients. Patient 2 presented three typical, active unilateral retinochoroiditis foci 6 years after the acute infection; HIV and HTLV1-2 remained negative. No complications were identified among the other patients.

Discussion

Since 1998, 65 cases of AT have been reported [5, 6, M. Demar, personal communication] in French Guiana. They involve wild *T. gondii* strains that circulate in this Amazonian area between wild felids (definitive hosts), their prey (intermediate hosts) and humans (accidental hosts) [5]. These

TABLE 4. Immunological and molecular diagnosis, treatment and outcome in the 11 patients

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6	Case 7	Case 8	Case 9	Case 10	Case 11	Summary
Toxoplasma serological test results at hospital admission												–
IgG, IU/L	953.9	282.9	0	216	66	10.8	89.8	174.2	158.3	86	154.2	–
IgM EIA Index (negative if <0.6)	9.04	12.55	0.12	10.43	8.3	13.34	7.57	11.98	12.57	7	11.1	–
Molecular diagnosis	Positive	Positive	Negative	Positive	Positive	Positive	Negative	Positive	Negative	Positive	Positive	9 Positive
Specimens tested	Blood	Blood	Blood, exp	BAL	BA, Mus, blood	BAL, blood	Blood (three times)	Blood, PF	Blood	Blood, BAL	Blood	–
Positive specimens	Blood	Blood	–	BAL	BA, mus	Blood	–	Blood, PF	–	Blood, BAL	Blood	–
Isolation in mice	Positive	Positive	Negative	Not done	Not done	Positive	Negative	Positive	Negative	Not done	Positive	5 Positive /8
Classification of *Toxoplasma* strain or DNA	Atypical (strain)	Atypical (strain)		Atypical (DNA)	Atypical (DNA)	Atypical (strain)		Atypical (strain)		Atypical (DNA)	Atypical (strain)	8 Atypical
Time to parasitic diagnosis after symptom onset (days)	13	10	–	20	11	22	–	30	–	17	30	——[a]
Specific treatment												
Duration of treatment (day)	45	60	30	91	30	45	45	45	45	26	5	——[b]
Time to apyrexia/TRT (days)	31	9	–	17	11	5	3	6	6	6	No apyrexia	——[c]
Non specific treatment												
Mechanical ventilation (MV)	No	No	Yes	Yes	Yes	Yes	Yes	No	Yes	No	Yes	7 Yes
Endotracheal tube			Yes	Yes	Yes	No	No		No		Yes	4 Yes
NIV (Noninvasive ventilation)						Yes	Yes		Yes		Yes	
PEEP (maxi, cm H20)			4	15	10	3	5		6		6	——[d]
Duration of MV (day)			1.1	16	5	3	4		2		3	——[e]
Blood transfusions	Yes	Yes	No	Yes	Yes	No	No	Yes	No	No	Yes	6 Yes
Inotropes/vasopressors	No	No	Yes	Yes	Yes	No	No	Yes	No	No	Yes	5 Yes
Central venous/arterial catheters	Yes	No	Yes	Yes	Yes	Yes	No	Yes	No	No	Yes	7 Yes
Extra renal epuration	No	No	No	No	No	No	No	No	No	No	Yes	1 Yes
Community-acquired infection	No	No	No	No	No	No	No	Yes	No	No	Yes	2 Yes
Type of infection	–	–	–	–	–	–	–	Mayaro virus	–	–	Strongyloidiosis	
Nosocomial-acquired infection	Pneumopathy	No	No	Septicaemia	No	Lymphangitis	No	No	No	No	No	3 Yes
Pathogens	P.A, MRSA			*Escherichia coli* (ESLB)		MRSA						
Site of infection	BAL			Bl		Bl, skin						
Evolution under treatment	Un	F	F	Un	Un	F	Un	F	Un	F	Un	5 F
Complications	Phlebitis Pulmonary emboly			Anaemia Renal colic Cerebral oedema	Rhabdomyositis Polymyositis		Hepatitis		Hyperprolactinaemia		MODS	
Death	No	No	No	No	No	No	No	No	No	No	Yes	Death: 1/11
Outcome												
Follow–up	7 years	7 years	7 years	6 years	No	3 years	No	5 years	5 years	3 years		
Complications	No	Chorioretinitis	No	No	No	No	No	No	No	No		

BA, bronchial aspirate; Bl, blood; BAL, broncho alveolar lavage; Exp, expectoration; Mus, muscle biopsy; PF, pleural fluid. Type of insufficiency: R, respiratory; H, haemodynamic; Re, renal. TRT, treatment; P.A, *Pseudomonas aeruginosa*; MRSA, methicillin-resistant *Staphylococcus aureus*; E.C, *Enterobacter cloacae*; NIV, non-invasive ventilation; PEEP, positive end expiratory pressure; ESBL, extended spectrum betalactamase-producing; MODS, multiple organ dysfunction syndrome; F, favourable; Un, unfavourable.

[a]Time to molecular diagnosis (days): mean = 19.12; median = 18; 95% CI, 13.72–24.52; interquartile range (Q1–Q3),12.5–24.

[b]Duration of treatment (days): mean = 42.4; median = 45; 95% CI, 29.7–55.2; Q1–Q3, 30–45.

[c]Time to apyrexia/TRT (days): mean = 10.4; median = 6; 95% CI, 4.7–16.1; Q1–Q3, 6–11.

[d]PEEP: mean = 7; median = 6; 95% CI, 3.9–10; Q1–Q3, 4.5–8.

[e]Duration of MV (days): mean = 4.8; median = 3; 95% CI, 3.13–6.6; Q1–Q3, 2–4.

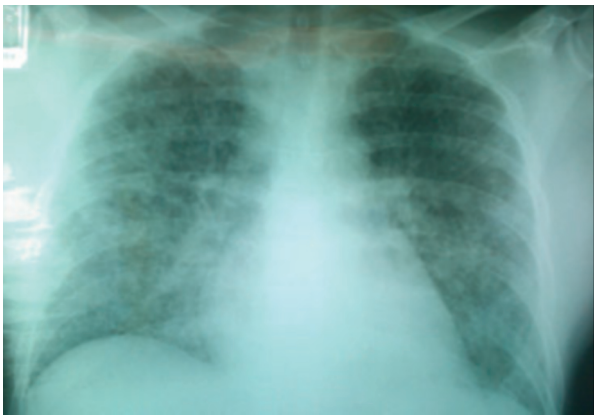

PHOTO 1. Thoracic imaging of Case 4 at ICU admission (chest X-Ray).

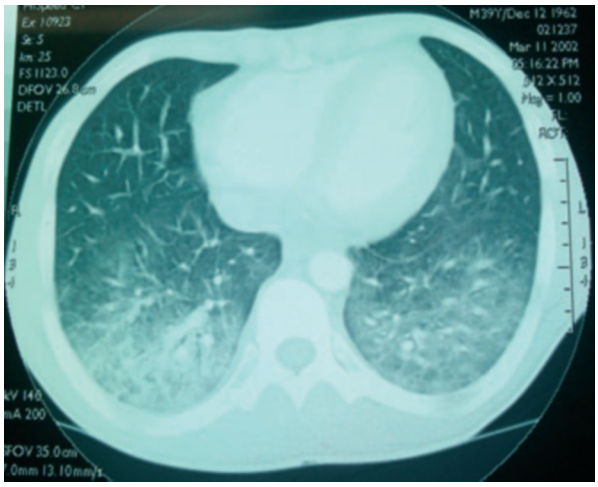

PHOTO 2. Thoracic imaging of Case 2 at ICU admission (chest computed tomography).

strains exhibit an atypical and unique multilocus genotype and present an unusual virulence in mice upon primary isolation [6–10]. At the same time, little was known about the genotypes of strains causing chronic and asymptomatic toxoplasmosis as they have silent expression in immunocompetent adults. The seroprevalence of uncomplicated toxoplasmosis was estimated at 72% among 179 adults hospitalized for reasons other than acute toxoplasmosis from 1999 to 2001 [6]. A recent genetic study, including large numbers of *T. gondii* isolates from domestic and wild animals and from patients suffering with AT, genetically individualized the 'anthropized' and the 'wild' *T. gondii* environmental population with respectively low and high genetic diversity [16]. Genotypes from both environments cannot be classified into the three main lineages described in North America or Europe.

AT presented as a significant infectious syndrome with enlargement of the liver, the spleen and/or the lymph nodes. Even though other organs can be affected such as the heart, central nervous system (CNS) or eyes, the prognosis is linked to lung involvement, which has been consistently reported with variable levels of severity [5,6]. Our series of 11 French Guianese patients with disseminated toxoplasmosis underscores the central role of life-threatening pneumonia. Molecular typing of isolates confirmed their unique and divergent genotypes. Clinical history was consistent with an acute increase in pulmonary signals up to respiratory distress syndrome associated with severe sepsis-like infection. Heart, eye and/or muscle impairment emphasized this severity that has only previously been reported in immunocompromised patients [17–19]. Moreover, patients should be monitored regularly for the rest of their lives, due to the possible reactivation in immunodeficient and non-immunodeficient patients. Despite recent findings in mouse experimental models [20], pulmonary involvement during *Toxoplasma* infection is rare in immunocompetent subjects. It is mainly observed in immunocompromised patients [12,17–19,21–23]. Pomeroy and Filice [22] reviewed 46 cases of Toxoplasma pneumonia, of which 15 were among non-immunodeficient patients. This has been followed by recent sporadic reports [6,24–27].

Neither pulmonary symptoms nor specific findings on chest radiography were found to distinguish *Toxoplasma* pneumonia from pneumonia due to more common pathogens. A high level of LDH can be an adjunctive marker, as previously discussed [28]. Diagnosis was usually established by visualizing tachyzoites in lung tissue or fluids; however, the sensitivity of the method is inconsistent [29,30] and may be dependent on the amount of *T. gondii* in specimens. PCR analysis is increasingly regarded as the most sensitive method for the diagnosis of *Toxoplasma* pneumonia [12]; this was similarly observed in our series. In addition, blood seemed to be most suitable for detecting *T. gondii*, even 30 days after infection onset (cases 8, 11). The *T. gondii* infection had spread to the lungs, inducing necrotizing pneumonitis [31], mostly as part of toxoplasmosis dissemination to multiple organs via parasitaemia, which can lead to septic shock. Wild Amazonian strains appear to be more aggressive with specific pulmonary tropism. This particular clinical feature may also be the result of an overstimulation of immune response, the human immune system being unsuited to such strains. So far, no particular predisposing factors for *T. gondii* lung involvement have been described elsewhere.

Several hypotheses, other than those relating to virulent strains, can be suggested to explain this severity. (i) A delay in introducing disease management (i.e. patient 11, who died,

did not received adequate care early enough). Indeed, patients that survived, despite increased severity (SAPS II, PSI), were managed within 2–3 weeks after the onset of symptoms (patients 3, 4, 5 and 7). (ii) *Toxoplasma gondii* parasitic stage (i.e. ingestion of oocysts). All patients displayed several risk factors for toxoplasmosis exposure. Unfortunately, none of these risk factors allowed the identification of the infective stage, except in patient 2, who was vegetarian. (iii) Host susceptibility; however, except for patient 1 who had sickle cell disease, well-known to promote complications in infection, no other patients had predisposing factors and almost all ethnic groups were represented. The *inoculum* effect may also be mentioned. One of the most severely ill patients (patient 4) consumed pieces of raw game meat (carpaccio), which could contain a significant amount of living parasites. However, this couldn't be proved as no pathological or molecular studies have been carried out on meat. Further *in vivo* investigation is essential to understand the basic mechanism of virulence of these novel isolates.

Conclusion

Although AT remains prevalent in French Guiana, the number of new life-threatening cases has decreased since 2004 (nine patients reported from 2002 to 2004 vs. two patients from 2005 to 2010; M. Demar, unpublished data), probably due to the population and physician awareness. AT is well known in French Guiana and physicians should be aware of such severe cases of acquired toxoplasmosis, which should be suspected in any lung infections with a long lasting high fever in patients who live in or have recently visited the Amazonian region or more generally the neotropical rain forest.

Authorship/Contribution

Magalie Demar participated in the collection, analysis and interpretation of data, the study design and conception and the writing of this report. Didier Hommel participated in the collection of clinical and epidemiological and biological data and the reading of this report. Félix Djossou participated in the collection of clinical and epidemiological data, and the reading of this report. Christian Peneau participated in the performing of biological analysis and the reading of this report. Rachida Boukhari participated in the collection of biological data and the reading of this report. Dominique Louvel participated in the collection of clinical and epidemiological data and the reading of this report. Anne-Marie Bourbigot participated in the collection of clinical and biological data and the reading of this report. Nasser Valery participated in the collection of clinical and biological data and the reading of this report. Daniel Ajzenberg participated in the management and performing of molecular analysis, the study design and conception and the writing of this report. Marie Laure Darde participated in the management and performing of molecular analysis and the design, conception and writing of this report. Bernard Carme participated in improving the study design and the conception, writing and reading of this report.

Transparency Declaration

There was no funding for this study and all the data were generated as part of routine activities from the medical services and from the laboratories. The authors have no dual or conflict of interests related to this article.

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