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Epilepsy Research 41 (2000) 205–211

Epilepsy Research

www.elsevier.com/locate/epilepsyres

Interleukin-6 and Interleukin-1 receptor antagonist in cerebrospinal fluid from patients with recent tonic–clonic seizures

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Received 28 January 2000; received in revised form 19 April 2000; accepted 23 April 2000

Abstract

We have previously reported increased concentrations of interleukin (1L)-6 in CSF from patients with tonic–clonic seizures, where increased cytokine production most likely is a consequence of neuronal epileptic activity associated with seizures. The biological effects of IL-6 are mediated by other cytokines, which are studied here in addition to IL-6. The purpose of this study was to analyze levels of soluble cytokines from plasma and CSF from patients with newly developed tonic–clonic seizures. The concentrations of IL-6, IL-1 receptor antagonist (IL-1RA), IL-1β, tumor necrosis factor (TNFα) and nerve growth factor (NGF) were measured from plasma and CSF from 22 patients with newly developed tonic–clonic seizures within 24 h from the seizure and 18 controls. The mean concentrations of IL-6 were significantly increased in CSF ($P < 0.001$) and plasma ($P < 0.01$) after tonic–clonic seizures, there was some indication of increased concentrations of IL-1RA and no significant change in NGF, IL-1β or TNFα. Our study shows that cytokine network is activated in patients after recent tonic–clonic seizures. We provide evidence of intrathecal production of IL-6 associated with electrical seizure activity.

Keywords: Cytokines; Seizures; Interleukin-6; Interleukin-1 receptor antagonist; Growth factors

1. Introduction

Many cytokines and their receptors are expressed in the brain both during normal development and in adulthood, suggesting a crucial function for these factors in brain maturation and

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0920-1211/00/$ - see front matter
PII: S0920-1211(00)00140-6

proper function of neurons and glia, Inflammatory cytokines play a role in central nervous system (CNS) disorders. In stroke (Beamer et al., 1995) and inCNS trauma (McClain et al., 1991) cytokines are produced in response to cellular destruction, in bacterial menigitis by invading inflammatory cells (Waage et al., 1989) and in autoimmune disorders like multiple sclerosis by microglial or other inflammatory cells (Olsson, 1994).

The significance of the cytokine network also in epilepsy is supported by experimental studies, where increased expression of several cytokines has been reported in brains of animals treated with a convulsant agent, kainic acid (Minami et al., 1990, 1990; Nishiyori et al., 1997; Eriksson et al., 1998). Interestingly, application of recombinant interleukin-1 receptor antagonist (IL-1RA) reduced the neuronal damage in kainic acid induced seizures, suggesting that some of the cytokines may be neuroprotective (Panegyres and Hughes, 1998). Kainic acid causes cellular damage in the hippocampus, and cytokine expression may be determined by a response to neuronal injury. We have previously reported increased concentrations of interleukin-6 (IL-6) in CSF from patients with newly developed tonic–clonic seizures without any evidence of infection or CNS trauma, where increased cytokine production most likely is a consequence of neuronal epileptic activity associated with seizures (Peltola et al., 1998).

The biological effects of IL-6 are, however, to a large extent mediated by other cytokines, which are known to be induced by IL-6. In view of this we have here studied concentrations of IL-1RA and nerve growth factor (NGF), which are linked to the functions of IL-1β and IL-6 (Tilg et al., 1994; Kossman et al., 1996; Arend et al., 1998). Furthermore, we have determined the levels of IL-6 at early time points following an occurrence of tonic–clonic seizure in adult patients. None of these patients or controls were included in our previous study. The results show that IL-6 concentrations rapidly increase after seizures with minor effects on IL-1RA and no change in CSF-NGF.

2. Subjects and methods

2.1. Patients

A total of 22 consecutive patients with previously undiagnosed and untreated tonic–clonic or partial secondarily generalized seizures were studied within 24 h after the seizure. Ten patients had recurrent and 12 patients had single seizures. Six patients had seizures brought on by alcohol withdrawal. We excluded patients with seizures associated with electrolyte disturbances, metabolic causes, acute brain diseases or trauma. The mean age in the seizure group was 37 years (range 15–60). After the first epileptic seizure the patients usually underwent EEG and computed tomography (CT) or magnetic resonance imaging (MRI) examinations. Two patients with cerebral tumors (meningioma and glioblastoma multiforme) were diagnosed, while the remaining patients had normal CT/MRI findings. There was no evidence of recent systemic or CNS infection in any of the patients. All patients were fully informed of the risks and potential benefits of the CSF examination, and informed consent was obtained from each subject. The study protocol was approved by the Ethics Committee of Tampere University Hospital.

The control samples were obtained from 18 adult patients (mean age 38 years; range 16–56 years) on whom lumbar puncture (LP) was performed to exclude neurological disease and who yielded normal neurological examination and laboratory findings.

Lumbar CSF was obtained between 09:00 and 14:00. The first 2 ml of CSF was used for routine examination and a further 200 μl for the present study. Blood was collected within 10 min of lumbar puncture in a Vacutainer EDTA vacuum tube and centrifuged at 3000 rpm for 10 min. The plasma and CSF samples were stored at -70°C until analysis. Hemolyzed samples were not included for analysis.

2.2. Interleukins and TNFα

IL-1β, IL-6 and TNFα concentrations in the sample were measured using enzyme linked im-

munosorbent assay (ELISA) kits (Pelikine Compact, Central Laboratory of the Netherlands Red Cross Blood Transfusion Service, Amsterdam, The Netherlands). IL-1RA concentrations were also determined using an ELISA kit (R&D Systems, Minneapolis, MN). The assays were performed according to manufacturers' instructions. The sensitivity of the assay for IL-1β was 0.8 pg/ml; that for IL-6 was 0.4 pg/ml, that for IL-1RA 22 pg/ml and for TNFα 2 pg/ml.

2.3. Nerve growth factor

The concentration of nerve growth factor in the CSF samples was determined using a sensitive two-site NGF-ELISA assay. Briefly, 0.04 μg primary monoclonal anti-NGF antibody 27/21 (Roche, Bromma, Sweden) in 50 mM carbonate buffer was plated to each well in an EIA plate (Dynatech Laboratories, Virginia) and incubated overnight at 4°C. The next day, after extensive washes with washing buffer (50 mM Tris pH 7.0, 150 mM NaCl, 5 mM $MgCl_2$, 0.1% Triton-X), the samples and NGF-standards (0.1–250 pg/ml) were applicated onto the wells after 2 h blocking (1% BSA in carbonate buffer) at room temperature. Three wells were used for each sample and each standard concentration of NGF. Following the overnight incubation and washing, secondary β-galactosidase coupled with an anti-NGF antibody (Roche, Bromma, Sweden) was added in a concentration of 0.1 U/ml to the wells and again incubated overnight. Before adding 200 μM 4-methylumbellifrenyl-β-D-galactoside (MUG) (Sigma, Stockholm, Sweden) to the wells, the plate was first washed with the washing buffer followed by washes with a substrate buffer (100 mM sodium phosphate buffer pH 7.3, 1 mM $MgCl_2$). Fluorescence signal was measured 1, 2 and 3 h after adding MUG. Typically there was a linear correlation between the fluorescence signal and NGF concentration in the range 0.1–250 pg/ml.

2.4. Statistical methods

The mean and standard deviations were calculated for continuous variables. Statistical significance of differences between two groups was tested by independent two-tailed test. Associations between variables were assessed with Pearson's correlation coefficient. All analyses were performed using a microcomputer and the Statistica/Win package (version 5.1; Statsoft, Tulsa, OK). A P value of less than 0.05 was considered statistically significant.

3. Results

The mean concentrations of IL-6 were elevated both in plasma and CSF, and there was also some

Table 1
The mean concentrations of cytokines in cerebrospinal fluid and plasma from patients with recent tonic–clonic seizures ($n = 22$) and controls ($n = 18$)[a]

	Patients CSF (pg/ml)	Controls CSF (pg/ml)	P	Patients plasma (pg/ml)	Controls plasma (pg/ml)	P
IL-1β (mean ± S.D.)	0.04 ± 0.1	0.1 ± 0.3	0.42	0.4 ± 0.6	1.8 ± 5.9	0.27
IL-1RA (mean ± S.D.)	60.4 ± 56.8	33.0 ± 21.4	0.068	680.1 ± 1037.7	274.8 ± 171.0	0.12
TNF-α (mean ± S.D.)	0.1 ± 0.1	0.1 ± 0.1	0.34	0.3 ± 0.5	1.7 ± 6.0	0.26
IL-6 (mean ± S.D.)	22.0 ± 31.9	1.8 ± 0.5	0.001	3.8 ± 3.7	1.3 ± 1.1	0.01
NGF (mean ± S.D.)	14.5 ± 10.6	18.2 ± 11.7	0.32	ND	ND	

[a] CSF, cerebrospinal fluid; IL-1β, interleukin-1β; IL-1RA, interleukin-1 receptor antagonist; TNF-α, tumor necrosis factor-α; IL-6, interleukin-6; NGF, nerve growth factor; S.D., standard deviation; ND, not done.

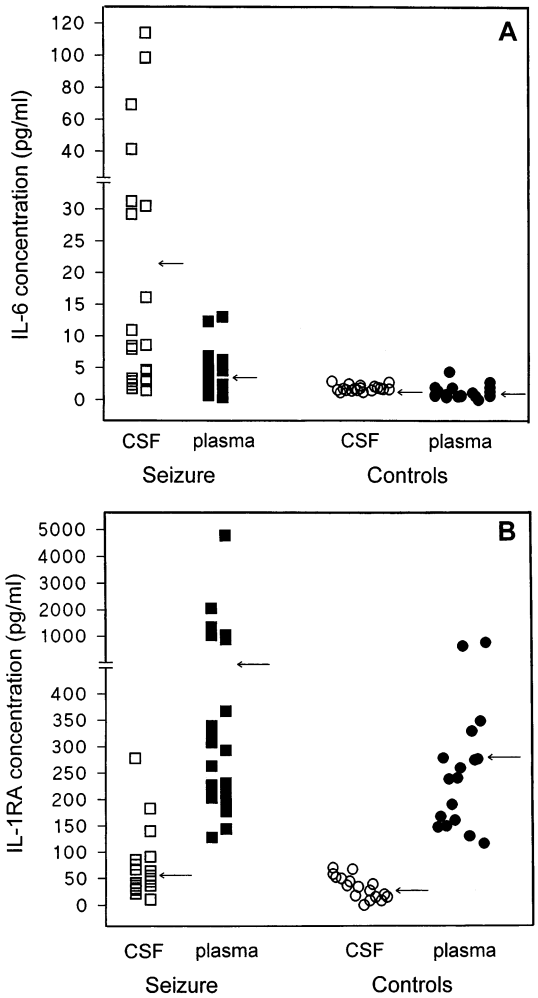

Fig. 1. (A) CSF and plasma concentrations of interleukin-6 from patients with recent tonic–clonic seizures and controls. (B) CSF and plasma concentrations of interleukin-1 receptor antagonist from patients with recent tonic–clonic seizures and controls. The mean concentrations are indicated by an arrow.

indication of increased concentrations of IL-1RA (Table 1). The distribution of plasma and CSF levels of IL-6 and IL-1RA is shown in Fig. 1. The concentrations of IL-6 were greater than 3.7 pg/ml (mean + 2 S.D. for the control group) in 15 out of 22 CSF samples and greater than 2.4 pg/ml (mean + 2 S.D. for the control group) in nine out of 22 plasma samples. The number of elevated values in the seizure group was statistically significant both in CSF ($P < 0.0001$) and plasma ($P = 0.01$). CSF levels of IL-1RA were elevated in five out of 22 patients with seizures, whereas none of the control group showed increased values ($P = 0.057$). In plasma, six out of 22 patients had increased IL-1RA levels compared with two out of 18 control patients ($P = 0.24$), Levels of IL-6 in CSF did not correlate with any other cytokine concentrations in CSF and there was no correlation between plasma and CSF concentrations of IL-6 or IL-1RA. There was a weak correlation between plasma concentrations of IL-1β and IL-1RA ($r = 0.358$, $P = 0.025$). There was no correlation between CSF concentrations of IL-6 and CSF cell counts.

For an evaluation of blood–brain barrier damage, ratios of CSF albumin to serum albumin were investigated, but none of the patients showed increased values (normal < 0.009).

4. Discussion

This study shows an increase in the CSF and plasma concentrations of the cytokine IL-6 after tonic–clonic seizures with a small effect on IL-1RA and no significant change in NGF. The patients did not have any laboratory or clinical evidence of infections. These results confirm and extend our previous findings on IL-6 upregulation following seizures in humans. IL-6 concentration was elevated especially in CSF and plasma concentrations were also increased but to a lesser extent. The mean concentrations of IL-1RA were two-fold compared to controls both in CSF and plasma, but the difference barely failed to reach statistical significance, probably due to the small sample size.

IL-6 is a cytokine known to be rapidly upregulated following different kinds of tissue trauma and inflammation. In CNS, IL-6 was found to be induced within hours in the rat facial nucleus following an axotomy of the facial nerve and well before other cytokines (Kiefer et al., 1993). This suggests that IL-6 may act as an activation signal for other cytokines in brain tissue and IL-6 may be elevated only transiently. In view of this, time is an important factor to consider when determining the CSF levels for IL-6. In our previous study elevated CSF levels of IL-6 were detected in 27%

of patients with seizures occurring within 72 h before sampling, on the other hand all increased IL-6 levels were observed in patients sampled within 15 h from the seizure (Peltola et al., 1998). In this study the percentage of patients with increased concentrations of IL-6 was clearly higher (82%) when patients were sampled within 24 h from the seizure. In experimental studies the expression of IL-6 mRNA was detected 2 h after the seizures and increased at 4 h after the seizure (Minami et al., 1991). It must be kept in mind, however, that animal data on mRNA time courses are not directly comparable to CSF protein levels of cytokines. The elimination of IL-6 from CSF is most likely quite rapid, since in an experimental model where radioligand bound IL-6 was applicated intrathecally, the elimination half-life of IL-6 was 42 min presumably via venous drainage (Banks et al., 1994). Previously CSF and plasma concentrations of IL-6 have been studied in children with febrile seizures; results were within normal limits (Ichiyama et al., 1998). In that study CSF samples were taken on days 1–2 (mean 1.1 ± 0.2), but in our previous study none of the samples studied later than 24 h from the seizure were positive.

As in our previous study, no abnormalities were found in either CSF or plasma concentrations of TNFα and IL-1β. The IL-1 family comprises IL-1α, IL-1β, IL-1 receptor antagonist (IL-1RA) and IL-1 receptors type I and II (IL-1R). IL-1RA downregulates the actions of IL-1β. However, the importance of the IL-1 family in seizures is supported by experimental studies. Increased mRNA levels of IL-1β, IL-1RA and IL-1R have been observed by in situ, hybridization in kainic acid induced seizures (Minami et al., 1990; Nishiyori et al., 1997; Eriksson et al., 1998). In the same model application of recombinant IL-1RA produced neuroprotective effects (Panegyres and Hughes, 1998). Our failure to measure increased concentrations of IL-1β in CSF does not exclude a role for IL-1β in uncomplicated seizures. Several effects of IL-1β are known to have been produced in an autocrine/paracrine fashion by very low quantities of the cytokine, and it is possible that CSF concentration is very insensitive to changes in local production of the cytokine (Schneider et al., 1998). In children with febrile seizures the concentrations of IL-1β have been within normal limits (Lahat et al., 1997; Ichiyama et al., 1998). It has recently been demonstrated, with the use of sensitive techniques, that the expression of the IL-1β gene is affected by physiological changes in the activity of discrete populations of hippocampal neurons in long term potentiation (Schneider et al., 1998). It is therefore plausible that epileptic discharges most likely also cause an activation of IL-1β production in an early phase after the seizure. In kainic acid and pentyleneterazol induced seizures maximal levels of IL-1β mRNA have been found to be expressed in the brain 1.5–3.5 h after the seizures (Minami et al., 1990), but in our study the earliest samples were drawn 4–6 h after the seizure.

In experimental studies, increased IL-1RA expression is detected later than increased expression of IL-1β. Kainic acid induced expression of IL-1RA mRNA was first detected 5 h after kainic acid administration and was markedly increased at 24 h after the seizure (Eriksson et al., 1998). In our study the difference in concentrations of IL-1RA between the seizure and the control group barely failed to reach statistical significance. In other neurological disorders elevated IL-1RA concentrations have been detected more readily than elevated IL-1β levels. Increased values have been measured in stroke, where the mean concentrations of IL-1RA in plasma were of the same magnitude as in the seizure group in our study: 354 pg/ml in stroke patients versus 680 pg/ml in patients with recent seizures (Beamer et al., 1995).

Neurotrophic factors such as NGF are important during development but they are also required for supporting neurons after injury and for the maintenance of neuronal properties. Many cytokines, especially interleukins, have direct neurotrophic effects and they can also stimulate production of certain neurotrophic factors. IL-1 and IL-6 have both been shown to stimulate the production of NGF under various conditions (Lindholm et al., 1987; Lindholm, 1992; Kossman et al., 1996). Increased neurotrophic factor production has been demonstrated in several experimental seizure models (Zafra et al., 1990; Gall, 1993). A recent study reported that a neurite stimulating

effect was induced by the CSF of epileptic patients (Akoev et al., 1996). Likewise, in West syndrome, a severe epileptic syndrome of the infancy, some patients had very high levels of NGF in the CNS (Riikonen et al., 1997). In the present study on adult patients we failed to observe any differences in the NGF levels between the control and the seizure group. However it has to be kept in mind that neither the kinetics nor the mode of production and release of NGF in the adult human brain are known and other time points need to be studied as well.

The cytokines measured in CSF can be either of peripheral blood or intrathecal origin. IL-6 is produced by a variety of cells, including fibroblasts, monocytes, T cells, B cells, microglia, endothelial cells, neurons and astrocytes (Gruol and Nelson, 1997). The mean concentrations of IL-6 in our study were higher in CSF than in plasma (22.0 pg/ml vs. 3.8 pg/ml) supporting the hypothesis of intrathecal production of IL-6. Seizures may increase blood–brain barrier (BBB) permeability, but none of the patients had an elevated albumin CSF/serum ratio, suggesting intact BBB (Ruth, 1984). With IL-1RA, the antagonist, the source of production in our patients is more difficult to assess. The IL-1RA concentrations are ten-fold in plasma compared with CSF, and IL-1RA is known to cross the BBB (Gutierrez et al., 1994). In other neurological disorders elevated levels of cytokines have been measured in different types of CNS trauma or infection. In our patients there was no evidence of CNS trauma or infection, and the mechanism of production is most likely associated with electric brain activity whose duration is usually less than 5 minutes.

Both experimental and human data suggest that cytokine network is activated in association with seizures. Cytokines seem to play a role in normal physiology connected with neuronal activity and NMDA receptor activation, as illuminated with the role of IL-1β in LTP, while LTP is attenuated with application of IL-1RA (Schneider et al., 1998). On the other hand exogenous IL-1β exacerbates neuronal damage induced by glutaminergic receptor activation, whereas external application of recombinant IL-1RA protects from kainic, acid induced neuronal damage (Lawrence et al., 1998; Panegyres and Hughes, 1998). IL-6 can also play a dual role: it prevents NMDA receptor-mediated neurotoxicity in cultured hippocampal neurons, but it also enhances NMDA-induced neurotoxicity and cell death in cerebellar granule neurons (Yamada and Hatanaka, 1994; Qiu et al., 1998). In addition, IL-6 can induce astrogliosis (Yong, 1996). Cytokines can either increase or protect from the neuronal or glial damage associated especially with prolongated seizures.

As shown here, IL-6 and to a lesser extent IL-1RA, increase in CSF from patients with recent tonic–clonic seizure. IL-6 is instrumental as an activating factor in brain tissue contributing to the synthesis and action of other cytokines. Different experimental and human seizure models need to be studied in more detail in order to get a better understanding of the importance of IL-6 and other cytokines in human epilepsy.

Acknowledgements

The authors thank Pekka Tenkilä MA, for reviewing the manuscript for fluency in English.

References

Akoev, G.N., Chalisove, N.I., Ludino, M.I., et al., 1996. Epileptiform activity increases the level of nerve growth factor in cerebrospinal fluid of epileptic patients and in hippocampal neurons in tissue culture. Neuroscience 75, 601–605.

Arend, W.P., Malyak, M., Guthridge, C.J., Gabay, C., 1998. Interleukin-1 receptor antagonist: role in biology. Ann. Rev. Immunol. 16, 27–55.

Banks, W., Kastin, A., Gutierrez, E.G., 1994. Penetration of interleukin-6 across the murine blood–brain barrier. Neurosci. Lett. 179, 53–56.

Beamer, N.B., Coull, B.M., Clark, W.M., Hazel, J.S., Silberger, J.R., 1995. Interleukin 6 and interleukin-1 receptor antagonist in acute stroke. Ann. Neurol. 37, 800–804.

Eriksson, C., Winblad, B., Schultzberg, M., 1998. Kainic acid induced the expression of interleukin-1 receptor antagonist mRNA in the rat brain. Mol. Brain Res. 58, 195–208.

Gall, C., 1993. Seizure induced changes in neurotrophin expression- implications for epilepsy. Exp. Neurol. 124, 150–166.

Gruol, D.L., Nelson, T.E., 1997. Physiologlcal and pathological roles of interleukin-6 in the central nervous system. Mol. Neurobiol. 15, 3307–3339.

Gutierrez, E.G., Banks, W.A.S., Kastin, A.J., 1994. Blood-borne interleukin-1 receptor antagonist crosses the blood–brain barrier. J. Neurolmmunol. 55, 153–160.

Ichiyama, T., Nishikawa, M., Yoshitomi, T., Hayashi, T., Furukawa, S., 1998. Tumor necrosis factor-a, interleukin-1, and interleukin-6 in cerebrospinal fluid from children with prolonged febrile seizures. Neurology 50, 407–411.

Kiefer, R., Lindholm, D., Kreutzberg, G.M., 1993. Interleukin-6 and transforming growth factor-β1 mRNAs are induced in rat facial nucleus following motoneuron axotomy. Eur. J. Neurosci. 5, 775–781.

Kossman, T., Hans, V., Imhof, H.G., Trentz, O., Morganti-Kossman, M.C., 1996. Interleukin-6 released in human cerebrospinal fluid following traumatic brain injury may trigger nerve growth factor production in astrocytes. Brain Res. 7131, 143–152.

Lahat, E., Livine, M., Barr, J., Katz, Y., 1997. Interleukin-1 levels in serum and cerebrospinal fluid of children with febrile seizures. Pediatr. Neurol. 17, 34–36.

Lawrence, C.B., Allan, S.M., Rothwell, N.J., 1998. Interleukin-1β and the interleukin-1 receptor antagonist act in the striatum to modify excitotoxic brain damage in the rat. Eur. J. Neurosci. 110, 1188–1195.

Lindholm, D., Heumann, R., Meyer, M., Thoenen, H., 1987. Interleukin-1 regulates synthesis of nerve growth factor in non-neuronal cells of rat sciatic nerve. Nature 3, 658–659.

Lindholm, D., 1992. Regulation of neurotrophic factor synthesis in CNS by various cytokines. J. Neurochem. 40, 130.

McClain, C., Cohen, D., Phillips, R., Ott, L., Young, B., 1991. Increased plasma and ventricular fluid interleukin-6 levels in patients with head injury. J. Lab. Clin. Med. 118, 1.

Minami, M., Kuraishi, Y., Yamaguchi, T., et al., 1990. Convulsants induce interleukin-1β messenger RNA in rat brain. Blochem. Biophys. Res. Commun. 171, 832–837.

Minami, M., Kuraishi, Y., Satoh, M., 1991. Effects of kainic acid on messenger RNA levels of IL-1α, IL-6, TNFα and LIF in the rat brain. Biochem. Biophys. Res. Commun. 176, 593–598.

Nishiyori, A., Minami, M., Satoh, M., 1997. Type 2 interleukin-1 receptor mRNA is induced by kainic acid in the rat brain. Mol. Brain Res. 501, 237–245.

Olsson, T., 1994. Role of cytokines in multiple sclerosis and experimental autoimmune encephalomyelitis. Eur. J. Neurol. 1, 7–19.

Panegyres, P.K., Hughes, J., 1998. The neuroprotective effects of the recombmant interleukin-1 receptor antagonist after excitotoxic stimulation with kainic acid and its relationship to the amyloid precursor protein gene. J. Neurol. Sci. 154, 123–132.

Peltola, J., Hurme, M., Miettinen, A., Keranen, T., 1998. Elevated levels of interleukin-6 may occur in cerebrospinal fluid from patients with recent epileptic seizures. Epilepsy Res. 31, 129–133.

Qiu, Z., Sweeney, D.D., Netzeband, J., Gruol, D.L., 1998. Chronic interleukin-6 alters NMDA receptor-mediated membrane responses and enhances neurotoxicit developing CNS neurons. J. Neurosci. 1, 10445–10456.

Riikonen, R., Soderstrom, S., Vanhala, R., Ebendal, T., Lindholm, D., 1997. West'syndrome — cerebrospinal fluid NGF and effect of ACTH. Pediatric. Neurol. 17, 224–229.

Schneider, H., Pitossi, F., Balschun, D., Wagner, A., Del Rey, A., Besedovsky, H.O., 1998. A neuromodulatory role of interleukin 1 in the hippocampus. Proc. Natl. Acad. Sci. 95, 7778–7783.

Ruth, R.E., 1984. Increased cerebrovascular permeability to protein during systemic kainic acid seizures. Epilepsia 25, 259–268.

Tilg, H., Trehu, E., Atkins, M.B., Dinarello, C.A., Mier, J.W., 1994. Interleukin-6 (IL-6) as an anti-inflammatory cytokine Induction of circulating IL-1 receptor antagonist and soluble tumor necrosis factor receptor p55. Blood 83, 113–211.

Yamada, M., Hatanaka, H., 1994. Interleukin-6 protects cultured rat hippocampal neurons against glutamate-induced cell death. Brain Res. 643, 173–180.

Yong, V.W., 1996. Cytokines, astrogliosis, and neurotrophism following CNS trauma. In: Ransohoff, R.M., Benveniste, E.N. (Eds.), Cytokines and the CNS. Boca Raton, CRC Press, pp. 309–327.

Waage, A., Halstensen, A., Shalaby, R., et al., 1989. Local production of tumor necrosis factor ct, interleukin 1 and interleukin 6 in meningococcal meningitis. J. Exp. Med. 170, 1859–1867.

Zafra, F., Hengerer, B., Leibrock, J., Thoenen, R., Lindholm, D., 1990. Activity-dependent regulation of BDNF and NGF mRNA in the rat hippocampus is mediated by nonNXMA glutamate receptor. EMBO 9, 545–3550.