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CASE REPORT

# *Toxoplasma gondii* primary infection in renal transplant recipients. Two case reports and literature review

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## Summary

Toxoplasmosis after solid organ transplantation is a complication associated with high morbidity and mortality. Universal prophylaxis with trimethoprim–sulfamethoxazole (TMP-SMX) is effective to prevent post-transplant toxoplasmosis. We report two cases of renal transplant recipients with negative antibodies against *Toxoplasma gondii* pretransplant who developed toxoplasmosis after TMP-SMX discontinuation. We have also performed a review of published cases of primary toxoplasmosis after renal transplantation. Of 20 cases reviewed, 11 were male and the mean age was 37.8 years (SD = 13.8). Donor serology for *T. gondii* was determined in 15 donors, two of them (13%) with negative immunoglobulin (Ig)G and four (27%) with positive IgG and IgM antibodies. Fever was present in 85% of primary toxoplasmosis and hematologic abnormalities were observed in 69% of the cases. Ten patients died (50%). All patients with fatal outcomes had clinical evidence of toxoplasmosis during the early post-transplant period (first 90 days), while no patient with late toxoplasmosis died ($P = 0.003$). Primary toxoplasmosis is associated with high mortality rates and TMP-SMX prophylaxis can delay the onset of symptoms resulting in an improvement of prognosis.

## Background

*Toxoplasma gondii* has been recognized as a potential donor-to-host transmission infection after solid organ transplantation and the diagnosis of toxoplasmosis is usually made after the first month post-transplantation [1–3]. However, this disease occurs mainly in seronegative heart transplant recipients from seropositive donors, as the myocardium is one of the sites were cysts are located [4,5]. Toxoplasmosis transmitted from the donor has also been described after liver and renal transplantation, although is much more infrequent [6].

It has been suggested that administering routinely trimethoprim–sulfamethoxazole (TMP-SMX) prophylaxis for *Pneumocystis jirovecii* may be enough for preventing donor-to-host transmission of *T. gondii* after solid organ transplantation in areas with low prevalence of toxoplasma infection [7]. However, the strategy for preventing toxoplasmosis in donor/recipient serology mismatch for nonheart solid organ transplantation in areas of high seroprevalence of toxoplasma remains undetermined.

Herein we report two cases of primary *T. gondii* infection in two renal transplant recipients. In addition, a review of the published cases of primary toxoplasmosis in renal transplant recipients was performed.

## Case 1

A 39-year-old male patient, with end stage renal failure caused by focal segmental glomerulosclerosis, underwent

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kidney transplantation from cadaveric donor in June 2008. The postoperatory period was uneventful and no acute rejection episodes occurred. *Pneumocystis jirovecii* prophylaxis with (TMP-SMX) was administered as protocol up to 6 months post-transplantation. Immunosuppression consisted of Janus kinase 3 (JAK-3) inhibitor monotherapy.

Two months after transplantation, the patient was readmitted because of anemia, requiring a blood transfusion. Immunosuppressive regimen was switched to tacrolimus plus mofetil mycophenolate (MMF). The bone marrow aspirate was normal and anemia was considered to be an adverse effect of JAK-3 treatment [8].

Eight months after transplantation, he reported flu-like symptoms with headache and nonproductive cough. Shortly afterwards, he was admitted to the hospital with a 38 °C fever, moderate renal failure and cough. Physical examination showed no further abnormalities. The serum creatinin was 2.43 mg/dl, lactate dehydrogenase 599 U/l, C-reactive protein 5 g/l. There was a mild leukopenia (3000 cells/$mm^3$) with normal thrombocyte count and 132 g/l hemoglobin. Although his clinical condition was good during hospitalization, he remained febrile.

A chest X-ray showed slight bilateral interstitial infiltrates. He received empirical treatment with broad-spectrum antibiotics including levofloxacin (500 mg/day) and high-doses of intravenous TMP-SMX (160/800 mg/8 h). Cultures of bronchoalveolar lavage fluid (BAL) for fungi, bacteria and viruses and stains for *P. jirovecii* were negative, therefore TMP-SMX was discontinued after 3 days. DNA amplification of respiratory viral pathogens and cytomegalovirus pp65 antigenemia in BAL were both negative.

He remained febrile for four additional weeks. Toxoplasmosis was suspected because of serologic mismatch between donor and recipient (D+/R−). Serum immunoglobulin (Ig)M antibodies against *T. gondii* 1 month after admission were positive with negative IgG antibodies. Pyrimethamine at a dose of 75 mg/day (loading dose 100 mg) and sulfadiazine at a dose of 1 g four times daily was started and 2 days later the patient was afebrile. Cranial CT scan and chest X-ray were both normal 1 week after initiation of anti-toxoplasma treatment. The levels of immunosuppressors remained within therapeutic ranges with no modifications. This scheme was maintained for six additional weeks and secondary prophylaxis with TMP-SMX (160/800 mg three times a week) was administered for three additional months. *Toxoplasma gondii* antibodies seroconversion (negative IgM plus positive IgG antibodies) was confirmed 1 month after the diagnosis. The patient remained asymptomatic 1 year after transplantation.

## Case 2

A 57-year-old male patient with end-stage renal failure caused by nephroangiosclerosis received a renal transplantation from cadaveric donor in July 2008. The postoperatory period was uneventful and no acute rejection episodes occurred. Immunosuppression consisted in antilymphocyte globulins, tacrolimus and MMF at standard doses. He had a negative serology for CMV and for *T. gondii* (both IgG and IgM) but the donor had positive IgG antibodies for both. Creatinine nadir was 1.5 mg/dl. *Pneumocystis jirovecii* pneumonia prophylaxis with TMP-SMX during 6 months post-transplantation and prophylaxis for CMV with valganciclovir adjusted to renal function (450 mg twice daily) for 3 months post-transplantation were administered as protocol.

Five months after transplantation, he was readmitted because of fever, leukopenia and high transaminases. Qualitative DNA amplification for CMV in serum was positive and intravenous ganciclovir was administered for 2 weeks with good clinical and virologic outcome.

One year after transplantation, the patient reported 2 months of fever with diaphoresis and weight loss. Chest and abdominal computed tomography (CT) scan showed enlarged retroperitoneal lymph nodes and an enlarged spleen. The bone marrow biopsy showed no abnormalities. The fever and the rest of symptoms disappeared without specific treatment in 1 week and he was discharged.

Four months later, he was admitted again with a 38 °C fever and diaphoresis. Physical examination showed no relevant abnormalities and liver function tests were normal (AST 27 IU/l, ALT 26 U/l, alkaline phosphatase 295 U/l, GGT 134 IU/l, total bilirubin 0.6 mg/dl). Blood cultures were negative. An abdominal CT scan showed enlarged spleen without pathologic lymph nodes. A new bone marrow biopsy showed no evidence of parasites. Positron emission tomography was normal.

*Toxoplasma gondii* primary infection was confirmed by serology. The patient had positive IgM antibodies with negative IgG antibodies. Cranial magnetic resonance imaging showed no abnormalities. The patient was treated with pyrimethamine 100 mg per day and sulphadiazine 1 g/6 h during 4 weeks. Two weeks after treatment initiation, he was readmitted with pancitopenia (platelet count $21 \times 10^9$, leukocyte count $1.5 \times 10^{-9}$, hemoglobin 10.3 mg/dl) that was attributed to the treatment. For this reason, treatment was switched to atovaquone 750 mg/8 h with resolution of the hematologic abnormalities. After 15 days of atovaquone, the dose was reduced to 750 mg/12 h and it was maintained for one additional month. Two months after the diagnosis, IgG antibodies against *T. gondii* became positive. After completing the treatment, the patient remained asymptomatic.

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## Methods

We have performed a systemic review via MEDLINE of all published cases of toxoplasmosis after renal transplantation. In the literature, a total of 42 kidney transplant recipients have been described to develop toxoplasmosis post-transplantation. Of these, 20 were either considered reactivations of toxoplasmosis or the pretransplant serology for toxoplasma of the recipient was unknown, as a result these patients were excluded from this review. Two recipients who were considered pediatric (age under 16 years) were also excluded. Finally, 20 kidney recipients with primary toxoplasmosis post-transplantation (18 in the literature and the present two cases) were analyzed.

## Results

Table 1 shows the main features of the 20 reported cases of primary toxoplasmosis after renal transplantation. Of the 20 cases reviewed, 11 were male and the mean age was 37.8 years (SD = 13.8). Donor serology for *T. gondii* was available from 15 donors, two of them (13%) with negative IgG and four (27%) with positive IgG and IgM antibodies. In the literature, information regarding active prophylaxis against toxoplasma was available for seven recipients and none of them developed toxoplasmosis under active prophylaxis. The median days of the onset of symptoms after transplantation was 25 (interquartile range 15.5–180 days), and six cases (30%) were diagnosed after 3 months post-transplantation. Fever was present in 85% of cases of primary toxoplasmosis and clinical manifestations were varied. Hematologic abnormalities were present in 69% of patients, predominating leukopenia and thrombocytopenia. Ten patients died (50%). All patients with fatal outcomes initiated the clinical manifestation during the early post-transplant period (first 90 days), while no patient with late toxoplasmosis died ($P = 0.003$).

## Discussion

Although primary infection with *T. gondii*, mainly transmitted from the donor, may be infrequent in renal transplant recipients, the mortality of this complication is high. In the published cases, mortality reaches 50% (10 deaths out of 20 cases). Thus, efforts to identify patients at risk or to make an early diagnosis are required.

There were various clinical presentations of primary toxoplasmosis in renal transplant recipients. Fever was the most frequent clinical sign (85%), followed by pulmonary affectation. In the radiographic images of *T. gondii* pneumonitis interstitial infiltrates were usually present. Neurologic symptoms such as somnolence, confusion and altered consciousness, seizures, headache, drowsiness and lethargy were commonly unspecific.

The sources of infection in this series did not only include the transmission through the graft. On the one hand, two donors had negative serology for toxoplasma, and on the other, thirty percent of patients had late toxoplasmosis suggesting an environmental source for the acquisition of infection. There are different routes for toxoplasmosis acquisition, being the most important one the consumption of water or material contaminated with cat feces and the ingestion of undercooked meat. Seronegative transplant recipients for toxoplasma must avoid these sources of infection and physicians should warn patients about these hygienic measures during all the post-transplant period. It is also important to point out that 4/15 (27%) donors with available serology for toxoplasma had positive IgM antibodies against *T. gondii*. Seronegative recipients for toxoplasma should not receive a graft from a donor with positive IgM antibodies for toxoplasma as the risk to develop post-transplant toxoplasmosis may be high.

Prevention of primary toxoplasmosis after kidney transplantation is controversial. One report suggests that conventional doses of TMP-SMX for the prevention of *P. jirovecii* pneumonia should be enough for the prevention of toxoplasmosis in nonheart transplant recipients. However, patients can develop the infection after prophylaxis discontinuation. One possibility is to give TMP-SMX during the first 6 months post-transplantation in recipients with negative antibodies against toxoplasma to retard the onset of symptoms. Physicians should, therefore, be aware of the possibility of toxoplasmosis in the case of compatible clinical symptoms after discontinuation. In fact, our two patients developed clinical symptoms compatible with toxoplasmosis within 6 months after the discontinuation of prophylaxis. In these cases, TMP-SMX was efficacious to retard the onset of the infection but not to prevent it. Moreover, all deaths reported in the literature occurred in patients with early toxoplasmosis, reinforcing the importance of giving TMP-SMX post-transplantation in seronegative recipients with close clinical and serologic monitoring until patients reach the minimum net state of immunosuppression.

Diagnosis of primary toxoplasmosis is difficult and it may be underestimated. In 6 of 18 published cases, the final diagnosis was obtained at necropsy (30%), and none of these six patients received any anti-toxoplasma drug. In other review in noncardiac transplant recipients including primary and reactivated toxoplasmosis, autopsy and serology were the two most frequent ways to diagnose toxoplasmosis [25] because at that time molecular diagnosis was not available. Toxoplasmosis can be diagnosed by serology and direct methods. Primary

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**Table 1.** Clinical characteristics of primary toxoplasmosis after kidney transplantation reported in the literature (1966–2009).

| Case (reference) | Year | Age/gender | Donor serology for *Toxoplasma gondii* | Immunosuppressive regimen | Active prophylaxis with any drug active against *Toxoplasma gondii* | Days post-transplant of initiation of symptoms | Fever | Visceral involvement | Clinical manifestations | Diagnosis | Treatment | Outcome |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 (10) | 1966 | 20/M | NA | AZA + PDN | Unknown | 0 | Yes | CNS, lungs, myocardium, skeletal muscle, liver, ureter, testis, parathyroid and own kidneys | Leukopenia, thrombocytopenia, seizures, headaches, abnormal movements, pneumonia | Autopsy | None | Death |
| 2 (11) | 1970 | 44/F | NA | AZA + PDN + irradiation | No | 31 days | No | CNS, myocardium | Tachycardia, ECG changes of myocardial ischemia, bloody stools, coma, abnormal movements, seizures | Autopsy | None | Death |
| 3 (12) | 1977 | 36/M | NA | ATG + AZA + PDN | Unknown | 13 months | Yes | – | Leukopenia, rash, lethargy, anemia | Serology | None | Alive |
| 4 (13) | 1983 | 20/F | IgG+ | AZA + PDN | Unknown | 15 days | Yes | – | Cough, leukopenia | Serology | Sulfisoxalone | Death |
| 5 (13) | 1983 | 30/F | IgG+ | AZA + PDN | Unknown | 28 days | Yes | – | Pneumonia, leukopenia, thrombocytopenia, coma, seizures | Serology | None | Death |
| 6 (14) | 1986 | 30/M | IgG– | ATG + AZA + PDN | Unknown | 16 days | Yes | – | Lymphocytosis, headaches, hepatic and muscular cytolysis | Serology | Pyrimethamine, sulfadoxine | Alive |
| 7 (15) | 1987 | 16/F | IgG+ IgM+ | CyA + PDN | Unknown | 10 days | Yes | – | Seizures, malaise, arthralgias, myalgia, pericarditis | Serology | Iv TMP/SMZ following by oralsulfadfiazine + pyrimethamine | Alive |
| 8 (16) | 1990 | 42/M | IgG+ | OKT3 + PDN + AZA | No | 14 days | Yes | – | Abdominal pain, pneumonia, leukopenia, thrombocytopenia, hepatolysis | Bone marrow apirate, bronchoalveolar lavage | None | Death |
| 9 (17) | 1995 | 37/F | IgG+ IgM+ | ATG + AZA + CyA + PDN → OKT3 | No | 16 days | Yes | – | Renal failure, hemolytic anemia, thrombocytopenia, leukopenia, hepatic cytolysis, hypoxemia | Serology, bone marrow aspirate | None | Death |
| 10 (18) | 1997 | 30/F* | NA | PDN + ATG + CyA | Unknown | 27 days | Yes | Lungs, liver, myocardium, brain and pancreas | Leukopenia, thrombocytopenia, hepatic cytolysis, pulmonary infiltrates. | Autopsy | None | Death (39) |
| 11 (18) | 1997 | 40/M* | IgG+ IgM+ | ATG + CyA | Unknown | 23 days | Yes | – | Thrombocytopenia, leukopenia | Serology, Toxoplasma DNA detection by PCR in serum (day 31), bone marrow apirate | Pyrimethamine + sulfadiazine (empirical on day 32) | Alive |
| 12 (19) | 2002 | 22/M | IgG+ | CyA + AZA + PDN | Unknown | 20 days | Yes | Retinocoroiditis Bilateral interstitial pneumonitis | Pancytopenia | Serology | TMP/SMZ → clindamycin + pyrimethamine | Alive |
| 13 (20) | 2004 Kidney–pancreas | 51/F | NA | ATG + CyA + AZA + PDN | Unknown | 21 months | No | | Vague neurologic symptoms, blurring of vision, headache | Serology | Pyrimethamine + folinic acid + sulfadiazine → clindamycin | Alive |

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**Table 1.** continued

| Case (reference) | Year | Age/gender | Donor serology for *Toxoplasma gondii* | Immunosuppressive regimen | Active prophylaxis with any drug active against *Toxoplasma gondii* | Days post-transplant of initiation of symptoms | Fever | Visceral involvement | Clinical manifestations | Diagnosis | Treatment | Outcome |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 14 (21) | 2005 | 58/F | IgG+ | PDN + FK + MMF | No (TMP/SMZ prophylaxis during the first 3 months post-transplantation) | 4 months | Yes | Brain abscesses (magnetic resonance imaging) Chorioretinitis | Syncope, headache, generalized weakness slurred speech | Serology | Pyrimethamine + sulfadiazine | Alive |
| 15 (22) | 2006 | 38/M | IgG+ IgM+ | ATG + PDN + AZA | Unknown | 10 days | Yes | Lungs, liver, heart, brain | Hemophagocytic syndrome, skin rash, arthralgias, liver dysfunction, shock | Bone marrow biopsy. autopsy | None | Death (21) |
| 16 (23) | 2008 | 60/M* | IgG+ | ATG + MMF + PDN + CyA | Unknown | 23 days | No | Heart, liver, lungs | Bilateral pneumonia, respiratory failure, shock | Autopsy | None | Death (30) |
| 17 (23) | 2008 | 59/F* | IgG+ | Basiliximab + CyA + MMF + PDN | Unknown | 28 days | Yes | Heart, liver, lungs, brain | Bilateral pneumonia, thrombocytopenia, respiratory failure, shock | Autopsy | None | Death (32) |
| 18 (24) | 2008 | 26/M | IgG+ | AZA + CyA + PDN + ATG → FK + MMF | No | 11 years | Yes | – | Skin eruptions, hemophagocytic syndrome, left-ventricular failure | Serology + bone marrow biopsy | Clindamycin + pyrimethamin + sulfamethoxazole/ trimethoprim | Alive |
| 19 Case 1 | 2009 | 40/M | IgG− | JAK-3 → FK + MMF | No (TMP/SMZ prophylaxis during the first 6 months post-transplantation) | 8 months | Yes | – | Interstitial pneumonia, leukopenia | Serology | Pyrimethamine + sulfadiazine | Alive |
| 20 Case 2 | 2009 | 57/M | IgG+ | ATG + MMF + FK | No (TMP/SMZ prophylaxis during the first 6 months post-transplantation) | 16 months | Yes | – | Fever of unknown origin, esplenomegaly | Serology | Pyrimethamine + sulfadiazine Atovaquone | Alive |

M, male; F, female; NA, not available; AZA, azathioprine; PDN, prednisone; ATG, anti-thymocyte globulins; CyA, cyclosporin A; FK, tacrolimus; MMF, mofetil mycophenolate; TMP/SMZ, trimethoprim/sulfamethoxazole; CNS, central nervous system.



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toxoplasmosis can be diagnosed by the presence of positive IgM antibodies with compatible clinical symptoms in a transplant patient with previous negative IgG antibodies against *T. gondii*. However, serology is not useful in patients with reactivated toxoplasmosis. Direct observation of tachyzoites in biopsy samples provides a definitive diagnosis but the sensitivity is low. Polymerase chain reaction (PCR) amplification of the 35-fold repetitive B1 gene for detection of *T. gondii* DNA in body fluids and tissues has successfully been used to diagnose cerebral and disseminated toxoplasmosis in AIDS patients [26]. PCR amplification in blood leads to sensitivities and specificities for diagnosis cerebral toxoplasmosis that can reach 80% and 98%, respectively [9]. Other studies have shown the utility of PCR amplification in brain tissue, cerebrospinal fluid, vitreous and aqueous fluids and BAL in patients with AIDS [27]. In renal transplant patients with a high clinical suspicion of toxoplasmosis in which serology is not useful to make a diagnosis, PCR amplification of *T. gondii* DNA in blood as well as in other affected tissues may be an optimal diagnostic strategy.

In conclusion, toxoplasmosis in seronegative renal transplant recipients is a complication that may cause important morbidity and mortality. Physicians must be aware of this possibility and should apply preventive measures and make an early diagnosis in the case of compatible symptoms.

## Authorship

CC, MM and AM: designed the research/study. MM, LL, GM and IH: performed the research/study. NE, FC, FO, JMM and JMC: contributed important reagents. MM, NE and VT: collected the data. CC, AM, JMM and JMC: analyzed the data. CC, MM, JMM, FC and AM: wrote the paper.

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