Zagazig University Medical Journal
www.zumj.journals.ekb.eg
https://doi.org/10.21608/zumj.2023.238187.2910 Volume 30, Issue 3, May 2024
Manuscript ID ZUMJ-2309-2910 (R1)
DOI 10.21608/ZUMJ.2023.238187.2910

ORIGINAL ARTICLE

Cerebrospinal Fluid Nitric Oxide as a Diagnostic Inflammatory Marker of Central Nervous System Inflammatory Condition

Alaa Zedan Ibrahim[1], Naglaa Ali Khalifa[2], Sana Abo Abdallah Ahmed Sola[3*], Mohamed RagabAbdellatif[1]
[1] Pediatrics Department, Faculty of Medicine, Zagazig University, Egypt
[2] Clinical Pathology Department, Faculty of Medicine, Zagazig University, Egypt
[3] Pediatrics Department, Faculty of Medicine, Garian University – Libya

***Corresponding author:**
Sana Abo Abdallah Ahmed Sola
Email: ssola9637@gmail.com

Submit Date 2023-09-21
Revise Date 2023-10-03
Accept Date 2023-10-05

ABSTRACT

Background: Incidences of death and morbidity are high worldwide for acute neuroinflammatory disorders like encephalitis. The most helpful fluid for examining brain metabolism is cerebral spinal fluid, which also offers a crucial chance to identify neuro-inflammation in conditions affecting the human central nervous system. This study aimed to assess the role of nitric oxide as a cerebrospinal fluid metabolomics in diagnosis of neuroinflammatory diseases.

methods: This case-control study was conducted in the neurology unit in the pediatric and clinical pathology departments of Zagazig University Hospital on 52 cases with neuroinflammatory conditions, they were equally divided into two groups; Control group included 26 patients (16 males and 10 females) with gray and white matter diseases and a case group of 26 patients (12 males and 14 females) with inflammatory neurological diseases. Cerebrospinal fluid was analysis by ELISA to assay the level of Nitric oxide in samples.

Results: This study revealed that cerebrospinal fluid Nitric oxide levels, along with CSF Protein and CSF Cell Count, were significantly elevated in children with inflammatory neurological diseases compared to those with grey and white matter neurological conditions $p<0.05$. CSF Nitric oxide, with a cut-off level of ≥60.38 (mol/l), emerges as a highly sensitive (100%) and reasonably specific (88.5%) biomarker for discriminating neuroinflammatory diseases in children from other neurological conditions.

Conclusions: Can be used CSF Nitric oxide as a valuable tool for diagnosing and monitoring neuroinflammatory disorders in pediatric patients, offering a potential avenue for early intervention and improved patient care.

Keywords: Cerebrospinal fluid; Central Nervous System; Nitric oxide levels

INTRODUCTION:

Worldwide, acute neuroinflammatory illnesses like encephalitis have high rates of mortality and morbidity **[1,2]**. Inflammation of the CNS is referred to as neuroinflammation, and it can be brought on by infection, autoimmune, traumatic brain damage, toxic metabolites, or degeneration. In cases of acquired inflammation or infection, invading immune cells such infiltrating lymphocytes or monocytes are what trigger the inflammatory response. Additionally, resident immune cells in the brain like microglia can mediate inflammation, which may result in neuronal injury or healing **[3]**.

Encephalitis is estimated to affect 500 000 people per year, with over 100 infectious or autoimmune factors. Invading microorganisms and cells trigger an inflammatory reaction in the central nervous system (CNS), which is necessary for the destruction of neuronal tissue and development of encephalitis. The negative effects of encephalitis have drawn a lot of interest to the study of biomarker development and the pathophysiological mechanisms behind the disorders **[4]**. Clinical signs, neuroimaging, and routine cerebrospinal fluid

(CSF) testing can all be used to diagnose neuroinflammation in certain individuals, however in some people, routine testing comes up negative. There is now growing proof that neurodevelopmental problems cause inflammation of the brain (such as autism spectrum disorder), Psychiatric diseases including schizophrenia and depression, as well as neurodegenerative conditions like Alzheimer's disease. For the purpose of identifying inflammation in specific patients, translatable biomarkers of neuroinflammation are urgently needed. Metabolomics is a method that is quickly developing and is being utilized more frequently as a distinctive "fingerprint" in CNS illnesses. To provide insight into the underlying metabolic mechanisms, this potent instrument examines the variations of endogenous metabolites in biofluids and subsequent chemometrics data management **[5,6].** The most practical matrix for investigating metabolic abnormalities in the brain is CSF **[7].** Liquid chromatography combined with high resolution mass spectrometry has emerged as the method of choice for precisely curating massive volumes of data due to the invention and advancements in high throughput analytical instrumentation **[8-10]).** The goal of this work was to assess the role of nitric oxide as a cerebrospinal fluid metabolomics in diagnosis of neuroinflammatory diseases.

METHODS

After protocol approval by our Local Ethics Committee (IRB # 9195-12-1-2022) faculty of medicine, Zagazig university, this study was carried out in neurology unit in the pediatric and clinical pathology departments at Zagazig University Hospital, during the period from February 2022 to July 2022. It enrolled 52 children with neuroinflammatory conditions. The children were divided into two groups arranged by age (1 month to 9 years). Group I (control group): included 26 patients (16 males and 10 females) with gray and white matter diseases and Group II (Case group): included 26 (12 males and 14 females) with inflammatory neurological diseases. Written informed consent was obtained from all participants parents or relative and this study after telling them the nature of the study. The study's protocol complied with the Helsinki Declaration, which is the World Medical Association's code of ethics for research on humans. The inclusion criteria were age between 1 month and 9 years, both sexes, and patients with neuroinflammatory conditions. Excluded from the criteria were patients who received immune modulatory therapy before the lumbar puncture. Patients with brain tumors, central nervous system trauma, and cerebral palsy.

All participants were subjected to full history taking and full clinical examination including brain Computed tomography (CT)/or magnetic resonance imaging (MRI) and investigation included Complete blood count with differential count by automatic analyzer (XN 330 sysmex). C-reactive protein (CRP) assessed using Cobas c311. Cerebrospinal fluid (CSF) analysis (glucose, total leukocytic count, protein) by enzyme-linked immunosorbent assay (ELISA) to assay the level of Nitric oxide in samples.

Sample collection:

Collect sample of blood about 4 cm, 1cm was taken for Complete Blood Count (CBC) analysis and 3cm left to clot and centrifuge serum were separated assayed for C reactive protein **.**collect sample of CSF, All CSF samples was taken before starting immune modulatory therapy (corticosteroids or intravenous immunoglobulin), but not before treatment with anti-microbials or antiseizure medication. CSF samples were drawn from lumbar puncture (spinal tap) one sample was done cell counter for TLC and was centrifuged immediately to do glucose, protein. Another CSF samples was centrifuged also and stored at -80°C and assayed by enzyme-linked immunosorbent assay (ELISA) for Nitric oxide concentrations were measured using an automated procedure, with a nitric oxide kit based on the Griess reaction**.** After reaction with the Griess reagent, the nitric oxide concentrations (mol/l) were derived spectrophotometrically.

Assessment of nitric oxide by Eliza technique:

The kit uses a double-antibody sandwich enzyme-linked immunosorbent assay (ELISA) to assay the level of Nitric oxide in samples. Add Nitric oxide to monoclonal antibody Enzyme well which is pre-coated with Nitric oxide antibody, incubation; then, add Nitric oxide antibodies labeled with biotin, and combined with Streptavidin-HRP to form immune complex; then carry out incubation and washing again to remove the uncombined enzyme. Then add Chromogen Solution A, B, the color of the liquid changes into the blue, and at the effect of acid, the color finally becomes yellow. The chroma of color and the concentration of the Human Nitric oxide of sample were positively correlated.

STATISTICAL ANALYSIS:

SPSS is used for data management. Released in 2015 by IBM Corp., Version 23.0 of IBM SPSS Statistics for Windows IBM Corp., Armonk, NY

more than two groups of normally distributed variables were compared using the f(Anova)-test. Two groups of non-normally distributed variables were compared using the Mann-Whitney U test. A chi-square test was used to compare the percentage of category variables. Receiver Operating Characteristic (ROC) curve to draw roc curve; the true positive rate (Sensitivity) is plotted on (y) axis and false positive rate (100-Specificity) on(x) axis, optimum cut-off was determined using the Youden index. P-values greater than 0.05 were deemed statistically significant (S), whereas P-values lower than 0.05 were deemed statistically insignificant (NS).

RESULTS:

Table 1; define that Causes of neuro inflammatory diseases in studied children, were (34.6%) viral infection, Bacterial, for each followed autoimmune diseases (30.8%). Diagnosis of neuro inflammatory diseases were 30.8% Meningitis, 23.1% Encephalitis, followed by 11.5% Transverse myelitis, Multiple sclerosis for each, lastly 7.7% Cerebilitis, ADEM, GBS for each.

Table 2; defined that children with inflammatory neurological diseases had significant higher percent positive Kering's sign, unsteady gait $p<0.05$, While there were significant increase irritability, decreased motor power, in addition affecting muscle tone (hypotonia and hypertonia) in children with grey and white matter neurological diseases, $p<0.05$.

Table 3; showed that there was significant difference in imaging in children with inflammatory neurological diseases compared to children with grey and white matter neurological diseases, $p<0.05$. the current table defined those 7 children (26.92%) had Cerebral edema in children with inflammatory neurological diseases, while more percent children with grey and white matter neurological diseases had imaging finding indicate hydrocephalus, dilated ventricles lesions, $p<0.05$. Otherwise, there was no difference in other imaging finding in both groups, $p>0.05$.

Table 4; showed that there was significantly higher value in Cerebrospinal fluid (CSF) Protein, CSF Cell Count (cells/µl), CSF Nitric oxide (mol/l) in children with inflammatory neurological diseases compared to children with grey and white matter neurological diseases, $p<0.05$. Otherwise, there was no difference in other CSF finding in both groups, $p>0.05$.

Table 5; showed that there was no difference Cerebrospinal fluid (CSF) Nitric oxide (mol/l) value regarding causes of inflammatory neurological diseases in studied children, $p>0.05$.

Table 6; showed that cerebrospinal fluid (CSF) Nitric oxide at cut off level ≥60.38(mol/l) show sensitivity 100%, specificity 88.5% and accuracy 94.2% to discriminate neuroinflammatory diseases in children versus grey and white matter neurological diseases.

Figure 1: Receiver operating characteristic (ROC) curves of CSF Nitric oxide (mol/l).

Table 1: Causes and diagnosis of neuro inflammatory diseases.

	Cases group n.26	
	N	%
Causes of neuro inflammatory diseases		
Viral infection	9	34.6
Bacterial infection	9	34.6
Autoimmune	8	30.8
Diagnosis of neuro inflammatory diseases		
Meningitis	8	30.8
Encephalitis	6	23.1
Transverse myelitis	3	11.5
Multiple sclerosis	3	11.5
Cerebilitis	2	7.7
ADEM	2	7.7
GBS	2	7.7

Table 2: Neurological examination of studied children

	Cases group n.26		Control group n.26		χ 2	P- value
	n	%	n	%		
Neck stiffness	11	42.31	5	19.23	3.25	0.071
Kering's sign	8	30.77	0	.00	f	0.004*
Brudzinski's sign	5	19.23	0	.00	f	0.051
Bulged anterior fontanel	8	30.77	12	46.15	1.3	0.25
Irritable	11	42.31	20	76.92	6.5	0.011*
consciousness level						
normal	9	34.61	4	15.38		
Confusion	9	34.61	9	34.61	3.39	0.49
Disorientation	1	3.85	2	7.69		
Delirium	2	7.69	2	7.69		
Lethargy	5	19.23	9	34.61		
Loss of equilibrium	11	42.31	6	23.08	2.18	0.14
Unsteady gait	14	53.85	5	19.23	6.7	0.01*
Muscle tone						
Normotonia	14	53.84	2	7.69		
Hypotonia	7	26.92	13	50.0	17.37	0.0001*
Atonia	2	7.69	0	0.0		
Hypertonia	3	11.54	11	42.31		
Decrease motor power	13	50.00	21	80.77	5.4	0.02*

*χ 2 Chi-square test *P value < 0.05: Significant. P value ≥ 0.05: no significant*

Table 3: Imaging of the studied patients

	Cases group n.26		Control group n.26		χ 2	P- value
	n.	%	n.	%		
Hydrocephalus	3	11.54	10	38.46	5.03	0.025*
Dilated ventricles	2	7.69	8	30.77	4.45	0.035*
Tonsillar herniation	3	11.54	5	19.23	0.59	0.44
Cerebral edema	7	26.92	0	.00	f	0.01*
Space occupying lesion	5	19.23	5	19.23	-	-
Midline shift	4	15.38	2	7.69	f	0.67
Interventricular Hemorrhage	1	3.85	7	26.92	f	0.051
Multifocal lesion in gray matter	2	7.69	7	26.92	f	0.14
Widespread lesion in white matter	4	15.38	9	34.62	2.56	0.11

*χ 2 Chi-square test. f: Fisher exact test*P value < 0.05: Significant. P value ≥ 0.05: no significant*

Table 4: Cerebrospinal fluid (CSF) of the studied patients

	Case Group n.26	Control Group n.26	u-test	P- value
CSF Glucose (mg/dl) Median (range)	53(12-71)	40(1-73)	1.320	0.187
CSF Protein (mg/dl) Median (range)	136.9(60-400)	61(13.9-535)	3.478	0.001*
CSF Cell Count (cells/µl) Median (range)	650(10-8000)	11.5(5-300)	4.286	0.0001*
CSF Nitric oxide (mol/l) Median (range)	86.83(68.9-126)	45(31.37-70.24)	6.040	0.0001*

u: Mann-Whitney U. *Data are expressed as median, Range.*
P value < 0.05: Significant (*). P value ≥ 0.05: no significant

Table 5: Cerebrospinal fluid (CSF) Nitric oxide according to causes

	Viral infection n.11	Bacterial infection n.12	Autoimmune n.3	f-test	P- value
CSF Nitric oxide (mol/l) Mean ± SD	91.9±17.57	88.58±11.89	82.36 ±10.61	1.029	0.373

F: Anova test, P *value ≥ 0.05: no significant*

Table 6: Performance of CSF Nitric oxide in predicting neuroinflammatory diseases in children versus grey and white matter neurological diseases

Cut off level	Studied groups			Sensitivity	Specificity	PPV	NPV	Accuracy
		Cases n.26	control n.26					
CSF Nitric oxide ≥60.38(mol/l)	Cases	26	3	100%	88.5%	89.7%	100%	94.2%
	Control	0	23					

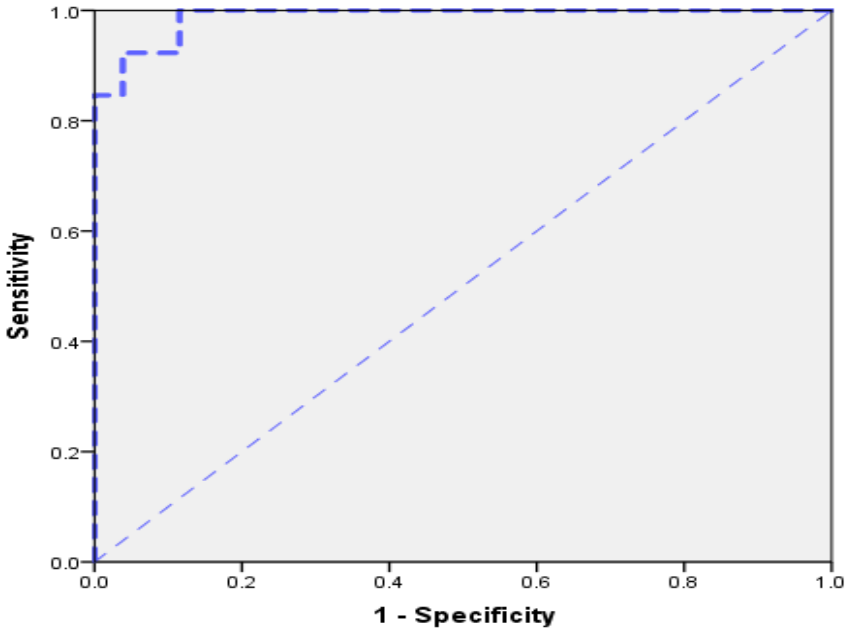

Figure 1: Receiver operating characteristic (ROC) curves of CSF Nitric oxide (mol/l).

The area under the curve (AUC) is 0.988 with 95% confidence interval (0.969-1), p=0.0001, so CSF Nitric oxide (mol/l) was excellent predictor for neuroinflammatory diseases in children

DISCUSSION:

In our study, as regard causes of neuro inflammatory diseases in studied children, were (34.6%) viral infection, Bacterial, for each followed autoimmune diseases (30.8%)**.** Diagnosis of neuro inflammatory diseases were 30.8% Meningitis,23.1% Encephalitis, followed by 11.5% Transverse myelitis, Multiple sclerosis for each, lastly 7.7% Cerebilitis, ADEM, GBS for each.

Curcio et al[11] identified the following diagnoses: acute cerebellar ataxia (ACA) (n=14; 14.3%), acute demyelinating encephalomyelitis (ADEM) (n=13; 13.3%), multiple sclerosis (MS) (n=18; 18.4%), anti-*N*-methyl-D-aspartate receptor encephalitis (anti-NMDAR encephalitis) (n=15; 15.3%), encephalitis not otherwise specified (E-NOS) (n=12; 12.2%), and "Other" (n=26; 26.5%). "Other" included acute transverse myelitis, neuromyelitis Optica, CNS lupus, primary CNS vasculitis, Rasmussen's encephalitis, opsoclonus myoclonus ataxia syndrome, and clinically isolated syndrome.

In our study, there was significant higher percent of headache, back pain, pain in lower limb in children with inflammatory neurological diseases than children with grey and white matter neurological diseases, p>0.05. While there was significant higher percent of delayed motor millstone, convulsion in children with grey and white matter neurological diseases than inflammatory neurological diseases children, p<0.05. Otherwise, there was no difference in disease symptoms in children with inflammatory neurological diseases and children with grey and white matter neurological diseases, p>0.05,

In children, visual and motor impairment, seizures, and constitutional symptoms like fever and vomiting are the most typical presenting symptoms **[12]**. The optic nerve, brainstem, and spinal cord are the most typical locations for a main presentation **[12]**. Vomiting has been recorded as a presenting symptom in 38% of juvenile NMO patients, indicating that NMO can specifically impact the area postrema **[12]**. Most kids with AQP-4 seropositive NMO have at least one episode, it should be highlighted of ON (83%) or LETM (78%) **[13].**

In our study, there was significant difference in imaging in children with inflammatory neurological diseases compared to children with grey and white matter neurological diseases, p<0.05. the current table defined those 7 children (26.92%) had Cerebral edema in children with inflammatory neurological diseases, While more percent children with grey and white matter neurological diseases had imaging finding indicate hydrocephalus, dilated ventricles lesions, p<0.05. Otherwise, there was no

difference in other imaging finding in both groups, p>0.05.

According to **Svenungsson et al [14]** who sought to examine the relationship between cerebral fluid and systemic and intrathecal production of proinflammatory cytokines (CSF) nitric oxide (NO) release in patients with neuropsychiatric lupus erythematosus (NPLE). They discussed the MRI results of NPLE patients. MRI abnormalities such WML, atrophy, and cerebral infarctions were present in most of the patients.

Relapses and remissions of the clinical disease with neurological impairments, along with numerous subclinical episodes as shown by MRI, are the hallmarks of MS **[15]**. Most cases evolve into chronic stages over time. Uncertainty exists on whether the relapses observed by the MRI are subclinical due to a different type of intracranial inflammation, localization in clinically silent areas, or a combination of the two **[16].**

In our study, CSF Protein and CSF Cell Count values were significantly higher (cells/µl), Compared to children with grey and white matter neurological illnesses, children with inflammatory neurological diseases had higher levels of CSF nitric oxide (mol/l), p<0.05. Other than that, neither group's CSF findings differed from the other, p>0.05.

This is in accordance with **Danilov et al [17]** who demonstrated showed when compared to non-inflammatory controls, NO products are more prevalent in the CSF in all categories of MS patients. It's interesting to note that nitrite levels were somewhat elevated throughout progressing disease and more elevated during clinical exacerbation than remission. It was clear that nitrite analyses in patients with progressing illness produced values much higher than those in patients experiencing remission.

Furthermore, **Svenungsson et al [14]** reported that, our patients had elevated NO metabolite levels in their CSF, which were found to be related to the severity of their neuropsychiatric illnesses. As a result, CSF NO metabolites may be used as a marker for NPLE and as a tool to track the effects of treatment. It is particularly intriguing since some patients with NPLE only have modest, subjective symptoms and have considerably elevated NO metabolite levels in the CSF.

The lack of elevated NO oxidative products could be attributable to the use of less sensitive techniques like the Griess reaction assay, the exclusion of patients who were not in remission, blood contamination of the samples, or the fact that measurements were only made in plasma. These chemicals' plasma levels are influenced by dietary consumption and endothelially produced NO. Changes in the CNS compartment would need to be quite large for them to be identified in plasma samples because nitrite and nitrate levels are significantly lower in CSF **[18]**.

In our study, there was no difference CSF Nitric oxide (mol/l) value regarding causes of inflammatory neurological diseases in studied children, p>0.05.

Citrulline and NOx concentrations were not correlated, and this finding may be due to the fact that all NOS isoforms synthesize both metabolites in equimolar levels regardless of the presence of CNS illness or inflammation. The literature claims that when there is an infection or inflammation, cytokines cause iNOS expression **[19]**. Therefore, NO can be generated by both enzymes during infection and inflammation since cytokines can also stimulate neuronal NOS **[20]**.

Pérez-Neri et al [21] independent of the presence of infection or inflammation, support a direct connection between citrulline and NO production. According to reports, patients with CNS infections had unchanged CSF NOx concentration **Ramírez-Bermudez et al[22]**. Patients with multiple sclerosis (CNS inflammation) do not experience this, though. Different biochemical profiles for infectious and inflammatory processes are possible **[23].**

In our study we found that, CSF Nitric oxide at cut off level ≥60.38(mol/l): show sensitivity 100%,specificity 88.5%and accuracy 94.2% to discriminate neuroinflammatory diseases in children versus grey and white matter neurological diseases.

CONCLUSIONS:

These findings underscore the promise of CSF Nitric oxide as a valuable tool for diagnosing and monitoring neuroinflammatory disorders in pediatric patients, offering a potential avenue for early intervention and improved patient care. Further studies with large number of patients should be done to support our results for better outcome.

REFERENCES:

1- **Feng G, Zhou L, Li F, Hu Y, Wang X, Tian X**. Predictors of outcome in clinically diagnosed viral encephalitis patients: a 5-year prospective study. Biomed Res Int. 2020; Article ID 2832418, 9 pages. https://doi.org/10.1155/2020/2832418

2- Venkatesan A, Geocadin RG. Diagnosis and management of acute encephalitis: A practical approach. Neurol Clin Pract**. 2014**; 4: 206–15.

3- Yan J, Kuzhiumparambil U, Bandodkar S, Dale RC, Fu S. Cerebrospinal fluid metabolomics: Detection of neuroinflammation in human central nervous system disease. Clin Trans Immunol., **2021;**10(8), e1318.

4- Ellul M, Solomon T. Acute encephalitis - diagnosis and management. Clin Med (Lond). **2018;** 18: 155–9.

5- French CD, Willoughby RE, Pan A, Wong SJ, Foley JF, Wheat LJ et al. NMR metabolomics of cerebrospinal fluid differentiates inflammatory diseases of the central nervous system. PLoSNegl Trop Dis. **2018**; 12: e0007045.

6- Vasilopoulou CG, Margarity M, Klapa MI. Metabolomic analysis in brain research: Opportunities and challenges. Front Physiol**. 2016**; 7: 183.

7- Wu J, Wuolikainen A, Trupp M, Jonsson P, Marklund SL, Andersen PM et al. NMR analysis of the CSF and plasma metabolome of rigorously matched amyotrophic lateral sclerosis, Parkinson's disease, and control subjects. Metabolomics. **2016**; 12: 101.

8- Liu X, Zhou L, Shi X, Xu G. New advances in analytical methods for mass spectrometry-based large-scale metabolomics study. Trends Analyt Chem. 2019; 121: 115665.

9- Beale DJ, Pinu FR, Kouremenos KA, Poojary MM, Narayana VK, Boughton BA et al. Review of recent developments in GC–MS approaches to metabolomics-based research. Metabolomics. **2018**; 14; 1-31.

10- Mehl F, Gallart-Ayala H, Konz I, Teav T, Oikonomidi A, Peyratout G et al. LC-HRMS data as a result of untargeted metabolomic profiling of human cerebrospinal fluid. Data Brief**. 2018**; 21: 1358–62.

11- Curcio AM, Bain JM, Beck ES, Vargas WS. Pediatric Inflammatory and Autoimmune Neurologic Disorders at a Tertiary Medical Center. J Child Neurol. 2020; 35(14):949-52.

12- Chitnis T, Ness J, Krupp L, Waubant E, Hunt T, Olsen CS et al. Clinical features of neuromyelitis Optica in children: US Network of Pediatric MS Centers report. Neurol. **2016**; 86, 245–252.

13- McKeon A, Lennon VA, Lotze T, Tenenbaum S, Ness JM, Rensel M et al. CNS aquaporin-4 autoimmunity in children. Neurol. **2008**, 71, 93–100.

14- Svenungsson E, Andersson M, Brundin L, Van Vollenhoven R, Khademi M, Tarkowski A et al. Increased levels of proinflammatory cytokines and nitric oxide metabolites in neuropsychiatric lupus erythematosus. Ann Rheum Dis. 2001 Apr 1;60(4):372-9.

15- Kraus J, Engelhardt B, Chatzimanolis N, Bauer R, Tofighi J, Kuehne BS. Cell surface bound and adhesion molecules in CSF and blood in multiple sclerosis: correlation with MRI-measures of subclinical disease severity and activity. J Neuroimmunol. 2002;122 (1 – 2), 175 –85.

16- Kaunzner UW, Gauthier SA. MRI in the assessment and monitoring of multiple sclerosis: an update on best practice. Ther Adv Neurol Disord. 2017 Jun;10(6):247-61.

17- Danilov AI, Andersson M, Bavand N, Wiklund NP, Olsson T, Brundin L. Nitric oxide metabolite determinations reveal continuous inflammation in multiple sclerosis. J Neuroimmunol. 2003 Mar;136(1-2):112-8.

19- Alderton WK, Cooper CE, Knowles RG. Nitric oxide synthases: structure, function and inhibition. Biochemical journal. 2001 Aug 1;357(3):593-615.

20- Michal G, Schomburg D, editors. Biochemical pathways: an atlas of biochemistry and molecular biology. John Wiley Sons/ 2012; Oct 30.

21- Pérez-Neri I, Ramírez-Bermúdez J, Ojeda-López C, Montes S, Soto-Hernández JL, Ríos C. Las concentraciones de glutamina y citrulina reflejan la síntesis del óxido nítrico en el sistema nervioso humano. Neurología. 2020 Mar 1;35(2):96-104

22- Ramírez-Bermudez J, Perez-Neri I, Montes S, Ramirez-Abascal M, Nente F, Abundes-Corona A, Soto-Hernandez JL, Rios C. Imbalance between nitric oxide and dopamine may underly aggression in acute neurological patients. Neurochem Res. 2010 Oct; 35:1659-65.

23- Calabrese V, Scapagnini G, Ravagna A, Bella R, Foresti R, Bates TE et al. Nitric oxide synthase is present in the cerebrospinal fluid of patients with active multiple sclerosis and is associated with increases in cerebrospinal fluid protein nitrotyrosine and S-nitrosothiols and with changes in glutathione levels. J Neurosci Res. 2002 Nov 15;70(4):580-7.

To Cite :

Ibrahim, A., Khalifa, N., Ahmed Sola, S., Abdellatif, M. Cerebrospinal Fluid Nitric Oxide as a diagnostic inflammatory Marker of Central Nervous System Inflammatory Condition. ***Zagazig University Medical Journal*****, 2024; (794-802): -. doi: 10.21608/zumj.2023.238187.2910**