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Case Report

Molecular diagnosis of toxoplasmosis at the onset of symptomatic primary infection: A straightforward alternative to serological examinations



Maude F. Lévêque^{a,b}, Delphine Chiffre^b, Chloé Galtier^c, Sahar Albaba^b,
Christophe Ravel^{a,b}, Laurence Lachaud^{a,b}, Anaïs Iemmi^c, Pierre Flori^d, Yvon Sterkers^{a,b,e,*}

^a Research Unit "MiVEGEC" (CNRS 5290/IRD 224/UM), University of Montpellier, Montpellier, France

^b Department of Parasitology-Mycology, University Hospital Centre of Montpellier, Montpellier, France

^c Department of Cardiology and Vascular Diseases, University Hospital Centre of Montpellier, Montpellier, France

^d Department of Parasitology-Mycology, University Hospital Centre of Saint-Etienne, Saint-Etienne, France

^e Molecular Biology 'Pole' of the National Reference Centre for Toxoplasmosis, Montpellier, France

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ABSTRACT

Myopericarditis is a rare but well-documented clinical presentation of primary *Toxoplasma gondii* infection in immunocompetent patients. Here, early detection of *Toxoplasma* DNA in the peripheral blood by PCR allowed the diagnosis of acute toxoplasmosis while serological tests were negative. Additional serological evaluations 2 weeks later confirmed the diagnosis and showed that cardiac manifestations occurred before seroconversion. This highlights the importance of a second serological control in the case of a suspected active infection. Overall, we show here that PCR testing for *Toxoplasma* is a sensitive and straightforward alternative to serological examinations.

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Introduction

Toxoplasma gondii is the causative agent of toxoplasmosis and one of the most prevalent zoonotic agents. It infects approximately one-third of the human population globally (Montoya and Liesenfeld, 2004). This parasite spans distinct biological barriers during acute infection and eventually persists as tissue cysts to escape the host immune system. Humans are generally infected through the consumption of undercooked meat or vegetables and water contaminated by cat faeces.

Uncontrolled parasite replication during primary infection in the foetus and reactivation of tissue cysts in the immunocompromised host can lead to life-threatening involvement of one or more organs. Myocarditis is a well-known complication of toxoplasmosis, most often associated with HIV infection (Kirchhoff et al.,

2004). Cardiac clinical manifestations result from active parasite invasion of the pericardium and myocardium, producing local inflammatory responses and potential subsequent cardiomyopathies (Nunes et al., 2017).

This article reports the diagnosis of *Toxoplasma* myopericarditis, occurring before seroconversion for *T. gondii*, by detection of parasite DNA in the peripheral blood of a patient with no laboratory or clinical evidence of immunosuppression. The diagnosis of acute toxoplasmosis in immunocompetent subjects is discussed.

Case report

A 23-year-old man was admitted to the Emergency Room of the Academic Hospital of Montpellier on March 13, 2018. He complained of intense chest pain not relieved by paracetamol, which had started 2 days ago. At the time of admission, a physical examination was normal and the patient appeared feverish, with normal blood pressure and a rhythmic heart beat of 87 bpm. The pain was typical of pericarditis: it was retrosternal, constrictive,

* Corresponding author at: Research Unit "MiVEGEC" (CNRS 5290/IRD 224/UM), University of Montpellier, Montpellier, France.

E-mail address: Yvon.sterkers@umontpellier.fr (Y. Sterkers).

increased with deep inspiration, and was relieved by anteflexion and after treatment with acetylsalicylic acid 1 g. There was no evidence of pericardial friction, which is usually fickle, fleeting, and does not eliminate the diagnosis if absent. An electrocardiogram and echocardiogram showed no particular abnormality. Troponin, a marker of myocardial necrosis, was elevated at 31.3 ng/l (reference value <14 ng/l) as was C-reactive protein (CRP) at 76.1 mg/l (reference value <5 mg/l), suggesting an infectious myopericarditis in the absence of an alternative aetiology.

The patient was transferred to the cardiology intensive care unit the day after admission. Serological and molecular testing was negative for herpes simplex virus 1 and 2, hepatitis B and C virus, HIV, and human herpes virus 6 and 8, and consistent with past exposure to rubella virus, cytomegalovirus, parvovirus B19, and Epstein–Barr virus. The results of immunological analyses were normal (lymphocytes, plasma immunoelectrophoresis, autoantibodies).

Three days after the appearance of the symptoms, serological tests for toxoplasmosis were negative; however, PCR to detect *T. gondii* DNA in the peripheral blood was positive (Table 1). Recent primary infection by the parasite was therefore considered the aetiological factor of the myopericarditis. The patient did not present any symptoms of cervical lymphadenopathy or myalgia, which may occur during the initial stages of *T. gondii* infection (Montoya and Liesenfeld, 2004). However, he complained of intermittent abdominal pain a few days before his admission to the emergency room.

During his hospitalization, the patient's clinical condition improved and levels of troponin and CRP normalized 3 days after the appearance of the cardiac symptoms. Treatment based on acetylsalicylic acid 1 g three times a day for 1 month, lansoprazole 15 mg once a day for 1 month, colchicine 1 mg once a day for 3 months, and bisoprolol 1.25 mg once a day for 3 months was prescribed. The patient did not receive any drugs for toxoplasmosis and was discharged 4 days after admission with an appointment for cardiac magnetic resonance imaging. This revealed probable lateral myocarditis. To confirm the diagnosis of toxoplasmosis, biological follow-up was requested. Two weeks after the pericarditis episode with elevation of troponin levels, PCR no longer detected *T. gondii* DNA in the peripheral blood (Table 1), suggesting that the circulation of parasites had been very transient. However, IgG and IgM were found positive, with a consistent low antigen affinity of IgG at 0.14 index (high >0.6), confirming a recent *Toxoplasma* seroconversion (Table 1).

Discussion

Toxoplasma primary infection is asymptomatic and harmless for the vast majority of healthy individuals, causing mononucleosis-like symptoms with cervical lymphadenopathy in 10% of subjects (Montoya and Liesenfeld, 2004). Primary infections with atypical

strains from South America can be associated with severe clinical involvement in the immunocompetent host (Sobanski et al., 2013). In the case presented here, the patient's history revealed that he had eaten raw horsemeat at a restaurant less than 2 weeks before the appearance of his cardiac symptoms, while no other risk factor was found. The meat served at the restaurant during this period of time had come from Europe and was, therefore, unlikely contaminated by atypical strains. Despite the attempt to genotype the *Toxoplasma* DNA isolated from the patient's blood, it was not possible to confirm this assumption due to the low parasite load.

Toxoplasma pericarditis and myocarditis have already been documented in immunocompetent patients (Bousquet et al., 2016; Chandenier et al., 2000; Montoya et al., 1997; Mroczek-Czernecka et al., 2006; Paspalaki et al., 2001; Pergola et al., 2010; Roubille et al., 2012; Sano et al., 2000), although the number of cases is low. In these previous cases, serological testing was generally used to link the cardiac complications to toxoplasmosis. Indeed, guidelines for the diagnosis of acute toxoplasmosis in immunocompetent patients state the requirement for two serological tests separated by 3 weeks. However, as illustrated here, a number of symptomatic primary infections may be undiagnosed at the first serological examination, due to the delayed synthesis of specific antibodies.

This case highlights the importance of performing blood PCR analysis, which is likely to be the first positive biological test, soon after the appearance of the cardiac symptoms. Indeed, serological conversion for toxoplasmosis was evidenced after the patient's recovery from his cardiac disorders (i.e., relief of chest pain and normalization of troponin levels). Therefore, in the absence of molecular diagnosis and the second serological control, the patient's transient disorders would not have been associated with active toxoplasmosis. The identification of *Toxoplasma* as the aetiological factor of myopericarditis is, however, necessary in order to exclude other probably more severe diagnoses.

In conclusion, when symptomatic *Toxoplasma* primary infection is suspected, physicians should consider blood PCR as a sensitive and straightforward approach for diagnosis.

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Table 1

Detection of *Toxoplasma gondii* DNA from peripheral blood and the course of anti-*Toxoplasma* antibody levels (initial cardiac symptoms: 11 March).

Date	PCR ^a	CMIA IgG ^b	ICT IgG–IgM ^c	CMIA IgM ^d	ISAGA IgM ^e
14 March	Positive (Cp: 35.2) ^f	0.1	Not done	0.05	3+
27 March	Negative ^g	2.50	Positive	5.10	12+

CMIA, chemiluminescent microparticle immunoassay; ICT, immunochromatography technology; ISAGA, immunosorbent agglutination assay; Cp, crossing point.

^a The PCR was first described by Reischl et al. (2003). Briefly, the PCR target is rep529i; the sensitivity was described as detecting down to 2.5×10^{-2} parasite equivalents. In the present authors' hands, using spiked amniotic fluid, the limit of detection in the serial dilution assay was 0.1 *Toxoplasma*/ml (Varlet-Marie et al., 2014).

^b Architect Abbott, Germany; limit of positivity 3 IU/ml.

^c LDBIO Diagnostics, France.

^d Architect Abbott, Germany; limit of positivity 0.6 index.

^e bioMérieux, France; limit of positivity 9+, maximum 12+.

^f Extraction (Hohlfeld et al., 1994); PCR (Reischl et al., 2003).

^g Extraction: EasyMag, bioMérieux, France; PCR (Reischl et al., 2003).

and interpretation of the data, the writing of the manuscript, or in the decision to submit the manuscript for publication.

Ethical approval

We read and complied with the policy of the journal on ethical consent. The study was conducted in accordance with the regulations of the local medical ethics committee of the Hospital University Centre (CHU) of Montpellier, France, in line with the revised Declaration of Helsinki.

Conflict of interest

The authors declare no conflict of interest.

Author contributions

Study design: MFL and YS; data collection: MFL, DC, CG, SA, AI, PF, YS; data analysis: MFL, DC, PF, YS; writing: MFL, DC, CR, LL, PF, YS.

References

- Bousquet A, Bigaillon C, Dumitrescu N, Larréché S, Godreuil C, Mestiri R, et al. Acute myocarditis in an immunocompetent young man: don't forget *Toxoplasma gondii*. *Int J Cardiol* 2016;214:358–9.
- Chandenier J, Jarry G, Nassif D, Douadi Y, Paris L, Thulliez P, et al. Congestive heart failure and myocarditis after seroconversion for toxoplasmosis in two immunocompetent patients. *Eur J Clin Microbiol Infect Dis* 2000;19:375–9.
- Hohlfield P, Daffos F, Costa JM, Thulliez P, Forestier F, Vidaud M. Prenatal diagnosis of congenital toxoplasmosis with a polymerase-chain-reaction test on amniotic fluid. *N Engl J Med* 1994;331:695–9.
- Kirchhoff LV, Weiss LM, Wittner M, Tanowitz HB. Parasitic diseases of the heart. *Front Biosci J Virtual Libr* 2004;9:706–23.
- Montoya JG, Liesenfeld O. Toxoplasmosis. *Lancet Lond Engl* 2004;363:1965–76.
- Montoya JG, Jordan R, Lingamneni S, Berry GJ, Remington JS. Toxoplasmic myocarditis and polymyositis in patients with acute acquired toxoplasmosis diagnosed during life. *Clin Infect Dis* 1997;24:676–83.
- Mroczek-Czernecka D, Rostoff P, Piwowarska W. Acute toxoplasmic perimyocarditis in a 67-year-old HIV-negative woman—a case report. *Przegl Lek* 2006;63:100–3.
- Nunes MCP, Guimarães Júnior MH, Diamantino AC, Gelape CL, Ferrari TCA. Cardiac manifestations of parasitic diseases. *Heart Br Card Soc* 2017;103:651–8.
- Paspalaki PK, Mihailidou EP, Bitsori M, Tsagkaraki D, Mantzouranis E. Polyomyositis and myocarditis associated with acquired toxoplasmosis in an immunocompetent girl. *BMC Musculoskelet Disord* 2001;2:8.
- Pergola G, Cascone A, Russo M. Acute pericarditis and myocarditis by *Toxoplasma gondii* in an immunocompetent young man: a case report. *Infez Med Riv Period Eziologia Epidemiol Diagn Clin E Ter Delle Patol Infett* 2010;18:48–52.
- Reischl U, Bretagne S, Krüger D, Ernault P, Costa J-M. Comparison of two DNA targets for the diagnosis of Toxoplasmosis by real-time PCR using fluorescence resonance energy transfer hybridization probes. *BMC Infect Dis* 2003;3:7.
- Roubille F, Roubille C, Lattuca B, Gervasoni R, Vernhet-Kovacsik H, Leclercq F. Recent toxoplasmosis infection with acute myopericarditis and persistent troponin elevation in an immunocompetent patient. *Cardiol Res* 2012;3:189–91.
- Sano J, Saitoh H, Kobayashi Y, Ikeda M, Kodani E, Takayama M, et al. Toxoplasma pericarditis without immunosuppressant disorder detected by polymerase chain reaction of pericardial fluid: a case report. *J Cardiol* 2000;35:47–54.
- Sobanski V, Ajzenberg D, Delhaes L, Bautin N, Just N. Severe toxoplasmosis in immunocompetent hosts: be aware of atypical strains. *Am J Respir Crit Care Med* 2013;187:1143–5.
- Varlet-Marie E, Sterkers Y, Brenier-Pinchart M-P, Cassaing S, Dalle F, Delhaes L, et al. Characterization and multicentric validation of a common standard for *Toxoplasma gondii* detection using nucleic acid amplification assays. *J Clin Microbiol* 2014;52:3952–9.