Tocilizumab for the Treatment of Patients With Refractory Takayasu Arteritis

Yoshikazu Nakaoka, MD, Kaori Higuchi, MD, Yoh Arita, MD, Michio Otsuki, MD, Kaori Yamamoto, BVSc, Takahiro Hashimoto-Kataoka, MD, Taku Yasui, MD, Kuniyasu Ikeoka, MD, Tomohito Ohtani, MD, Yasushi Sakata, MD, Yoshiihi Shima, MD, Atsushi Kumanogoh, MD, Keiko Yamauchi-Takahara, MD, Toshio Tanaka, MD, Tadamitsu Kishimoto, MD, and Issei Komuro, MD

Summary

Treatment of refractory Takayasu arteritis (TA) remains an unresolved clinical issue. Patients usually respond to glucocorticoid (GC) therapy, but often relapse on tapering of the GC dose. The aim of the present study was to assess the safety and efficacy of the interleukin-6 (IL-6) receptor antibody tocilizumab (TCZ) in patients with TA refractory to conventional therapies including GC. Four patients with TA who had shown GC resistance received TCZ infusions (8 mg/kg) every 4 weeks a total of at least 24 times (range, 24 to 51). Clinical symptoms, the serum levels of acute phase proteins and IL-6, GC dosage necessary to maintain remission, and cross-sectional imaging by enhanced CT and MRI were assessed. All patients achieved good clinical response and rapid normalization of the acute phase proteins such as C-reactive protein and serum amyloid A during the therapy with TCZ. The mean dosage of prednisolone could be reduced from 21.3 mg/day to 1.5 mg/day. Although the serum IL-6 level was transiently elevated in all patients after several TCZ infusions, it gradually recovered to the initial level. Along with the decrease of serum IL-6, two patients exhibited significant reduction in thickened arterial lesions. No drug-related adverse effects were noted. In this small group of patients with refractory TA, TCZ therapy was effective and well-tolerated. Further larger studies should be conducted to confirm this finding. (Int Heart J 2013; 54: 405-411)

Key words: Interleukin-6 (IL-6), Cytokine, Inflammation, Monoclonal antibody, Relapsing

Takayasu arteritis (TA) is a chronic inflammatory vasculitis of unknown pathogenesis affecting large vessels, predominantly the aorta and its main branches. Coronary and pulmonary arteries are also affected. Continuous inflammation causes severe vascular damage and formation of wall thickening, fibrosis, stenosis, and thrombus formation, which may lead to fatal vascular accidents.

Glucocorticoids (GC) remain the principal therapy for TA. These drugs suppress the clinical signs and symptoms of inflammation when administered in moderate to high doses, but a sizable number of patients with these conditions relapse upon tapering of the GC dose or discontinuation. Such patients require retreatment and high cumulative doses of GC, resulting in substantial toxicity and morbidity. Thus, immunosuppressive drugs such as methotrexate (MTX), azathioprine (AZA), cyclosporine A (CsA), cyclophosphamide (CYC), and mycophenolate mofetil (MMF) have been studied with an attempt to control the disease activity and lower doses of GC. However, the results of the above immunosuppressive agents in refractory patients with TA are not still satisfactory. A pilot study using tumor necrosis factor (TNF-α)-inhibitors such as infliximab, etanercept, and adalimumab in refractory TA patients reported beneficial effects. However, the efficacy of TNF-α-inhibitors has not been evaluated in randomized trials. Furthermore, one group has recently reported that 33% of the patients experienced disease relapse while receiving TNF-α-inhibitors, suggesting that the efficacy of TNF-α-inhibitors for refractory patients with TA still remains elusive.

Interleukin-6 (IL-6), a pro-inflammatory cytokine, has a crucial role in the pathogenesis of large vessel vasculitis (LVV) including TA and giant cell arteritis (GCA), since the expression level of IL-6 has been reported to be greatly elevated in patients with LVV and to correlate positively with disease activity. In addition, the strong expression of IL-6 was shown...
to actually occur in the aortic tissue of patients with TA. These findings indicate that IL-6 might be a good candidate for a novel therapeutic strategy of TA. Several recent studies have reported that IL-6 receptor (IL-6R) blockade with the IL-6R monoclonal antibody tocilizumab (TCZ) might be effective for the treatment of patients with refractory TA. Since the mean time of TCZ administration in 3 recent pilot studies is a bit short (range, 3 to 12), the safety and efficacy of TCZ for patients with refractory TA need to be examined for longer periods. Here, we assessed the clinical, serological, and cross-sectional imaging outcomes of 4 patients with refractory TA who were administered TCZ more than 24 times.

**Methods**

**Study patients:** From June 2008 to February 2011, we enrolled 4 patients with TA who were diagnosed according to the Guideline for Management of Vasculitis Syndrome (Japanese criteria for Takayasu arteritis). All of these patients fulfilled more than 3 of the 1,990 American College of Rheumatology (ACR) criteria for Takayasu arteritis. Since all these cases were refractory to conventional therapies including GC, or included intolerable adverse effects from these agents, the ethics committee of Osaka University Hospital approved the use of TCZ for these patients, and informed consent was obtained from the patients and their family members after thorough discussion regarding the risk/benefit ratio of this therapeutic option. Before treatment with TCZ, two patients were treated exclusively with GC (cases 2 and 3), one was treated with GC in combination with CYC, AZA, or MTX (case 1), and one was treated with GC in combination with MMF, or CsA (case 4) (Table I).

**TCZ treatment and definition of remission and relapsing:** All of the patients received TCZ (8 mg/kg/body weight) infusions every 4 weeks. Clinical improvement was assessed by evaluating signs and symptoms of disease activity and ability to taper the GC dose. Drug tolerability was also assessed clinically and by laboratory tests at the time of enrollment and subsequently every 4 weeks during the treatment with TCZ.

Remission was defined as the absence of any clinical signs or symptoms of active TA. A relapsing or active disease was defined as the unequivocal presence of signs or symptoms of active TA. Active TA was considered to exist in the setting of new or worsened vascular lesions on the cross-sectional imaging by CT or MRI, new or worsened transient and otherwise unexplained ischemic manifestations, appearance of new bruits or new peripheral pulse asymmetries, and unexplained fever.

**Serological evaluation:** The serum levels of IL-6 were examined at baseline and at each time before TCZ infusion every 4 weeks. Serum specimens from all patients were separated by centrifugation at 3,000 rpm for 15 minutes and stored at -80°C until assayed. A commercial enzyme linked immunosorbent assay (ELISA) kit was used for the measurement of serum IL-6 (R&D Systems, Minneapolis, MN, USA).

Since acute phase proteins such as C-reactive protein (CRP) and serum amyloid A (SAA) are supposed to normalize under adequate IL-6 blockade, the levels of these acute phase proteins were measured at the same time point as the collection of samples for the measurement of serum IL-6 level in order to identify and validate the sufficient dose of TCZ.

**Human leukocyte antigen (HLA) typing:** Genomic DNA was extracted from white blood cells by standard techniques using ethylene diamine tetra-acetic acid. The extracted DNA samples were subjected to HLA genotyping for the detection of HLA-A, B and DRB1 loci by combining polymerase chain reaction (PCR) and sequence-specific oligonucleotide probe (SSOP) protocols with a Luminex 100 flow cytometer to quantitate fluorescently labeled oligonucleotides attached to color-coded microbeads using a Genosearch kit (MBL, Nagoya, Japan).

**Cross-sectional evaluation by enhanced CT and MRI:** Evaluation of the aorta and its branches by enhanced CT and MRI was carried out before initiating TCZ treatment and once a year during the treatment with TCZ to monitor local inflammation in the vessels.

CT images were obtained at end inspiration for all patients using various 64-detector CT by 0.5-0.625 mm collimation. Plain CT scans were performed and 100 mL of nonionic ioxidated contrast material was injected at a flow rate of 2.5 mL/sec. Axial images were reconstructed at 1.0-mm thickness.

MRI images were obtained with a field strength of 1.5T or 3T using a cardiac phased array coil. Cardiac gating black-blood first spin-echo images were acquired with breath-holding before and after contrast material injection at 0.1 mmol per kilogram of body weight with an injector at a rate of 2 mL/sec, followed by a 20-mL saline flush.

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Age</th>
<th>Sex</th>
<th>HLA typing</th>
<th>Historical symptoms observed before TCZ therapy</th>
<th>Disease duration (years)</th>
<th>No. of flare-ups</th>
<th>Prednisolone dosage (mg/day) at the initial TCZ infusion</th>
<th>Therapies used in combination with prednisolone prior to TCZ therapy</th>
<th>Comorbidities</th>
<th>Thickened arterial lesions observed in cross-sectional imaging (CT, MRI, Ultrasonography)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>20</td>
<td>F</td>
<td>A11, A24, B52, B67, DR8, DR15</td>
<td>Cervical pain, Malaise, Fever, Weight loss</td>
<td>1</td>
<td>4</td>
<td>35</td>
<td>CYC (pulse therapy 750 mg x 3 times), AZA (100 mg/day), MTX (6 mg/week)</td>
<td>Hypertension</td>
<td>Carotid, Aortic arch, Celiac artery</td>
</tr>
<tr>
<td>2</td>
<td>29</td>
<td>F</td>
<td>A2, A24, B52, B54, DR4, DR15</td>
<td>Cough, Erythema</td>
<td>2.4</td>
<td>1</td>
<td>18.75</td>
<td>None</td>
<td>None</td>
<td>Carotid, Subclavian, Aortic arch, Ascending aorta, Pulmonary artery</td>
</tr>
<tr>
<td>3</td>
<td>44</td>
<td>M</td>
<td>A2, B35, B51, DR8, DR15</td>
<td>Back pain, Claudication of the arms, Dyspnea, Headache, Dizziness, Back pain</td>
<td>1.7</td>
<td>1</td>
<td>18.75</td>
<td>None</td>
<td>None</td>
<td>Carotid, Subclavian, Vertebral, Superficial femoral artery, Superior mesenteric artery,</td>
</tr>
<tr>
<td>4</td>
<td>22</td>
<td>F</td>
<td>A2, A24, B52, B61, DR9, DR15</td>
<td></td>
<td>10</td>
<td>4</td>
<td>12.5</td>
<td>MMF (2000 mg/day), Osteoporosis, CsA (300 mg/day)</td>
<td>None</td>
<td>None</td>
</tr>
</tbody>
</table>
RESULTS

Baseline characteristics of the patients: The mean age of the 4 refractory TA patients (3 females, 1 male) at the time of TCZ therapy initiation was 29 years (range, 20-44 years). The HLA-B52 allele, which is closely associated with susceptibility to TA in the Japanese population, was positive in 3 patients as shown in Table I.\textsuperscript{22} The mean disease duration was 3.8 years (range, 1-10). The patients were receiving a mean dosage of prednisolone of 21.3 mg/day (range, 12.5-35 mg/day) at the initial infusion of TCZ. The patients experienced an average of 2.5 disease flare-ups which preceded their TCZ treatment (range, 1-4 flare-ups). Substantial cardiovascular comorbidity such as hypertension was found in one patient (case 1), and severe osteoporosis subsequent to the long-lasting GC therapy was observed in one patient (case 4) (Table I). All of the patients exhibited thickened vascular lesions in the thoracic aorta, its branches, or pulmonary arteries, through evaluation by MRI and enhanced CT as shown in Table I. Other baseline clinical characteristics of the patients are shown in Table I.

At the initiation of TCZ treatment, 3 patients presented with constitutional symptoms such as cervical pain and general malaise (case 1), back pain (cases 3 and 4), dizziness (case 4), and claudication of the arms on exercise (case 3) (Table II). The mean disease duration was 3.8 years (range, 1-10). The patients were receiving a mean dosage of prednisolone of 21.3 mg/day (range, 12.5-35 mg/day) at the initial infusion of TCZ. The patients experienced an average of 2.5 disease flare-ups which preceded their TCZ treatment (range, 1-4 flare-ups). Substantial cardiovascular comorbidity such as hypertension was found in one patient (case 1), and severe osteoporosis subsequent to the long-lasting GC therapy was observed in one patient (case 4) (Table I). All of the patients exhibited thickened vascular lesions in the thoracic aorta, its branches, or pulmonary arteries, through evaluation by MRI and enhanced CT as shown in Table I. Other baseline clinical characteristics of the patients are shown in Table I.

At the initiation of TCZ treatment, 3 patients presented with constitutional symptoms such as cervical pain and general malaise (case 1), back pain (cases 3 and 4), dizziness (case 4), and claudication of the arms on exercise (case 3) (Table II). The mean disease duration was 3.8 years (range, 1-10). The patients were receiving a mean dosage of prednisolone of 21.3 mg/day (range, 12.5-35 mg/day) at the initial infusion of TCZ. The patients experienced an average of 2.5 disease flare-ups which preceded their TCZ treatment (range, 1-4 flare-ups). Substantial cardiovascular comorbidity such as hypertension was found in one patient (case 1), and severe osteoporosis subsequent to the long-lasting GC therapy was observed in one patient (case 4) (Table I). All of the patients exhibited thickened vascular lesions in the thoracic aorta, its branches, or pulmonary arteries, through evaluation by MRI and enhanced CT as shown in Table I. Other baseline clinical characteristics of the patients are shown in Table I.

Serological responses to TCZ treatment: Four patients underwent TCZ infusions at Osaka University Hospital for an average of 32 times (range, 24-51 times). The acute phase proteins such as CRP and SAA returned to normal levels in all patients within 1 month after the first TCZ infusion. The concentrations of CRP and SAA declined from 1.37 mg/dL (range, 0.86-2.38) to 0.04 μg/mL (range, 0.04) and 96.3 μg/mL (range, 48-144) to 3.8 μg/mL (range, 3-4), respectively (Table II).

The serum levels of IL-6 were elevated before TCZ treatment in all subjects. The mean pretreatment serum concentration of IL-6 was 15.1 pg/mL (range, 9-21, normal range 0.3-4.0). In all the subjects, the mean IL-6 level further increased to 700.1 pg/mL (range, 83.1-1024) during the TCZ therapy. This increment was observed from 1 to 4 months after the initiation of TCZ administration. However, the mean serum levels of IL-6 gradually decreased during the continued TCZ treatments and returned to 12.7 pg/mL (range, 4.7-21.8), a level that was very similar to the IL-6 level observed before the initial TCZ infusion (Table II, Figures 1 and 2). Although the clinical symptoms, such as cervical pain, back pain and dizziness, had persisted during the period with increased serum IL-6 in cases 1, 3 and 4, these symptoms disappeared along with the decline of serum IL-6 level (Figures 1 and 2).

Clinical responses to TCZ treatment: The cross-sectional imaging by MRI and CT scanning revealed significant reduction in the thickening of vessel walls in 2 patients (cases 1 and 3). Representative follow-up MRI or CT images for case 1 and case 3 are shown in Figure 3 and Figure 4, respectively. Intriguingly, the timing of when the reduction of the thickened arterial lesion was observed coincided with the time when the increased level of serum IL-6 returned to the initial level after several infusions of TCZ (Figure 1 and Figure 2), suggesting that the continued IL-6 blockade by TCZ might have contributed to “reverse remodeling” of the thickened vessels in the patients via ameliorating vascular inflammation. On the other hand, the other 2 patients (cases 2 and 4) exhibited no worsening and no new lesions in the vessels during the TCZ therapy (Table II).

Furthermore, all the patients attained outstanding reductions in the prednisolone doses without any signs of clinical relapse of TA during the TCZ therapy. The mean prednisolone dosage was 21.3 mg/day (range, 12.5-35 mg/day) before starting TCZ therapy and was reduced to 1.5 mg/day (range, 0-4 mg/day) during the TCZ therapy (Table II).

Two patients were still undergoing TCZ treatment every 4 weeks (cases 1 and 4) at the end of follow-up of this study, whereas TCZ was discontinued along with the addition of MTX treatment after 25 and 27 administrations of TCZ in cases 2 and 3, respectively. These two patients are currently being treated with a combination of prednisolone (4 mg/day) and MTX (8 mg/week) and with only MTX (15 mg/week), respectively (Table II).

Adverse events: All patients tolerated the TCZ treatment without infusion reactions. Although TCZ has been reported to be associated with leukopenia and neutropenia,\textsuperscript{22} we did not observe any leukopenia or neutropenia in our subjects. No gastrointestinal perforations were observed. Although we observed no other adverse events related to TCZ treatment, one female patient (case 1) exhibited atypical genital bleeding and subsequent iron deficiency anemia during the TCZ treatment. Her hemoglobin level recovered within two months by oral intake of iron supplements even though TCZ therapy was continued every 4 weeks. This indicates that the atypical genital bleeding was not directly related to TCZ administration in this patient.

One patient (case 2) had a disease flare-up after discon-

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Active features</th>
<th>TCZ infusion (times)</th>
<th>CRP (mg/dL) before and after</th>
<th>SAA (μg/mL) before and after</th>
<th>IL-6 (pg/mL) level at initial/ maximal/ minimal point during TCZ therapy</th>
<th>Improvement of the thickened vessel walls evaluated by MRI or enhanced CT</th>
<th>Prednisolone dosage (mg/day) before and at the end of follow-up</th>
<th>Treatment at the end of follow-up</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Cervical pain, malaise</td>
<td>51</td>
<td>1.04/ &lt; 0.04</td>
<td>144/4</td>
<td>17.3/ 1024/ 4.7</td>
<td>+; (brachiocephalic artery, right common carotid artery)</td>
<td>35/2</td>
<td>TCZ, PSL (2 mg/dL) MTX (7.5 mg/wk)</td>
</tr>
<tr>
<td>2</td>
<td>none</td>
<td>25</td>
<td>1.19/ &lt; 0.04</td>
<td>48/4</td>
<td>21/ 83.1/ 10.1</td>
<td>-</td>
<td>18.75/4</td>
<td>PSL (4 mg/dL) MTX (8 mg/wk)</td>
</tr>
</tbody>
</table>
| 3        | Back pain, Claudi
cation of the arms | 27 | 2.38/ < 0.04 | 101/4 | 13/ 585/ 21.8 | +; (descending aorta) | 18.75/0 | MTX (15 mg/wk) |
| 4        | Back pain, Dizziness | 24 | 0.86/ < 0.04 | 92/ 3 | 9/ 1009/ 14.1 | - | 12.5/0 | TCZ, |
Figure 1. Clinical course of case 1 with refractory TA. The initial administration of TCZ was started on day 0. Changes in doses of prednisolone, MTX, serum levels of CRP and IL-6 are shown. The arrows at the top show the time points of MRI tests.

Figure 2. Clinical course of case 3 with refractory TA. The initial administration of TCZ was started on day 0. Changes in serum levels of doses of prednisolone, MTX, CRP and IL-6 are shown. The arrows at the top indicate the time points of CT.

Figure 3. MRI images of case 1. A: Before TCZ therapy, thickening of the vessel walls of the brachiocephalic trunk (arrow), left common carotid artery and left subclavian artery were observed. B: At 1 year after TCZ treatment, there was no significant change in the thickening of the brachiocephalic trunk (arrow). Intriguingly, at 2 years (C) and 3 years (D) after TCZ therapy, significant reduction in the thickening of the brachiocephalic trunk (arrow) was observed.

Figure 4. Enhanced CT images of case 3. A: Before TCZ therapy, thickening of the vessel walls of the descending aorta (arrow) was observed. B and C, A significant reduction in the thickening of the descending aorta (arrow) was observed at 1 year (B) and 2 years (C) after initiating TCZ therapy, compared with before TCZ therapy.
Continuing TCZ therapy. Although she tried to quit TCZ therapy after the 19th infusion of TCZ, dry cough, the symptom originally observed at the onset of TA, recurred at 9 weeks after cessation of TCZ under monotherapy with prednisolone (5 mg/day). After she was treated with TCZ 6 more times, MTX (10 mg/week) and prednisolone (4 mg/day) were prescribed as maintenance therapy. At follow-up 12-months after cessation of TCZ therapy, the patient remained asymptomatic and the serum inflammatory markers were within normal limits. This flare-up event indicated that extreme care should be considered when terminating TCZ treatment.

**Case summaries:** For more thorough information, we provide the case summaries of 2 of the study patients, cases 1 and 3, in Tables I and II.

**Case 1:** A 20-year-old female was admitted to hospital with complaints of cervical pain, general malaise, weight loss, and fever (39°C) in August 2007. She was clinically diagnosed with TA by CT showing marked thickening of the aortic arch, brachiocephalic trunk, and bilateral common carotid arteries in association with elevation of CRP (12.1 mg/dL) (Table I). At a prednisolone dosage of 50 mg/day, she became free from all symptoms and her CRP level normalized. A subsequent reduction in the prednisolone dosage to 35 mg/day was followed by a recurrence of the symptoms. Although high-dose methylprednisolone pulse therapy (1,000 mg/day for 3 consecutive days) and a subsequent increment in the prednisolone dosage to 50 mg/day were temporarily effective, immunosuppressing agents including CYC pulse therapy (1,000 mg/day for two times), AZA (100 mg/day), and MTX (7.5 mg/week), were not effective at reducing the prednisolone dosage under 35 mg/day (Table I). Thus, TCZ was considered as a therapeutic option and the patient was referred to our hospital.

Before the treatment with TCZ, MRI scans showed thickening of vessel walls of the aortic arch, brachiocephalic trunk, bilateral common carotid arteries, and left subclavian artery (Table II and Figure 3A). The serum levels of CRP, SAA, and IL-6 were 1.57 mg/dL, 144 μg/mL, and 17.3 pg/mL, respectively. Her medication was prednisolone (35 mg/day) and methotrexate (7.5 mg/week). TCZ infusion was initiated in June 2008. After a single infusion of TCZ, the serum levels of CRP and SAA were reduced to under 0.04 mg/dL and to 13.1 μg/mL, respectively (Figure 1). The prednisolone dosage was tapered gradually at every TCZ infusion and methotrexate was terminated after the 6th TCZ infusion. After the 12th TCZ administration, she returned to school after a 2-year absence.

When the dosage of prednisolone was reduced to 8.75 mg/day, the patient felt general fatigue and the elevation of CRP was observed, presumably due to the daily rush at school. Although it seemed like a flare-up, no significant increase in serum IL-6 or recurrence of other clinical symptoms including cervical pain and fever were noticed during this event (Figure 1). The temporal increase of prednisolone dosage to 20 mg/day and resumption of MTX effectively improved the symptoms and lowered the serum level of CRP.

The serum level of IL-6 transiently increased to 1,024 pg/mL after the first TCZ infusion, but it gradually decreased and returned to the initial level around the 15th TCZ infusion (Figure 1). Along with the decrease of the serum IL-6 level, a significant reduction in the thickened lesions of the right common carotid artery and brachiocephalic trunk was noticed by MRI examination at 2- and 3-year follow-ups after initiating the TCZ therapy, whereas no change was observed at 1-year follow-up compared with that before TCZ treatment (Figures 1 and 3). These findings suggest that long-term blockade of IL-6 by TCZ had reversed the thickened arterial lesions in this patient.

The dosage of prednisolone was successfully tapered to 3 mg/day without any signs of recurrence, although the anemia (hemoglobin 7.5 g/dL) due to the abnormal genital bleeding and the subsequent elevation of CRP were observed around the 43th TCZ infusion. The transient increase of prednisolone dosage to 10 mg/day and administration of an iron supplement improved the elevation of CRP and anemia, respectively. No recurrence of the symptoms or laboratory data was observed after the genital bleeding. This patient is asymptomatic under continued TCZ therapy in combination with prednisolone (2 mg/day) and MTX (7.5 mg/week) (Table II and Figure 1).

**Case 3:** A 44-year-old male was referred to our hospital in June 2009 with recent onset of dyspnea on effort, claudication and pulselessness in the left arm, and back pain. TA was clinically diagnosed by the presence of elevation of CRP (1.44 mg/dL) and thickened artery lesions in the aortic arch, ascending aorta, descending aorta, and left pulmonary artery revealed by CT scanning. His estimated right ventricular systolic pressure (RVSP) was 81 mmHg on echocardiography and brain natriuretic peptide (BNP) was 150 pg/mL. This indicates that his dyspnea was presumably provoked by right ventricular overload associated with the left pulmonary artery stenotic lesions. He was treated with prednisolone at an initial dosage of 50 mg/day. He was free from all symptoms and the CRP level was normalized at the prednisolone dosage of 50 mg/day. After treatment with prednisolone with gradual tapering for 2 months, estimated RVSP on echocardiography and BNP improved to 30 mmHg and 9.1 pg/mL, respectively. A subsequent reduction in the prednisolone dosage to 18.75 mg/day was followed by a recurrence of the symptoms such as back pain and increase of the CRP level (2.39 mg/dL). The treatment with TCZ was initiated in February 2010. After a single TCZ therapy, the serum levels of CRP and SAA were normalized to under 0.04 mg/dL and to 3 μg/mL. The back pain transiently deteriorated during the first 5 TCZ infusions consistent with the temporal elevation of serum IL-6. However, this symptom gradually improved during the continued TCZ therapy. The prednisolone was tapered off at the time of the 27th TCZ infusion (Figure 2). Furthermore, the CT examination revealed that the thickening of the descending aorta was significantly reduced at one or two years after the initiation of TCZ therapy, compared with that before initiating the TCZ therapy (Figure 4). After TCZ withdrawal, MTX (15 mg/week) was exclusively prescribed as maintenance therapy (Figure 2). At 1-year follow-up after cessation of TCZ therapy, the patient remained asymptomatic and did not show any signs of recurrence.

**Discussion**

We observed good clinical and laboratory responses in our 4 patients with TA refractory to GC and other conventional immunosuppressive agents. The patients in this study had been treated with TCZ more than 24 times and we could confirm the safety and efficacy of TCZ therapy for the treatment of patients with TA refractory to conventional therapies including...
GC. Intriguingly, two patients showed significant reduction in their arterial thickened lesions, in spite of tapering the dosage of GC or discontinuing GC. The responses to TCZ therapy in our patients suggest that IL-6 has a crucial role in the pathogenesis of TA.

IL-6 has been regarded by several previous reports as one of the causative cytokines for the pathogenesis of TA. First, the serum level of IL-6 has been reported to be elevated in active TA patients and to correlate with disease activity. Furthermore, IL-6 was strongly expressed in the aortic tissues of patients with TA. In addition, IL-6 can specifically induce the production of acute-phase proteins such as CRP and SAA and evoke systemic inflammatory symptoms and signs which are specific features of LVV. Our findings suggest that TCZ might be a potential therapeutic agent to ameliorate the inflammation and clinical symptoms of patients with refractory TA.

The responses to TCZ of our patients are consistent with recent reports using TCZ for the treatment of LVV including TA and GCA. In 2008, Nishimoto, et al reported the successful treatment of a patient with refractory TA accompanied by ulcerative colitis with TCZ. TCZ (4 mg/kg/weekly and bi-weekly) rapidly induced improvements in the clinical manifestations and laboratory data, along with partial recovery of the ischemic symptoms. In 2011, Seitz, et al reported that TCZ (8 mg/kg) was also effective in 7 patients with LVV (5 with GCA and 2 with TA) for a mean period of 4.3 months. Of these subjects, 2 who were newly diagnosed underwent TCZ monotherapy and 5 with a relapsing course were able to taper their GC dosage significantly from 29.5 mg/day at the first TCZ administration to 4.5 mg/day at 12 weeks. Evidence of active LVV on baseline magnetic resonance angiography entirely disappeared in 3 GCA patients and improved in 2 TA subjects after 3 months with TCZ treatment. In 2011, Salvareni, et al reported a pilot study of 4 patients with LVV (2 with GCA and 2 with TA) treated with TCZ (8 mg/kg/month) for 6 months followed by MTX maintenance therapy. Two patients were treatment-naive and the other two patients had relapsing disease. Three patients entered complete clinical remission and one case achieved partial response. In addition, all cases showed significant improvement in vascular fluorodeoxyglucose uptake seen on baseline PET-CT. In two relapsing patients who had been treated with GC at initial infusion of TCZ, prednisolone was tapered off or gradually decreased to 2.5 mg/day by the end of the TCZ treatment. Urzony, et al treated 10 patients with GCA (n = 7), TA (n = 2), and PMR (n = 1) with TCZ for a mean period of 7.8 months (range, 4-12 months). All patients entered and maintained clinical remission during TCZ therapy. The mean daily prednisone dosages before and after TCZ initiation were 20.8 mg/day (range, 7-34.3 mg/day) and 4.1 mg/day (range, 0-10.7 mg/day), respectively.

In this study, we treated 4 refractory patients with TA more than 24 times, which is much longer than previous studies reported. We observed a "reverse remodeling" effect of TCZ on the thickened arterial walls in 2 patients in our study through continuous long-term administration of TCZ (Table II, Figure 1 and Figure 2), while we observed no worsening of the thickened arterial lesions in the other 2 patients. Furthermore, GC was tapered off in two patients (cases 3 and 4) and the dosage of GC was gradually decreased to 2 mg/day (case 1) and 3 mg/day (case 2) during this study period (Table II). These findings suggest that the long-term blockade of IL-6 contributes to the improvement of clinical manifestations in patients with TA presumably through inhibition of the vicious inflammatory cycle driven by IL-6.

Although the serum IL-6 levels of the patients in our study increased significantly after beginning TCZ therapy, the long-term treatment with TCZ gradually reduced the serum level of IL-6 (Table II, Figure 1 and Figure 2). This change in IL-6 level is almost identical to that previously reported in other diseases such as Castleman’s disease and rheumatoid arthritis. The transient increase of serum IL-6 can be explained by the disturbance of the receptor-mediated clearance of IL-6 through the occupation of active sites in the IL-6 receptor with TCZ and might reflect the actual amount of IL-6 production in the inflamed arterial walls. Intriguingly, the timing of improvement in the arterial thickened lesions by the imaging test coincided with that of the reduction of serum IL-6 level in all the cases (Figure 1 and Figure 2). However, the reason why cases 2 and 4 did not show any reduction of the thickened arterial lesions remains unclear. The long disease duration observed in case 4 might have hindered TCZ-mediated improvement of vascular lesions due to the strong fibrotic changes. A larger cohort study would be required to elucidate the meaning of the serum IL-6 levels of the patients with TA during TCZ treatment.

IL-6 blockade by TCZ has a profound role in the therapeutics of LVV. In GCA, the vascular inflammation is primarily evoked by pro-inflammatory IL-17-secreting Th17 cells, which have been proven to be activated in active GCA patients. Th17 lymphocytes infiltrate large and medium-sized vessels by the chemotactic guidance of dendritic cells within the vascular walls. IL-6 is specifically required for the differentiation of naïve T cells into the Th17 cells in cooperation with transforming growth factor β (TGF-β), IL-1β, IL-23, IL-21, and TNF-α. IL-17 stimulates macrophages and vascular resident cells within the vascular walls such as fibroblasts, endothelial cells, and smooth muscle cells to produce pro-inflammatory cytokines, including IL-6. This signaling circuit, mediated by IL-6 and Th17 cells, constitutes a positive feedback loop which could be a major potential target of TCZ in patients with GCA. However, the role of Th17 lymphocytes in TA patients has been elusive to date and should be addressed in the near future.

Several groups reported that TNF-α inhibitors are effective for the treatment of refractory cases with TA, whereas the efficacy of TNF-α inhibitors for TA is still controversial due to the fact that 33% of patients experienced disease relapse while receiving TNF-α inhibitors. TNF-α blockade for the patients with rheumatoid arthritis (RA) led to significant reduction of serum IL-6 accompanied by the reduction of serum CRP. This finding suggests that the clinical benefit of TNF-α blockade in RA patients might be evoked through the reduction of IL-6. In addition, IL-6-R blocking antibody MR16-1 was the most effective blocking agent among the antagonists for IL-6-R, TNF-α, and IL-1 β-receptor to inhibit the synergistic induction of human SAA gene upon the combined stimulation with IL-6, TNF-α, and interleukin-1β (IL-1β). In a refractory patient with TA, Nishimoto, et al reported that the serum TNF-α level fluctuated during TCZ therapy while the disease activity was well-controlled. Taken together, these findings indicate that TNF-α is upstream of IL-6 in the inflammatory cytokine cascade and that TNF-α therapy may have a
therapeutic effect in refractory patients with TA via the indirect reduction of IL-6 production.

There are several limitations in our study. First, the small number of patients treated with TCZ; second, the absence of a control group; and third, the retrospective character of the study. Therefore, our results should be interpreted in a careful manner. However, the beneficial effects of TCZ observed suggest that IL-6 blockade by TCZ might be a promising therapeutic strategy for refractory patients with TA, although further studies would be required to elucidate the precise molecular mechanisms involved in the pathogenesis of TA.

ACKNOWLEDGMENT

We would like to thank Dr. Yasushi Fujino, Dr., Dr. Eku Shimosegawa, and Dr. Hiromatsu Suikikawa of Osaka University for their helpful discussions and Akiko Tanyama and Yuka Yoshimoto of Osaka University for their secretarial work.

REFERENCES