Sicca Syndrome Associated with *Tropheryma whipplei* Intestinal Infection

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Received 22 January 2002/Returned for modification 24 March 2002/Accepted 6 May 2002

The case of a 61-year-old woman with Whipple’s disease-associated sicca complex is reported. *Tropheryma whipplei* infection was diagnosed by histological and ultrastructural examination of the jejunal mucosa and sequence analysis of the bacterial 16S ribosomal DNA. The role of vitamin A malabsorption in sicca complex secondary to Whipple’s disease is discussed.

CASE REPORT

A 61-year-old Caucasian woman was admitted to the Department of Medical Sciences of “La Sapienza” Medical School of Rome, presenting with malaise, asthenia, fatigue, and weight loss (8 kg in the previous 8 months). Her symptoms also included a foreign-body sensation in her eyes, dry mouth, arthralgias, occasional abdominal pain with diarrhea, itching, and dry cough. No history of fever was reported. On physical examination, she appeared pale and emaciated. Skin and mucosae were dry. Mild tenderness and stiffness of feet, ankles, and knees were present, and the liver was moderately enlarged. Laboratory examinations showed microcytic and hypochromic anemia (erythrocytes, 3.7 × 10^12/mm^3; hemoglobin, 98 g/liter; mean corpuscular volume, 79 fl; hematocrit, 29.2%; serum iron level, 280 μg/liter; and serum ferritin level, 53 ng/ml). The platelet count was moderately increased (429 × 10^9/mm^3), while the white blood cell count was normal. The erythrocyte sedimentation rate was 46 mm/h. Hypoalbuminemia (22 g/liter) and decreased total serum proteins (55.7 g/liter) were also observed. The results of the search for antinuclear antibodies, antineutrophilic cytoplasmic antibodies, and Sjögren’s syndrome (SS) autoantibodies to RNA (anti-SSA/Ro and anti-SSB/La) were negative. A mildly positive result for rheumatoid factor (latex fixation test) and Waaler-Rose reaction (10 IU/ml) was observed. The Schirmer’s test showed decreased tear production (4 mm/5 min; normal values, >10 mm/5 min), with evidence of corneal dystrophy. Stool examination showed steatorrhea; neither blood nor protozoa were detected. Routine fecal cultures for enteric pathogens, including enteropathogenic bacteria, acid-fast bacilli, and fungi, were negative. A computerized abdominal tomography scan showed no alterations. Gastrointestinal endoscopy disclosed a mild hyperemia of the gastric antrum, with a negative urease test for *Helicobacter pylori*.

Minimal granulations were observed in the mucosa of the second portion of the duodenum.

Three specimens of enteric mucosa were processed for diagnostic purposes. The first was taken on hospital admission, when initially searching for evidence of gluten intolerance-related enteropathy; the others were taken 2 and 7 months after initiation of antimicrobial therapy. A hematocrit and eosin stain showed the morphological integrity of the jejunal mucosa (data not shown). The most relevant feature was the thickening of the chorion, with extensive infiltration of large and compacted macrophages showing granular, Ziehl-Neelsen-negative, periodic acid Schiff (PAS)-positive, diastase-resistant cytoplasm (Fig. 1A). Ultrastructural examination of the first jejunal specimen showed the presence of numerous elongated free bacilli in either the interstitium or the macrophagic cytoplasm (Fig. 1B). Bacterial cells showed a thick wall covering the membrane, with no interposed periplasmic space, typical of *Actinomyces* (Fig. 1C). The two biopsy specimens taken during antimicrobial therapy showed no significant variations in the histological aspect of the jejunal mucosa, while substantial reduction of the bacterial load and extensive structural damage of intracellular bacteria were observed (data not shown).

To search for the bacterial etiology of the disease, total DNA was extracted from the paraffin-embedded slices of the three jejunal specimens by xylene treatment and cell lysis with lysozyme and proteinase K (14). The nucleic acids were purified with QIAamp DNA-binding columns (Qiagen), and 5-μg samples were used for PCR amplifications with the *Tropheryma whipplei* 16S ribosomal DNA primers W3FE and W2RB (15). Amplicons migrating as a single DNA band of the predicted size (ca. 280 bp) upon agarose gel electrophoresis were obtained from all three samples (data not shown). To confirm bacterial identification, the PCR products from all of the three biopsy specimens were cloned in the pGEM-T Easy vector (Promega) and six randomly selected clones for each PCR product were analyzed by automated DNA sequencing. The nucleotide sequences obtained were the same for all plasmid inserts analyzed, with 100% identity within the 274-bp region 3′ of the 16S rRNA gene of *T. whipplei* (encompassing positions 986 to 1213 of the sequence released under GenBank accession number AF202891).
Subsequent to the diagnosis of Whipple’s disease (WD), the patient was treated with cotrimoxazole (trimethoprim, 160 mg; sulfamethoxazole, 800 mg), orally administered twice daily for 1 year (17). After 2 months of therapy, the patient’s body weight had increased 3 kg without any dietetic change. Arthralgias and gastrointestinal symptoms remitted, and both blood counts and erythrocyte sedimentation rate values returned to normal. After 7 months of therapy, the Schirmer’s test showed a net improvement of tear production (25 to 30 mm/5 min in both eyes) and blood chemistries stabilized at normal values. The patient regained good health and maintained her normal weight for 1 year of follow-up examinations. The result of a search for *T. whipplei* DNA in stool specimens was negative.

SS is a chronic progressive autoimmune disorder characterized by the triad of dry eyes, dry mouth, and autoimmune exocrinopathy (5, 6). In addition to the mouth and salivary glands, other parts of the gastrointestinal system may also be involved (16). The presence of oral and ocular symptoms without aberrant immune function is referred to as sicca syndrome or sicca complex (SC), which appears to behave somewhat differently from classic SS (6, 10). Up to 5% of people over 60 years of age suffer from SS, with a female-to-male incidence ratio of 9:1 (5). As described above, we encountered a patient with clinical manifestations of SC secondary to *T. whipplei* infection.

WD is a rare bacterial infection whose agent has recently been isolated in fibroblastic cultures (13). Clinical manifestations of WD are varied. The infection, which mainly involves the small bowel, may be accompanied by fever, anemia, chronic diarrhea, weight loss, malabsorption, and regional lymphadenopathy (4). Polyarthritis, cardiac manifestations, and central nervous system involvement have been reported (1, 19). The infection rarely occurs in the eyes (e.g., with uveitis, retinitis, and keratitis) or in the lungs, pleura, and skin (4). To our knowledge, SC has not previously been associated with WD.
In our patient’s case, the combination of oral and ocular signs and symptoms with a positive Schirmer’s test result, arthralgia, and fatigue initially suggested the diagnosis of SS. However, the absence of characteristic autoantibodies in the presence of SC manifestations ruled out an autoimmune origin for the disorder. Moreover, the gastrointestinal involvement, which represented a prominent complaint, prompted us to verify the clinical suspicion of WD. In this case, the diagnosis of WD was enforced by the concordance of the three recommended diagnostic criteria (4, 12), namely, the histological and ultrastructural evidence of bacillary forms referable to T. whipplei in the chorion infrate and the specific detection of T. whipplei DNA in biopsy specimens.

Subsequent biopsies, performed with the purpose of monitoring the clinical course during antibacterial chemotherapy, showed the persistence of mucosal lesions at the jejunal level, even though the dramatic reduction of T. whipplei load, both outside and inside cells, correlated with the regression of clinical symptoms. These findings are consistent with the specific detection of T. whipplei 16S ribosomal DNA in all assays of the two biopsy samples taken during the course of chemotherapy.

The unusual association between WD and SC is a novel and clinically relevant finding. Infectious disease-related SS and SC have been documented in patients infected with hepatitis C virus (8), Epstein-Barr virus (6) and H. pylori (2), but the underlying causality has not yet been elucidated. In H. pylori-associated SS, the local and systemic autoimmune response was attributed to cross-reactivity between human and bacterial heat shock protein 60 chaperones (2). It can therefore be speculated that a similar response to the T. whipplei heat shock protein 60 (9) might contribute to the pathogenesis of WD and to the onset of the disease-related inflammatory sequelae. A more plausible explanation for SC in our patient would involve hypovitaminosis A secondary to malabsorption. Many cases of SS and SC have been reported in association with hypovitaminosis, and the therapeutic efficacy of vitamin supplementation is well documented (7, 11, 18). Poor intestinal absorption of dietary fats, which is a hallmark of WD, dramatically reduces the uptake of liposoluble vitamins (3). Remarkably, vitamin A deficiency determines both conjunctival and corneal dystrophy (xerophthalmia and keratomalacia, respectively) and similar degeneration of oral and bronchial epithelia. SC could therefore represent an epiphenomenon of reduced vitamin A absorption secondary to WD; in our patient, the remission of SC following cotrimoxazole therapy strongly supports this link. In conclusion, we propose that SC should be included among the atypical clinical manifestations in patients with an active T. whipplei infection.

We thank A. Petrucca for expert technical assistance. This research was supported by grants from the Italian Ministry of Health, Progetti Finalizzati, and Ricerca Corrente IRCCS to P.V.

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