

Cytokines in Cerebrospinal Fluid of Children With West Syndrome*

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ABSTRACT

In this study we **aimed** to determine the levels of tumor necrosis factor beta (TNF- β), interleukin (IL) 1-beta (IL-1 β), IL-5, IL-10, and interferon gamma (IFN- γ) in CSF from children during the onset of West syndrome (WS). We observed elevated levels of IL-1 β and IFN- γ correlated to clinical, EEG, therapeutic response, and follow-up suggesting the involvement of immune response in WS. These **results** suggest that inflammatory and immunologic mediators may play a role in the pathophysiologic mechanisms of infantile spasms. Our findings may explain the perfusion and cognitive disfunctions and actions of adrenocorticotrophic hormone (ACTH), corticosteroids, and intravenous immunoglobulin (IVIg) observed in WS. In **conclusion**, WS results from association of neurophysiological mechanisms and structural abnormalities with participation of cytokines mainly in symptomatic group.

Keywords: epilepsy, West syndrome, infantile spasms, cytokines.

RESUMO

Citocinas no líquido cefalorraquiano de crianças com síndrome de West

O objetivo do presente trabalho foi quantificar os níveis do fator de necrose tumoral beta (TNF- β), as interleucinas: 1-beta (IL-1 β), cinco (IL-5) e dez (IL-10) e o interferon gama (IFN- γ), no líquido cefalorraquiano (LCR) de crianças durante o período ictal da síndrome de West (WS). Observamos níveis elevados de IL-1 β e IFN- γ associados aos achados clínicos, EEG, resposta terapêutica e evolução, sugerindo o envolvimento do sistema imune na WS. Os resultados indicam que os mediadores imuno-inflamatórios interferem na fisiopatogênese dos espasmos infantis, justificando as disfunções cognitivas e vasculares e a ação do ACTH, corticosteróides e imunoglobulina no tratamento da WS. Em conclusão, a WS forma sintomática resulta da associação de anormalidades estruturais e neurofisiológicas, com a participação de algumas citocinas pró-inflamatórias, principalmente no grupo sintomático.

Unitermos: epilepsia, síndrome de West, espasmo infantil, citocinas.

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INTRODUCTION

The West syndrome is characterized by infantile spasms, hypsarrhythmia, with or without previously normal psychomotor development. In the symptomatic group multiple etiologies have been described such as focal or diffuse cerebral lesions and anatomical or metabolic disorders or chromosome abnormalities. Cryptogenic group remains with unclear etiology. In both groups, neuropathological and ultrastructural findings are nonspecific, showing none to severe lesions, with or without glial changes secondary to causal etiology.^{1,2,3}

The involvement of the immune system has been described in patients with epilepsies and WS^{4,5,6,7} possibly due to brain lesions⁸ or a common genetically determined susceptibility⁹. Elevated CSF levels of immunoglobulin G were described in WS associated with poor prognosis.¹⁰ On the other hand, the presence of auto-antibodies against brain tissue was not convincingly demonstrated in human epilepsies.⁹

Cytokines are pleiotropic hormones produced by immune competent cells acting as intercellular mediators. They regulate the magnitude and type of effector mechanisms triggered during the host inflammatory reactions and immune response. In this study we aimed to determine the levels of IL-1 β , IL-5, IL-10, TNF- β and IFN- γ in CSF from children during the onset of WS.

SUBJECTS AND METHODS

Patients

We evaluated four infants aged from 4 to 12 months (median 5.5) with a diagnosis of WS. All patients were followed up to the ages 3-4 years. Patients were grouped according to the 1989 Classification. The cryptogenic group included one case without associated etiological factor and the symptomatic group included three patients

with prior signs of brain damage or computed tomography (CT) abnormalities. After the diagnosis, the treatment was started with natural or synthetic ACTH (zinc tetracosactide) 5 IU/kg/day, given intramuscularly.

The CSF specimens were collected for standard procedures, including cell count, and proteins measurement. About 0.5 ml of CSF was stored at -70°C until assayed for IL-1 β , IL-5, IL-10, TNF- β and IFN- γ by ELISA using commercially available monoclonal antibody pairs. Further CSF samples were obtained 4 to 14 days later from 2 children.

Control samples

Control CSF samples were obtained from ten children (aged 1-12 years; mean 8.2) who were evaluated in the emergency room with suspected involvement of CNS.

RESULTS

Clinical analysis

Neurological examination showed truncal hypotonia, microcephaly, and pathologic reflexes with psychomotor regression before onset of WS in three children (symptomatic group). All of these had diffuse cortical atrophy with ventricular enlargement secondary to perinatal asphyxia. A case of cryptogenic group remained without neuropsychomotor commitment. Routine CSF analysis did not show abnormalities in both groups.

All patients were treated with ACTH for 3 to 15 days (mean 10). Seizure remission occurred within 1 to 5 days (mean 3.2). Relapse of infantile spasms was observed in one case (SGS) from the symptomatic group. Chronic treatment was kept with daily valproate. Follow-up showed neurological abnormalities with severe mental retardation in three and normal educational level in the case of the cryptogenic group. All patients were seizure-free at 3 years follow-up. Persistent EEG findings were observed in one case (BRP).

Table 1. Some clinical findings and concentrations of cytokines in CSF of children with WS.

Patient/ Sex	Age	Seizure type before spasms	Age at spasm onset	Duration of ACTH treatment (days)	Time of spasms cessation (days)	Follow-up with improved seizures	Cytokines (pg/ml)				
							IL-1 β	IL-5	IL-10	TNF- β	IFN- γ
SGS/M	12 mo 19 d	none	4 mo	8	5	3 yr 7 mo	72	0	0	0	0
JWSS/M	6 mo 7 d	Clonic neonatal	6 mo	20	5	3 yr 11 mo	132	0	0	0	185
BRP/F	9 mo 23 d	none	1 mo	5	1	3 yr 10 mo	182	13	0	0	218
RMTF/F	5 mo 29 d	none	6 mo	7	2	3 yr 4 mo	0	0	0	0	0
Mean	8.7 mo		4.3 mo	10	3.2	3 yr 8 mo	102	0	0	0	100

Cytokine levels

Concentrations of IL-1 β , IL-5, IL-10, TNF- β and IFN- γ in CSF and some clinical features are detailed in Table 1. Detected IL-1 β levels were observed in all patients from the symptomatic group on admission; two out of those had concentrations higher than 100 pg/ml. Elevated IFN- γ levels were demonstrated in two children. Cytokines were not detected in CSF from control group.

There was no correlation between cytokines and leukocyte count and protein levels in CSF. Both EEG abnormalities and spasms disappeared more rapidly in the cryptogenic case with no detectable cytokine in CSF. Follow-up of cytokine levels were performed in two patients from the symptomatic group. Both boys became seizure-free within 5 days. The case 1 (SGS) showed normalization of IL-1 β levels in 8 days after hospitalization and for case 2 (JWSS) IL-1 β and IFN- γ levels were still detected after 14 days.

DISCUSSION AND CONCLUSION

The studies of cytokines in epileptic syndromes unveil important informations about the interface of immune cells and CNS. We have observed that IL-1 β and IFN- γ are present in CSF from patients with WS during acute phase and may explain some pathophysiologic abnormalities described in previous studies.

Production of IL-1 β by peripheral blood mononuclear cells from patients with epilepsy has been described in interictal period¹¹ and at CNS in experimental models during epileptic seizures.¹² Interleukin-1 also increased slow-wave sleep¹³ and may favors appearance of the hypsarrhythmic patterns seen more often in NREM sleep.¹⁴

As our patients did not have pleocytosis in CSF, elevated levels of IL-1 β and IFN- γ may be associated with glial and inflammatory reactions at sites of brain damage². The results of the various parameters showed correlation between IL-1 β and IFN- γ levels in admission and evolution CSF with the presence of CT scan abnormalities.

Some previously normal patients showed mental deterioration and visual-spatial deficits at the onset of the WS possibly associated with hypoperfusion involving the cerebral cortex^{15,16} or secondary to seizure activity.^{17,18} Experimental studies have demonstrated that IL-1 β may cause perfusion disturbances¹³ and block the processes of learning and memory by inhibiting the long-term potentiating in hippocampus even at low concentrations.¹⁹ One patient (cryptogenic group) had normal psychomotor development and undetectable IL-1 β and IFN- γ levels in CSF. These results suggest that cognitive dysfunctions observed in WS may be related to etiology, site of brain dysfunction, seizure activity, and/or

systemic or local production of this cytokine. Cryptogenic group was associated with a good intellectual development.²⁰

Because multifactorial characteristic spasms are resistant to some antiepileptic drugs and non-conventional treatments with variable success, frequent relapses, and severe side effects. ACTH, corticosteroids²¹ and immunoglobulins have been used empirically and their mechanisms of action still remain unknown.^{4,22,23} Otherwise, corticosteroid therapy have inhibitory effects on IL-1 β production and release^{24,25} and IFN- γ synthesis.²⁶ ACTH is the recommend treatment with better results when started early.²⁷ High-dose IVIg has been indicated in most intractable childhood epilepsies particularly after unsuccessful treatment with ACTH.⁴ Commercially available IVIg preparations contain high transforming growth factor- β (TGF- β 1 and TGF- β 2) concentrations which are potent down regulators of cellular immune response.

All our patients showed good response to ACTH with initial spasms remission between 1 to 5 days and one patient showed relapse with further control. Hypsarrhythmic patterns disappeared and normalization of EEG occurred early in the case with undetectable IL-1 β and IFN- γ levels. We believe that ACTH, corticosteroids or "immunoglobulin" cause favorable outcome in some patients with WS not only by probable action as neuroregulators,²⁴ but also by theirs immune regulatory properties. Activation of microglia may be present in others epileptic syndromes like in Landau-Kleffner syndrome,²⁸ whose clinical and neuroradiologic features are suggestive of cortical disfunction and oligodendrocytes involvement.²⁹

Although pathogenesis of WS remains unknown, our results pointed out to the possible role of cytokines, IL-1 β and IFN- γ , in neurophysiological and structural abnormalities mainly in symptomatic group.

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