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Cerebrospinal fluid levels of cytokines and soluble tumour necrosis factor receptor in acute disseminated encephalomyelitis

We determined the concentrations of interleukin-1β (IL-1β), IL-6, IL-10, interferon-γ (IFN-γ), tumour necrosis factor-α (TNF-α), and soluble TNF receptor 1 (sTNFR1) in CSF from 18 patients with acute disseminated encephalomyelitis (ADEM) to investigate the role of cytokines in the pathogenesis of the disease in the acute stage. Cytokines and sTNFR1 were measured by ELISA. The CSF IL-6, IL-10, TNF-α, and sTNFR1 concentrations were elevated in 16, 13, 3, and 11 of the 18 patients with ADEM, respectively. CSF concentrations of IL-10 and sTNFR1 correlated positively with each other (\(P<0.01\)). Myelin basic protein levels in CSF of the patients with elevated CSF sTNFR1 levels were significantly higher than those in CSF of the patients with normal CSF sTNFR1 levels (\(P<0.05\)). IL-1β and IFN-γ were not elevated in CSF. Our results suggest that IL-6 and TNF-α mediate inflammation in the central nervous systems in ADEM.

We speculated that TNF-α is related to demyelination and that IL-10 is induced to modulate TNF-α-induced inflammation in ADEM. Conclusion: these findings suggest that cytokines such as tumour necrosis factor-α, soluble tumour necrosis factor receptor 1, interleukin-6, and interleukin-10 are related to the pathogenesis of acute disseminated encephalomyelitis in the acute stage.

**Keywords** Acute disseminated encephalomyelitis · Cytokine · Soluble tumour necrosis factor receptor · Cerebrospinal fluid · Myelin basic protein

**Abbreviations** ADEM acute disseminated encephalomyelitis · IFN-γ interferon-γ · IL-1β interleukin-1β · IL-6 interleukin-6 · IL-10 interleukin-10 · MBP myelin basic protein · sTNFR1 soluble tumour necrosis factor receptor 1 · TNF-α tumour necrosis factor-α

**Introduction**

Acute disseminated encephalomyelitis (ADEM) is an acute inflammatory demyelinating disorder of the CNS commonly seen in children and young adults. ADEM is clinically characterised by the acute onset of neurological symptoms including alteration of consciousness, paresis, ataxia, seizures, behavioural changes, and urinary incontinence after a viral infection or immunisation [2]. MRI reveals increased signal intensity in T2-weighted images representing multiple white matter lesions [5]. CSF findings commonly include an elevated level of myelin basic protein (MBP) [2].

Histologically and clinically, ADEM resembles experimental auto-immune encephalomyelitis [4, 16,17]. Therefore, the myelin destruction in ADEM could result from an immune attack on oligodendrocytes [18]. ADEM may be an auto-immune disorder, however, the pathogenesis as yet not well known. To investigate the role of cytokines in the pathogenesis of ADEM, we determined the concentrations of interleukin-1β (IL-1β), IL-6, IL-10, interferon-γ (IFN-γ), tumour necrosis
factor-α (TNF-α), and soluble TNF receptor 1 (sTNFR1) in CSF of ADEM patients.

Subjects and methods

Subjects

Informed consent was obtained from the parents of the patients enrolled in this study. CSF samples were obtained from 18 patients with ADEM and 25 control subjects on admission to our hospital, Kurume University School of Medicine Hospital, Yamagata University School of Medicine Hospital, Akita University School of Medicine Hospital, and Tokyo Metropolitan Children’s Hospital from April 1997 to November 2000.

The diagnosis of ADEM was based on MRI findings consistent with a disseminated demyelinating process with/without an elevated MBP level in CSF (normal level, < 4.0 ng/ml). For a diagnosis of ADEM, T2 signal hyperintensity had to be present in at least two locations (Fig.1). The clinical features of the ADEM patients are presented in Table 1. There were nine males and nine females, aged from 2 years to 26 years (median 5.9 years). The day of onset of neurological symptoms was considered as the first day of illness. CSF samples were taken from the children with ADEM on days 2 to 16 (median 5.5 days) of illness before treatment. The patients with ADEM were treated with high-dose intravenous corticosteroid and recovered without neurological sequelae. The second CSF investigations were performed in 12 of the 18 patients at the convalescent stage (median 13.5 days after the first CSF study).

The control subjects were 25 afebrile and non-infected children with neurological disorders (12 males and 13 females, aged from 2 months to 14 years; median 5.8 years). Their CSF samples were obtained for routine analysis and they had normal CSF cell counts.

Determination of cytokine and sTNFR1 concentrations

The concentrations of IL-1β, IL-6, IL-10, IFN-γ, and TNF-α in CSF were determined with sandwich-type ELISA kits (R&D Systems, Minneapolis, Minn.). The detection limits were 3.9 pg/ml for IL-1β, 3.1 pg/ml for IL-6, 2.0 pg/ml for IL-10, 8 pg/ml for IFN-γ, and 4.4 pg/ml for TNF-α, respectively. The concentration of sTNFR1 in CSF was determined with a sTNFR1 ELISA kit (Bender Medsystems, Vienna, Austria), the detection limit being 0.05 ng/ml.

Statistical analysis

All values are given as mean ± SD. The differences in the results between groups were analysed with the Mann-Whitney U test. Correlations were analysed using the Spearman rank correlation coefficient test.

Results

The concentrations of IL-1β, IL-6, IL-10, IFN-γ, and TNF-α in CSF of control subjects were all below the detection limits. The concentration of sTNFR1 in CSF of control subjects was 0.94 ± 0.39 ng/ml. The concentration of IL-6 in CSF was elevated in 16 of 18 patients with ADEM in the acute stage (Fig.2A). The concentration of IL-10 in CSF was elevated in 13 of the 18 patients (Fig.2B). The CSF TNF-α level was elevated in 3 of the 18 patients (30.6, 6.1, and 5.9 pg/ml). The concentration of sTNFR1 in CSF was elevated in 11 of the 18 patients (Fig.2C). The CSF IFN-γ level was elevated in another of the 18 patients (128 pg/ml). There was no correlation between the IL-6, IL-10, or sTNFR1 level and the CSF protein level or CSF cell count. The CSF IFN-γ level was elevated in another of the 18 patients (128 pg/ml). There was no correlation between the IL-6, IL-10, or sTNFR1 level and the CSF protein level or CSF cell count. The CSF IL-6 and IL-10 levels in the convalescent stage were decreased compared to those in the acute stage (Fig.5).

Fig. 1 A In patient 3, an axial T2-weighted MRI (TR = 3,000 ms, TE = 93 ms) demonstrating high-intensity lesions (arrows). B In patient 4, an axial T2-weighted MRI (TR = 3,000 ms, TE = 93 ms) demonstrating high-intensity lesions (arrows). C In patient 5, an axial T2-weighted FLAIR MRI (TR = 6,000 ms, TE = 150 ms, TI = 2,000 ms) demonstrating high-intensity lesions. D In patient 7, an axial T2-weighted FLAIR MRI (TR = 6,000 ms, TE = 150 ms, TI = 2,000 ms) demonstrating high-intensity lesions