VIA MEDICA
Vol. 4, No. 2, pp. 107–111

ISSN 1506–9680
communi-cations

Are CSF neopterin levels a marker of disease activity in multiple sclerosis?

Kostas Kapinas[1], Magda Tsolaki[2], Demetrios Zaphiriou[2], Philip Grammaticos[3], Eleni Dedussi[3]

[1]3rd Department of Neurology, Aristotle University of Thessaloniki, Greece
[2]General Papanikolaou Hospital, Thessaloniki, Greece
[3]Nuclear Department, AHEPA Hospital of Thessaloniki, Greece

Abstract

The study aimed to evaluate neopterin levels in cerebro-spinal fluid (CSF) as a marker of disease activation and progression of multiple sclerosis (MS).

Neopterin, a substance known to be released from macrophages and monocytes at increased rates in cellular immune reactions, was investigated by radio- immunoassay, in the CSF of 19 patients with MS during exacerbations of the disease, in 34 patients with other neurological diseases (OND) and in 20 normal subjects used as controls. Poser's criteria were used for the diagnosis of MS.

Although elevated neopterin levels in the CSF of patients with MS during exacerbations have been reported by other investigators, we found such elevation in only 4 out of 19 patients with MS (21%), in 5 out of 34 patients with OND (14.7%), and in none of the control group. Student's t-test was used for statistical analysis. There was no significant difference in the CSF values of the MS patients, the patients with OND ($p > 0.05$) or the controls.

These results indicate that neopterin levels in CSF may not be considered a marker of disease activity in MS.

Key words: multiple sclerosis, neopterin, cerebro-spinal fluid

Introduction

Neopterin is a pyrazino-2,3 d-pyrimidine derivative formed from quanosintriphosphate (GTP) in the synthesis pathway of biopterin, which acts as a cofactor in neurotransmitter synthesis — i.e., in hydroxylation of phenylalanine, tyrosine and tryptophan [1, 2]. It is released from monocytes and macrophages after stimulation in vitro by gamma interferon (gamma IFN) from activated T cells [3]. Release of neopterin from peripheral blood mononuclear cells in vitro was shown to be induced through activation of T cells stimulated by alloantigens or by virally modified autologous cells [4].

Multiple sclerosis (MS) is a demyelinating disease of the central nervous system (CNS). Although the etiology of MS is still unclear, pathology studies have indicated that infiltrating cells are composed mainly of activated T cells and monocytes-macrophages. Elevated neopterin levels in the CSF of patients with MS during exacerbations have been reported [5, 6], while others did not find differences in CSF levels of neopterin in patients with MS, those with other neurological diseases and healthy controls [7]. Increased neopterin excretion in serum and urine has also been described in patients with viral infections [8, 9], rheumatoid arthritis, systemic lupus erythematosus, coeliac disease [10], malaria [11] and certain tumours [8]. Neopterin has also been used as a biochemical marker of cellular immune response and T-cell activation [1, 11] and to follow the clinical course during different therapeutic trials — e.g., with interleukin-2 in patients with acquired immune deficiency syndrome (AIDS) [12].

The purpose of this study was to determine if neopterin levels are increased in the CSF and if neopterin is a valuable marker of acute cellular immune response and should represent an objective way to monitor disease activity in MS.

Material and methods

Patients

Multiple sclerosis. Nineteen patients (11 female, 8 male, aged 25 to 60 years; mean age 40.4 years) were clinically diagnosed with MS as defined by the criteria of Poser, et al. [13] None of the patients was treated with immunotherapeutic or other related drugs during the study.

Control Subjects with OND. Fifty-four patients (20 female, 34 male, aged 21 to 81 years; mean age 53.7 years) with OND were studied. Of these patients 9 were with cerebral infarctions, 4 with Guillain-Barre syndrome, 4 with polyneuropathy, 8 with dementia, 4 with paraparesis, 2 with Parkinson's disease, 2 with meningioma and 1 with acoustic neurinoma.

Correspondence to: Ass. Prof. Magda Tsolaki
General Papanikolaou Hospital
Exohi, Thessaloniki, Greece
c/o Prof. Philip Grammaticos
Nuclear Medicine Department, AHEPA Hospital of Thessaloniki, Greece
Tel: (+3031) 993 131, fax: (+3031) 994 606,
e-mail: fgrammat@med.auth.gr

Healthy Control Subjects. Twenty individuals (8 female, 12 male, aged 29 to 53 years; mean age 45.7 years) with tension headache or psychoneurotic disorders constituted a control group for establishing the upper reference level of neopterin concentrations in CSF.

CSF samples were taken between 10:00 and 12:00 hours. 15 ml of CSF was obtained by lumbar puncture. Cells, protein and glucose levels in CSF were measured. The remaining CSF was stored at 20° C in a refrigerator where it was protected against light until use (in a Symbol 176\f "Symbol" C). Oligoclonal bands in CSF and serum were determined by agarose gel electrophoresis. Neopterin in CSF was measured with a standard radio immunoassay (RIA) within 6 months after sampling.

The upper reference limit of neopterin in CSF was 2.5 ng/ml. Total and differential counts of CSF cells were also performed. Determinations of IgG and albumin were carried out on unconcentrated CSF. Oligoclonal bands in serum and CSF were tested by agarose gel electrophoresis. Before electrophoresis, CSF was concentrated 80–100 times using an Amicron B10 macrosolute concentrator.

Statistical analysis

Mean neopterin levels in CSF among patients with MS and OND, and healthy control subjects were compared by the Student's t-test and regression analysis.

Results

CSF neopterin levels of 2.5 ng/mL or higher were found in 4 (21%) of 19 patients with MS, in 5 (14.7%) of 34 patients with OND and in none of 20 healthy control subjects. The patients with OND who had an elevated CSF neopterin level included 1 of 4 with paraparesis, 1 of 4 with polyneuropathy, 1 of 4 with Guillain-Barre syndrome, 1 of 8 with dementia (vascular dementia) and 1 of 9 with stroke.

The mean CSF neopterin levels for all MS patients did not significantly differ from those of the controls or of the patients with OND ($p < 0.05$).

Very high or high neopterin levels were found in one patient with paraparesis (14 ng/ml), in one with cervical disc (20 ng/ml), in one with neuropathy (12 ng/ml), in one who had a history of vascular dementia and in one with stroke (3 ng/ml). Very high neopterin levels were also found in MS patients (20, 20.8, and 8.4 ng/ml).

CSF neopterin levels, protein, cells and glucose in MS patients are shown in Table 1. Mean values and standard deviation of all neopterin values in patients and controls are shown in Table 2. Normal neopterin values are up to 2.5 ng/ml.

Table 1. CSF neopterin levels, protein, cells and glucose in MS patients

Patients	Age	Neopterin	Protein	Cells	Glucose
1. AM	25	20	10	0	66
2. DP	30	0.2	30	1	63
3. ZA	30	0.7	18	5	79
4. TP	41	8.4	18	1	66
5. IA	29	0.34	18	1	66
6. KM	33	0.6	77	0	88
7. BE	35	0.4	15	3	75
8. SK	60	8	18	60	71
9. SM	32	0.1	45	15	94
10. FA	60	0.18	20	4	65
11. AA	39	20	10	0	66
12. M	35	0.34	18	3	71
13. SA	38	5	22	0	70
14. PM	28	0.54	28	1	78
15. SG	35	0.6	70	1	55
16. FS	40	0.3	19	0	67
17. PP	38	0.3	25	3	75
18. ED	25	0.2	15	0	73
19. D	32	0.2	28	0	98

Table 2. Mean values and standard deviation of all neopterin levels in patients and controls

Groups	Neopterin [ng/ml]
MS	1.38 ± 3.38
Controls	1.85 ± 4.04
OND	0.34 ± 0.38
	p = 0.13

Discussion

It is known that symptoms and signs will only partly reflect the abnormal CNS test in MS patients. This has been demonstrated when autopsy materials are tested [14, 15] and also when clinical symptoms and signs are correlated to findings from magnetic resonance imaging (MRI) [16, 17]. Jacobs et al. (1986) showed that 75% of lesions detected on MRI were clinically silent and only 5% were definitely related to symptoms and signs.

Elevated serum neopterin levels occur in several acute and chronic infections including HIV, and haematologic malignancies and also during graft rejection [18]. Serum neopterin levels in patients with OND, peripheral neuropathies or MS, who were in clinically stable condition at the time of testing, were not significantly different from levels in healthy controls. However, mean neopterin levels in patients with OND were significantly higher ($p < 0.05$) than levels in patients with MS [7].

In other studies, CSF levels of neopterin were higher in patients with MS during exacerbation [5, 19, 20] and in the relapsing-remitting type of the disease [21]. The significant CSF increase in neopterin described in some references was not reflected in serum [19, 22], suggesting possible intrathecal production of neopterin.

In this study we found no statistical difference in neopterin CSF levels among patients with MS and OND, and the controls. In addition, we found very high neopterin levels in 5 patients with OND: one with paraparesis, one with polyneuropathy, one with Guillain-Barre syndrome, one with vascular dementia and one with stroke. We found no explanation for this finding although it could reflect non-specific immune activation due to damage in the central nervous system or in the peripheral nerves.

Conclusion

In conclusion, it seems that determination of neopterin CSF levels cannot be used as a marker of disease activity in MS patients.

References

1. Huber C, Fuchs D, Niedeiwieser D, et al. Neopterin, eine neue biochemischer marker zur klinischen erfassung ellularter immunoreaction. Klin Wschr 1984, 62: 103.
2. Kern P, Rokos H, Dietrich M. Raised serum neopterin levels and imbalances of T lymphocyte subsets in viral diseases, acquired immune deficiency and related lymphadenopathy syndromes. Biomed Pharmacother 1984, 38: 407–411.
3. Huber C, Batchelor JR, Fuchs D, et al. Immune response associated production of neopterin release from macrophages primarily under control of interferon — gamma. J Exp Med 1984, 160: 310–316.
4. Huber C, Fuchs D, Hausen A, et al. Pteridines as a new marker to detect human T cells activated by allogeneic or modified cells for major histocompatibility complex (MHC) determinants. J Immunol 1983, 130: 1047–1050.
5. Fierz W, Dommasch D, Niederwieser A. Neopterin in cerebrospinal fluid as a parameter of local immune reactions in the central nervous system. In: Lowenthal A, Raus J, eds. Cellular and Humoral Immunological Components of Cerebrospinal Fluid in Multiple Sclerosis. Plenum Press, New York 1987, 369–379.
6. Fredrikson S, Link H, Enoroth P. CSF neopterin as marker of disease activity in multiple sclerosis. Acta Neurol Scand 1987, 75: 352–355.
7. Bansil S, Mithen AF, et al. Elevated neopterin levels in Guillain-Barre syndrome: further evidence of immune activation. Arch Neurol 1992, 49: 1277–1280.
8. Wachter H, Hausen A, Grassmayr K. Erhohte asscheidung von neopterin in harn von patienten mit malignen tumoren und mit viruserkrankungen. Hoppe-Seyer's Z Physiol Chem 1979, 360: 1957–1960.
9. Reibnegger G, Fuchs D, Grubauer H, et al. Neopterin excretion during incubation period, clinical manifestation and reconvalescence of viral infection. In: Biochemical and Clinical Aspects of Pteridines, Walter de Gruyter, Berlin, 433–447.
10. Fuchs D, Granditsch G, Hausen A, et al. Urinary neopterin excretion in coeliac disease. Lancet 1983, 1: 463.
11. Margreiter R, Fuchs D, Hausen A, et al. Neopterin as a new biochemical marker for diagnosis of allograft rejection. Transplantation 1983, 36: 650–653.
12. Kern P, Dietrich M. Increase in serum neopterin levels in patients receiving recombinant interleukin-2 and recombinant interferon alpha. In: Biochemical and Clinical Aspects of Pteridines, vol. 4, Walter de Gruyter, Berlin, 1985, 335–340.
13. Poser MC, Paty, et al. New diagnostic criteria for multiple sclerosis: guidelines for research protocols. Annals of Neurology 1983, 13: 3, 227–231.
14. Gilbert JJ, Sadler M. Unsuspected multiple sclerosis. Arch Neurol 1983, 40: 533–536.
15. Ptiadke JG, Best PV. Atypically and clinically silent multiple sclerosis: a report of 12 cases discovered unexpectedly at necropsy. J Neuro Neurosurg Psychiat 1983, 46: 414–420.
16. Young IR, Hall AS, Pallis CA, et al. Nuclear magnetic resonance imaging of the brain multiple sclerosis. Lancet 1981, 11: 1063–1066.
17. Jacobs L, Kinkel WR, Polachini I, Kunkel RP. Correlations of nuclear magnetic resonance imaging, computerized tomography and clinical profiles in multiple sclerosis. Neurology 1986, 36: 27–34.
18. Fuchs D, Mausen A, Reibnegger G, et al. Neopterin as a marker for activated immunity: application in HIV infection. Immunol Today 1988, 9: 150–159.
19. Fredrikson S. Studies on activation variables in multiple sclerosis. Acta Neurologica, Scandinavica Supp 1988, 115, 77: 10–103.
20. Shaw CE, Dunbar PR, Macaulay HA, et al. Measurement of immune markers in the serum and cerebrospinal fluid of multiple sclerosis patients during clinical remission. J Neurol 1995, 242 (2): 53–58.
21. Ott M, Demisch L, Engelhardt W, et al. Interleukin-2 soluble interleukin-2-receptor, neopterin L-tryptophan and beta 2-microglobulin levels in CSF and serum of patients with relapsing-remitting or chronic progressive multiple sclerosis. J Neurol 1993, 241 (2): 108–114.
22. Fredrikson S, Eneroth P, Link H. Intrathecal production of neopterin in aseptic meningoencephalitis and multiple sclerosis. Clin Exp Immunol 1987, 67 (1): 76–81.