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Treatment of classic Whipple's disease: from *in vitro* results to clinical outcome

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Objectives: Patients with classic Whipple's disease have a lifetime defect in immunity to *Tropheryma whipplei* and frequently develop treatment failures, relapses or reinfections. Empirical treatments were tested before culture was possible, but the only *in vitro* bactericidal treatment consists of a combination of doxycycline and hydroxychloroquine.

Methods: Our laboratory has been a reference centre since the first culturing of *Tropheryma whipplei*, and we have tested 27 000 samples by PCR and diagnosed 250 cases of classic Whipple's disease. We report here the clinical course of patients who were followed by one of our group.

Results: Of 29 patients, 22 (76%) were previously treated with immunosuppressive drugs, 26 (90%) suffered from arthralgias and 22 (76%) exhibited weight loss. Intravenous initial treatment was paradoxically associated with an increased risk of failure ($P=0.0282$). Treatment with doxycycline and hydroxychloroquine (±sulfadiazine or trimethoprim/sulfamethoxazole) was associated with a better outcome (0/13 failures), whereas all 14 patients who were first treated with trimethoprim/sulfamethoxazole and referred to us ($P<0.0001$) experienced failure. Among the patients treated with doxycycline and hydroxychloroquine after previous antibiotic treatments, two presented with a reinfection caused by different *T. whipplei* strains. Finally, serum therapeutic drug monitoring allowed us to detect a lack of compliance in the only patient with failure among the 22 patients treated with lifetime doxycycline.

Conclusions: *In vitro* results were confirmed by clinical outcomes and trimethoprim/sulfamethoxazole was associated with failures. The recommended management is a combination of doxycycline and hydroxychloroquine for 1 year, followed by doxycycline for the patient's lifetime along with stringent therapeutic drug monitoring.

Keywords: *Tropheryma whipplei*, doxycycline, lifetime prophylaxis, antimicrobial therapy

Introduction

Classic Whipple's disease is a chronic disease caused by *Tropheryma whipplei*.[1] This disease primarily causes arthralgia and diarrhoea, but most organs can be involved,[2] and the disease is diagnosed by the histological involvement seen in small-bowel biopsies.[3] Empirical treatments were successively proposed that included treatment with chloramphenicol, tetracycline, penicillin G, streptomycin and trimethoprim/sulfamethoxazole.[1] Until 1985, tetracyclines constituted the maintenance treatment of choice, but a high rate of relapses, particularly with CNS manifestations, was described.[4] Thus, some researchers have considered an induction treatment with intravenous penicillin G and streptomycin followed by an oral trimethoprim/sulfamethoxazole regimen for 1 year to be a reasonable alternative.[5]

The successful culturing of *T. whipplei* in 2000 allowed researchers to test the *in vitro* antibiotic susceptibility of the pathogen,[6,7] and the proposed treatments were found to be likely to be inadequate. Ceftriaxone is not active in cell culture, imipenem is efficient against only one out of the three tested strains and trimethoprim is not active because *T. whipplei* lacks the coding sequence for dihydrofolate reductase, which is the target of trimethoprim.[8] Moreover, acquired resistance due to *folP* mutations, the target gene of sulfamethoxazole, has been described,[9,10] thus making co-trimoxazole completely ineffective. The combination of doxycycline and hydroxychloroquine was the only bactericidal treatment against cultured *T. whipplei* in cells owing to the alkalization of the vacuoles, as reported for *Coxiella burnetii*;[6,11] this combination therapy thus constituted the first rational and

non-empirical treatment. In parallel, a correlation between the *in vitro* data and clinical outcome has been reported in *C. burnetii* endocarditis.[12,13] A 15 day regimen of intravenous meropenem or ceftriaxone followed by an oral regimen of trimethoprim/sulfamethoxazole for 1 year has been proposed,[14] and more recently a 3 month regimen of co-trimoxazole has been reported to be highly efficacious.[15] In contrast, 35 cases of clinically acquired resistance and relapses while undergoing treatment with trimethoprim/sulfamethoxazole have been described in different studies,[9,10,16–26] including one relapse in a German patient.[15,27] Thus, we believe that it is not ethical to treat patients with this protocol in France because of the high rate of failures.[9,10,19]

The existence of familial cases,[1] the fact that most patients are Caucasian (although Africans are more exposed to the bacterium) and the significantly higher frequency of HLA-DRB1*13 and DQB1*06 alleles in patients[28] suggest a genetic predisposition towards infection with, consequently, a lifetime vulnerability to the bacteria. These findings have been confirmed by the recent report of one patient of ours who suffered seven successive relapses, including reinfection with a different strain.[29] As proposed in recurrent *Granulibacter bethesdensis* infections in chronic granulomatous diseases, we propose a lifelong prophylactic treatment with doxycycline to avoid relapses after a 1 year doxycycline/hydroxychloroquine regimen.[29,30] Here, we report our experience based on data on the follow-up of 29 patients whom one of us treated (D. R.) for classic Whipple's disease after they were referred to us for primary treatment or after failure or relapse.

Patients and methods

Patient inclusion

Since the first culturing of *T. whipplei* in 2000, we have diagnosed 250 *T. whipplei* infections at our centre (Unité des Rickettsies, Marseille, France), including 150 cases of classic Whipple's disease. Among them, some patients with classic Whipple's disease who were referred to one of us (D. R.) for management were assessed in this study. Some patients had been followed since the beginning of their treatment at our centre and some had been followed for a few months after the diagnosis, while other patients were referred to us after an initial failure of treatment or relapse;[2,19] this explains the differences regarding the initial biological examinations performed at the time of diagnosis. Each patient was followed clinically and biologically (see below). Among these 29 patients, 14 who were initially or secondarily treated with trimethoprim/sulfamethoxazole have had their management in previous studies.[10,19,29] All patients gave informed consent for this report. All patients were contacted by phone for this study to evaluate their current clinical status.

Positive diagnosis

Positive diagnosis was based on histological involvement, i.e. periodic acid–Schiff (PAS) staining and/or immunohistochemistry using specific antibodies against *T. whipplei*, as previously described,[3] in patients with clinical manifestations.[2]

Treatment

Regarding the induction treatment prescribed in other centres, the different antibiotic regimens prescribed were: (i) ceftriaxone (2 g once daily); (ii) ceftriaxone (2 g once daily) and gentamicin (3 mg/kg once daily); (iii) amoxicillin (4 g three times per day) and gentamicin (3 mg/kg once daily); and (iv) piperacillin/tazobactam (4 g three times per day). The oral antibiotic regimen followed by the patients was doxycycline alone for one patient, trimethoprim/sulfamethoxazole and rifampicin for one patient, trimethoprim/sulfamethoxazole (160 mg/800 mg/day), or doxycycline (200 mg/day) and hydroxychloroquine (600 mg/day) for 22 patients. In cases with neurological involvement (clinical manifestations and/or positive *T. whipplei* in CSF), trimethoprim/sulfamethoxazole or sulfadiazine (4 g/day) were added, except in the case of previous failure of these antibiotics. Patients suffering from an immune reconstitution inflammatory syndrome (IRIS) were treated with corticosteroid or thalidomide (200 mg/day) in cases of corticosteroid failure.[29] Regarding follow-up, the serum level of antibiotics was determined twice yearly. To prevent ocular complications caused by hydroxychloroquine, ophthalmologist consultation, including colour vision examination, was performed twice yearly.

Table 1. *T. whipplei* treatment failures

Failure type	Description
Immediate failure (occurring after <3 months of treatment)	caused by IRIS[40]
Late failure (occurring after >3 months of treatment)	caused by clinically acquired resistance during treatment[10]
Relapse (after cessation of treatment)	caused by the same strain of *T. whipplei*[19]
Reinfection (after cessation of treatment)	caused by another strain of *T. whipplei*[29]

Definition of failure

Four clinical statuses were distinguished: (i) immediate failure, occurring within 3 months from the start of treatment; (ii) late failure in treated patients defined as symptom reappearance during treatment; (iii) relapse, defined as a reappearance of *T. whipplei* infection following treatment cessation; and (iv) reinfection, defined as the reappearance of clinical manifestations due to a genetically different strain of *T. whipplei* (Table 1).[17]

Biological data

PCR

The molecular detection of *T. whipplei* was performed using real-time quantitative PCR (qPCR).[31] Prior to October 2001, conventional PCR was used as previously described.[31] From October 2001 to September 2003, the specimens were analysed by targeting the 16S–23S rRNA gene intergenic spacer and the *rpoB* gene, as described elsewhere.[32] When an amplified product was detected, sequencing was also systematically performed.[31] From October 2003 to March 2004, the specimens were tested by targeting repeated sequences of *T. whipplei*, as reported previously.[33] After April 2004, these repeated sequences were detected using specific oligonucleotide TaqMan probes for *T. whipplei* identification.[34] A positive result was defined as two positive qPCR results in assays targeting two different *T. whipplei* DNA sequences. Both positive (Twist-Marseille strain) and negative (sterile water) controls were used systematically. The human actin gene was also detected in parallel to verify the quality of the extracted DNA.[31]

Genotyping

A genotyping system based on four highly variable genetic sequences (HVGS: TW133, ProS, SecA, Pro184) found by a genome comparison of two different genomes (strains Twist and TW08/27) was developed.[35–37] This system has since been applied by our laboratory to different samples

because it showed a higher discriminatory power than prior typing methods, such as 16S–23S rRNA, hsp65, variable number tandem repeat (VNTR) and intergenic spacer (ITS) typing, and enabled us to increase typing resolution with respect to the epidemiological behaviour of this pathogen at the molecular level.[38]

Follow-up

Each patient assessed in this study had at least one clinical examination per year. We performed saliva and stool PCR assays at least twice a year. If the PCR was positive, we performed genotyping as soon as possible. Until 2010, we recommended performing a small-bowel biopsy each year that included PAS staining and immunohistochemistry. As we treated our patients for their lifetime, we performed stool and saliva PCR twice yearly and duodenal biopsies only in cases of clinical failure. Finally, we monitored the serum concentrations of antibiotics during maintenance treatment and then during lifetime prophylaxis (with objectives of 4 mg/L for doxycycline, 0.8 mg/L for hydroxychloroquine and 100 mg/L for sulfamethoxazole).

Cure criteria

The criteria have evolved in parallel with our knowledge. Most of the patients referred to us for management after treatment failure with trimethoprim/sulfamethoxazole were treated for 1 year, and cessation of treatment was decided based on the absence of clinical manifestations. Until 2010 and the report of a reinfection caused by a new strain of *T. whipplei*, we recommended ceasing treatment after obtaining full clearance of any macrophagic bacteria on histological analysis, highlighted by both negative PAS staining and immunohistochemistry.[29] Henceforth, we believe that it is not possible to avoid reinfections because of the lifetime susceptibility to *T. whipplei*, and we therefore proposed prophylaxis.[29]

Statistical analysis

Proportions were compared using the bilateral χ^2 test. Survival analysis (time free of failure) was performed according to the induction treatment or first maintenance therapy. Univariate analysis was performed using Kaplan–Meier curves and the log-rank test. Two Cox regression analyses were performed using a Cox regression model; the first analysis systematically incorporated age, sex and induction treatment, and the second included age, sex and maintenance therapy (doxycycline and hydroxychloroquine versus trimethoprim/sulfamethoxazole). A difference was considered significant at $P<0.05$. All analyses were performed using R software version 2.15.2.

Results

Patient characteristics

Among the 29 patients, 23 were male (79.3%). At the time of diagnosis, their mean age was 55 years (range 26–78 years). The mean interval between the diagnosis date and the last follow-up was 78.9 months (range 7–264 months). Currently, 1 patient has died, 25 patients have been regularly followed, and we have ceased monitoring 3 patients. The mean delay between the first symptoms and a positive diagnosis was 58 months (range 1–240 months). Nineteen of the 29 patients (65%) had a previous erroneous diagnosis of inflammatory rheumatism, 2 patients (7%) were diagnosed with sarcoidosis and 1 patient was diagnosed with giant cell arteritis (3.5%). Of the 29 patients with classic Whipple's disease at the time of diagnosis, most (59%) were treated with immunosuppressive drugs, 16 (55%) with corticosteroids and 6 (20%) with tumour necrosis factor inhibitors (Table 2).

Table 2. Clinical characteristics of the 29 patients

Clinical characteristics	No. (% or mean)
Sex male	23 (79.3%)
Age (years)	26–78 (55)
Duration of follow-up (months)	7–264 (78.9)
Time from first symptoms to diagnosis (months)	1–240 (58)
First erroneous diagnosis	
inflammatory rheumatism	19 (65%)
sarcoidosis	2 (7%)
giant cell arteritis	1 (3.5%)
Previous immunosuppressive treatment	17 (59%)
corticosteroids	16 (55%)
TNF-α inhibitors	6 (20%)
Arthralgia	26 (90%)
Diarrhoea	22 (76%)
Weight loss	22 (76%)
Positive PAS and or IHC on small-bowel biopsy	29 (100%)
Positive PCR from small-bowel biopsy	16/18 (88%)
Positive PCR from stool sample	14/15 (93%)
Positive PCR from saliva sample	13/15 (86%)
Positive PCR from CSF sample	4/12 (33%)

TNF, tumour necrosis factor; IHC, immunohistochemistry.

Clinically, 26 patients suffered from arthralgia (90%), 22 (76%) from weight loss (mean 8 kg, range 5–25 kg) and 22 (76%) from diarrhoea. Diagnosis was based on positive PAS staining and/or immunohistochemistry performed on small-bowel biopsies in all patients. At the time of initial diagnosis, *T. whipplei* PCR was positive in 16/18 (88%) small-bowel biopsies, 14/15 (93%) stool samples and 13/15 (86%) saliva samples. Four of the 12 CSF samples tested by PCR were positive (Table 2).

General data regarding treatment

Of the 29 patients, 7 were treated with an initial intravenous regimen. For maintenance treatment, 14 were initially treated with trimethoprim/sulfamethoxazole, 8 were treated with the combination of doxycycline and hydroxychloroquine, 5 were treated with doxycycline and hydroxychloroquine in combination with sulfadiazine or trimethoprim/sulfamethoxazole, 1 was treated with doxycycline, trimethoprim/sulfamethoxazole and rifampicin[19] and 1 was treated with doxycycline alone.[29] Using the definitions of failure given above in the Patients and methods section, none of the 13 patients who were first treated with doxycycline/hydroxychloroquine, doxycycline/hydroxychloroquine and sulfadiazine or trimethoprim/sulfamethoxazole underwent treatment failure, while the other 16 patients developed treatment failure. In detail, the patient who was treated with doxycycline/sulfamethoxazole/rifampicin died, the patient who was treated initially with doxycycline developed seven successive relapses,[29] and of the patients who were first treated with trimethoprim/sulfamethoxazole, two developed an IRIS, six developed clinically acquired resistance and six relapsed after the cessation of treatment (Table 3). One of these patients recently developed uveitis while under lifetime treatment with doxycycline. The serum antibiotic

Table 3. Main characteristics of treatment and outcome of each patient included in this study

Patient	IS	First line of maintenance treatment (duration in months)	PCR saliva/stools after 3 months of treatment	Outcome (time of failure, months)	Last line of maintenance treatment (duration in months)	Outcome (time of failure)	Lifetime treatment with doxycycline (duration in months)	Current outcome (duration of follow-up, months)
1	Y	D+H (48)	−/−	no failure	NA	NA	Y (18)	no failure
2	N	SXT (12)	NA	relapse (60)	D+H (19)	no failure	N	loss of follow-up (24)
3	Y	D+SXT+RIF (1)	NA	death	NA	NA	NA	death
4	Y	SXT (1.5)	−/−	IRIS (1.5)	D+H (36)	no failure	Y (12)	no failure
5	Y	SXT (8)	NA	acquired resistance (8)	D+H (44)[a]	no failure	Y (24)	no failure
6	Y	SXT (10)	NA	acquired resistance (10)	D+H (48)	no failure	Y (25)	no failure
7	N	D+H+SXT or Ad (15)	−/−	no failure	NA	NA	Y (23)	no failure
8	Y	SXT (9)	−/−	acquired resistance (9)	D+H (7)	no failure	Y (48)	no failure
9	N	SXT (22)	−/NA	relapse (40)	D+H (15)	no failure	N	loss to follow-up (18)
10	Y	SXT (18)	NA	relapse (12)	D+H+Ad (30)	no failure	Y (24)	uveitis (non-compliance)
11	N	SXT (12)	NA	relapses[b]	D+H (18)	reinfection (30 months)	Y (25)	no failure
12	Y	SXT (11)	NA	acquired resistance (11)	D+H+Ad (48)	no failure	Y (21)	no failure
13	Y	D (96)	NA	6 successive failures	D+H then AMX	7th failure: reinfection (7)	Y (22)	no failure
14	Y	D+H (53)	−/−	no failure	N	NA	Y (1)	no failure
15	Y	D+H (18)	NA	no failure	N	NA	Y (26)	no failure
16	N	D+H (5, in progress)	−/−	no failure	N	NA	NA	no failure D+H in progress
17	N	SXT (24)	NA	relapse (96)	D+H (15)	no failure	Y (24)	no failure
18	Y	SXT (6)	NA	acquired resistance (6)	D+H (24)	no failure	Y (24)	no failure
19	N	SXT (12)	NA	relapse (36)	D+H (36)	no failure	N	loss to follow-up (36)
20	N	D+H+SXT or Ad (36)	−/−	no failure	N	NA	Y (30)	no failure
21	Y	D+H+SXT or Ad (24)	−/−	no failure	N	NA	Y (25)	no failure
22	Y	D+H+SXT or Ad (12)	−/−	no failure	N	NA	Y (15)	no failure
23	N	SXT (19)	−/−	acquired resistance (19)	D+H (36)	no failure	N	no failure (48)
24	N	D+H+SXT or Ad (30)	−/−	no failure	N	NA	Y (22)	no failure
25	N	D+H (47)	−/−	no failure	N	NA	Y (24)	no failure
26	N	D+H (84)	−/−	no failure	N	NA	Y (5)	no failure
27	Y	D+H (48)	−/−	no failure	N	NA	Y (1)	no failure
28	Y	SXT (1)	−/−	IRIS (1)	D+H (24)	no failure	Y (22)	no failure
29	Y	D+H (36)	−/−	no failure	N	NA	N	no failure (50)

N, no; Y, yes; D, doxycycline; H, hydroxychloroquine; Ad, sulfadiazine; SXT, trimethoprim/sulfamethoxazole; RIF, rifampicin; AMX, amoxicillin, NA, not available; IS, immunosuppressive treatment.

[a]The *T. whipplei* PCR performed on the CSF sample of this patient was positive at the time of the failure. He has been treated with doxycycline and hydroxychloroquine because of the trimethoprim/sulfamethoxazole failure.

[b]This patient experienced two successive relapses, 2 and 40 months after cessation of treatment.

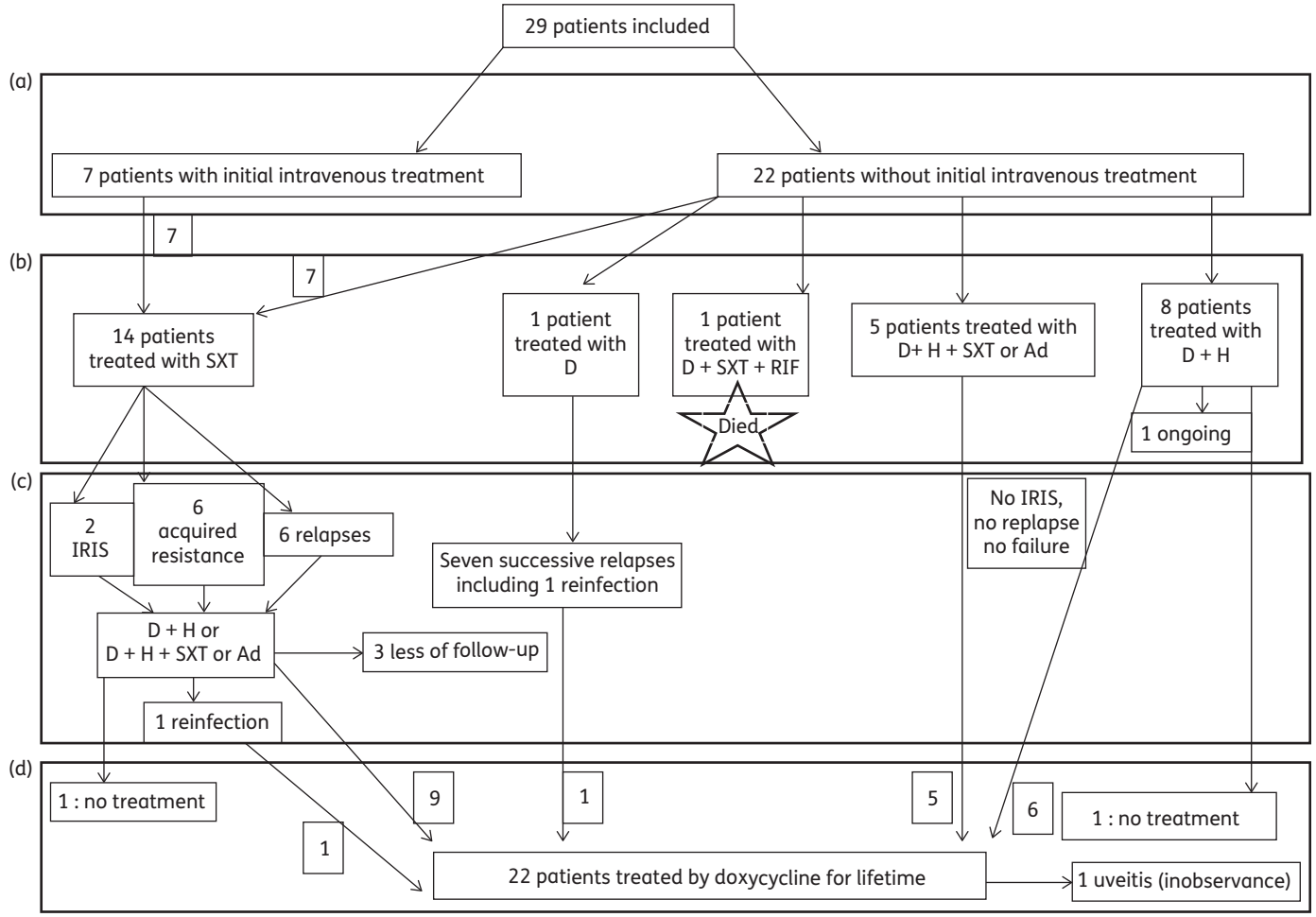

Figure 1. Treatment and follow-up of the 29 patients. (a) induction treatment; (b) first maintenance treatment; (c) other lines of treatment; (d) current treatment. SXT, trimethoprim/sulfamethoxazole; D, doxycycline; H, hydroxychloroquine; Ad, sulfadiazine; RIF, rifampicin.

concentrations demonstrated inadequate compliance with doxycycline treatment in this case. All of these data are summarized in Figure 1.

Side effects

One patient treated with trimethoprim/sulfamethoxazole presented with toxidermia, thus necessitating the cessation of treatment.[19,29] None of the patients treated with the combination of doxycycline and hydroxychloroquine presented side effects that necessitated ceasing treatment. Nevertheless, photosensitization caused by doxycycline necessitated caution for most of the patients, but no treatment had to be stopped.

Evaluation of intravenous therapy before the first antibiotic regimen

Among the 29 patients, 7 were initially treated with intravenous antibiotics. Four patients were treated with ceftriaxone, one was treated with ceftriaxone and gentamicin, one was treated with amoxicillin and gentamicin and one was treated with piperacillin/tazobactam for a period of 15–30 days. Induction treatment was associated with an increased risk of failure (6/7 versus 10/22), as indicated by Kaplan–Meier curves (Figure 2), a log rank test ($P=0.0282$) and in a Cox regression analysis adjusted for age, sex and the presence of an induction treatment [hazard ratio (HR) 3.16, 95% CI 0.11–0.92, $P=0.035$].

Evaluation of the first-line treatment against classic Whipple's disease

A regimen with doxycycline and hydroxychloroquine (eight patients) or doxycycline, hydroxychloroquine and trimethoprim/sulfamethoxazole or sulfadiazine (five patients) was associated with a better outcome, as no failure occurred in this group (0/13), whereas 14/14 failures were reported in the trimethoprim/sulfamethoxazole group. This result was confirmed by the examination of the Kaplan–Meier curve (Figure 3) and the log-rank test ($P<0.0001$). Among the 16 patients who experienced a treatment failure, 11, including the 2 patients who presented an IRIS, had been previously treated with an immunosuppressive treatment. None of the three patients who had a positive *T. whipplei* PCR in CSF and were treated with doxycycline, hydroxychloroquine and trimethoprim/sulfamethoxazole or

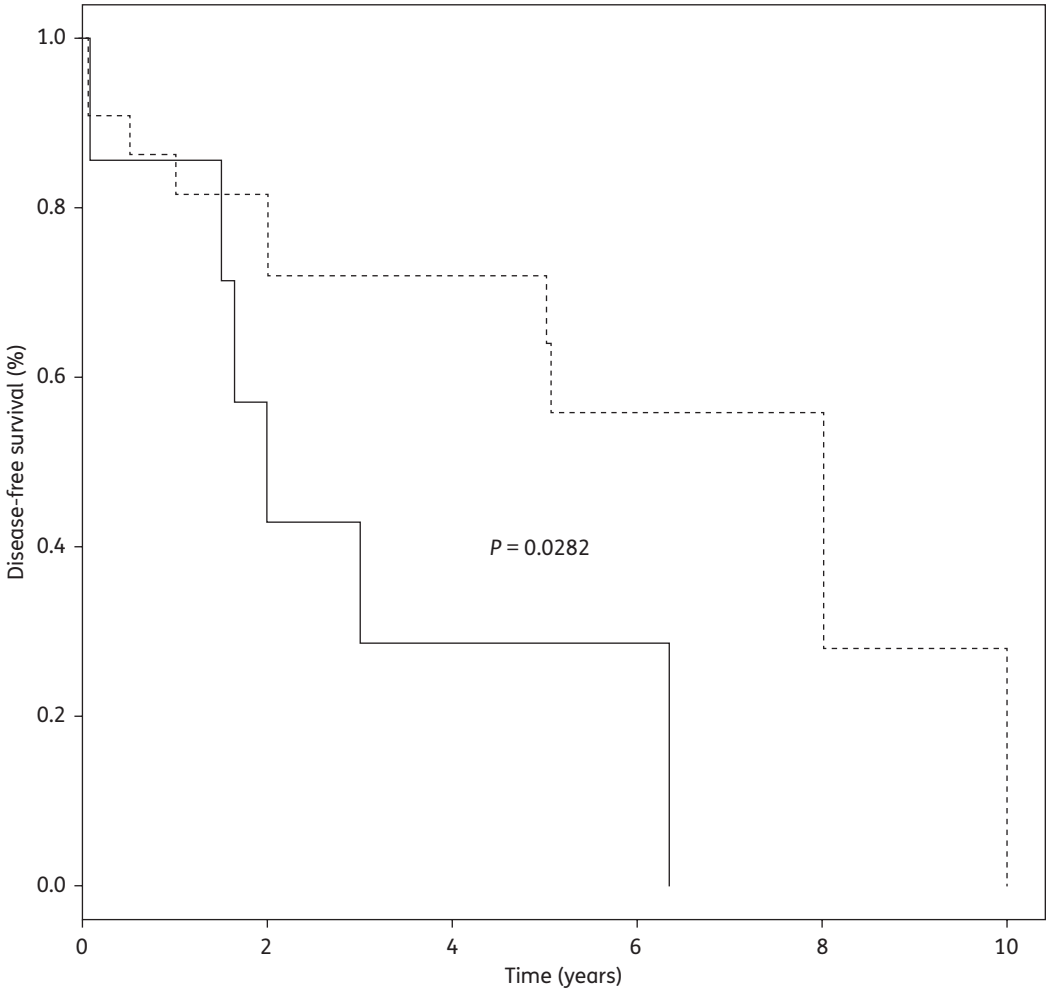

Figure 2. Kaplan–Meier curve comparing induction treatment (continuous line) and no induction treatment (dashed line).

sulfadiazine developed a relapse except for one patient treated with trimethoprim/sulfamethoxazole.

Evaluation of follow-up treatment in patients who had an initial IRIS, failure or relapse

Among the 16 patients who experienced an IRIS, clinical resistance or failure, 1 died[19] and 1 presented seven successive failures, including reinfection with another strain[29] (Figure 1). The 14 patients previously treated with trimethoprim/sulfamethoxazole were ultimately treated with doxycycline/hydroxychloroquine ± sulfadiazine (Table 3). Among them, three patients were lost to follow-up, one patient was currently without antibiotics and without relapse at 3 years of follow-up, and one other patient was reinfected with a genetically different strain of *T. whipplei*. This patient is currently receiving lifetime doxycycline treatment. In addition, one patient who had presented a positive PCR on a CSF sample at the time of failure was effectively treated with doxycycline and hydroxychloroquine, as previously reported (Table 3).[10,19]

Among the 16 patients who experienced an initial failure, in addition to the patient who died and the 3 patients who were lost to follow-up (Figure 1), only 1 patient was currently without antibiotics and 11 patients were treated with doxycycline for their lifetimes.

Among them, only one patient presented with uveitis, 2 years after the cessation of hydroxychloroquine. Nevertheless, the doxycycline serum concentrations revealed that this patient was non-compliant with the medication, thus highlighting the need to monitor the antibiotic serum levels in cases of relapse.

Evaluation of doxycycline for lifetime treatment

Currently, 22 of our 29 patients have been treated with doxycycline, prescribed for their lifetime, for a mean of 20 months (range 1–48 months). None of these patients has had a clinical or a biological relapse, with the exception of the patient presenting with uveitis.

Laboratory monitoring during treatment

After 3 months of initial treatment, negative PCR results were observed in the 16 patients for whom stool samples were tested and in the 17 patients for whom saliva samples were tested.

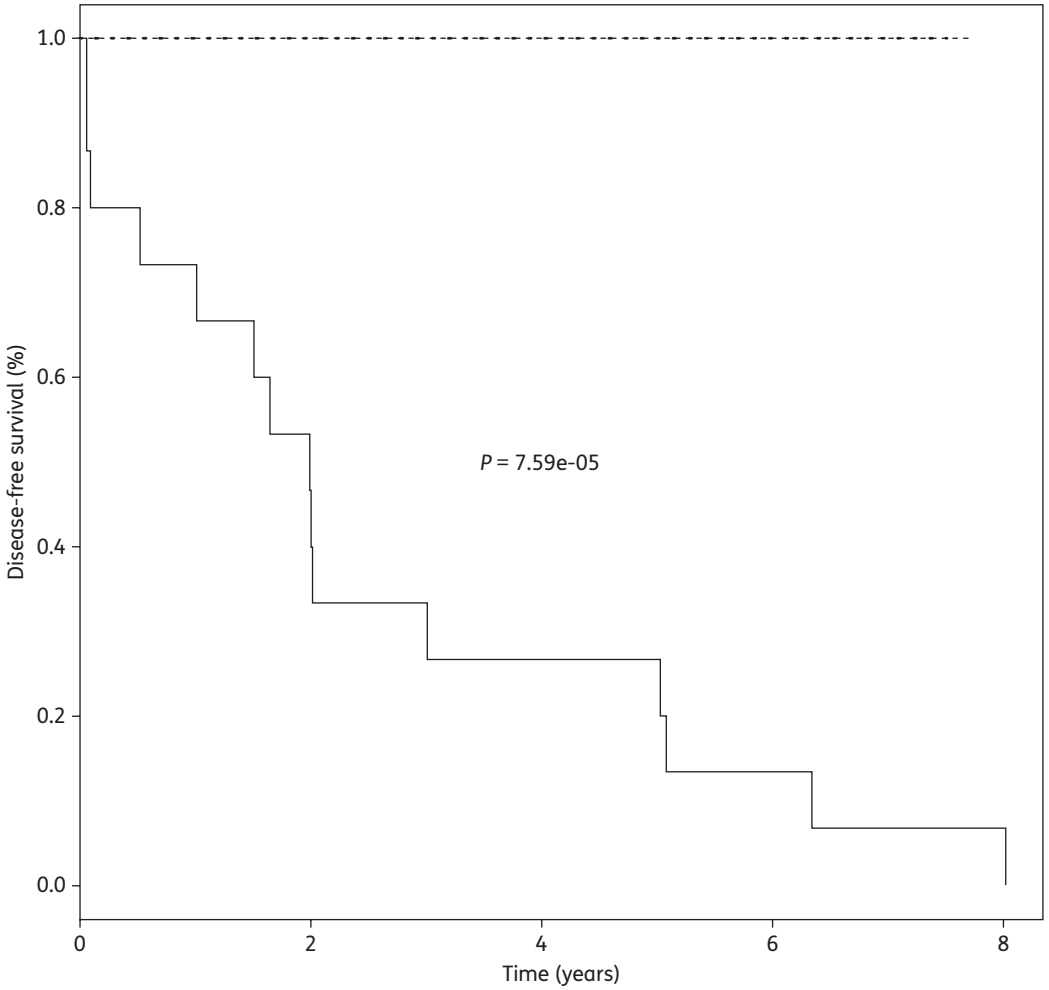

Figure 3. Kaplan–Meier curve comparing first maintenance treatment with hydroxychloroquine and doxycycline (black points) hydroxychloroquine and doxycycline and trimethoprim/sulfamethoxazole or sulfadiazine (dotted line) versus trimethoprim/sulfamethoxazole (continuous line).

Regarding the analysis performed on the small bowel biopsy, a discordance existed between the histological analysis and the PCR results. After at least 1 year of treatment, 16 patients presented with persistence of positive immunohistochemistry, although the PCR performed on the same biopsy was negative.

Discussion

We are confident in these cohort study results because we have much experience in the diagnosis and follow-up of Whipple's disease, with 27 000 specimens tested since the first culturing of the causative bacterium.[31] All PCRs were performed with stringent protocols, and the same pathologist performed all PAS staining and immunohistochemistry examinations.[3] In addition, one of the authors (D. R.) personally followed each patient reported here, enabling standardized management, which is not possible in a multicentre study.[19] Antibiotic serum level monitoring should be systematic, as highlighted by our patient who presented with uveitis because of inattention to doxycycline treatment. Finally, we do not believe that a multicentre randomized study is adequate for a rare disease treated with a lifetime antibiotic regimen. We believe that this seminal study, based on *in vitro* data and the clinical outcome, may serve as a basis for future studies.[12]

Here, in a single-centre study using well-defined diagnostic tools, we report a high level of failure with trimethoprim/sulfamethoxazole, revealing that this combination is definitively not a treatment for Whipple's disease in France.[9,10,19,29] Differences from the results obtained in other studies could be explained by the heterogeneity of management in multicentre studies, differences in strain susceptibility or genetic differences in the hosts.[14,15] We confirmed the patients' clinical outcome and the efficacy of the combination of doxycycline and hydroxychloroquine, which caused no major side effects. Finally, three patients suffered a relapse that was caused by different *T. whipplei* strains (including one not reported in this study), justifying lifetime prophylactic treatment with doxycycline to avoid reinfection,[29] as in other recurrent infections caused by permanent immune defects.[30] In addition, we were able to show the early exaggeration of the disease[2] defined by an IRIS that is better treated with thalidomide than with corticosteroids.[39] This finding is consistent with a recent

study regarding the immunopathology of IRIS that suggested that the mechanism of thalidomide action in these cases is the down-regulation of tumour necrosis factor-α expression, which is critical in IRIS.[39,40]

In conclusion, this is the first study confirming the similarities between *in vitro* results and clinical outcome in classic Whipple's disease. Currently, the recommended treatment for classic Whipple's disease should be a 1 year combined treatment with doxycycline and hydroxychloroquine followed by lifetime treatment with doxycycline. The low number of cases and the need for a very long follow-up for such a disease, which is caused by a lifelong susceptibility, will necessitate the confirmation of these results by other studies.

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Transparency declarations

None to declare.

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