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Neuroendocrine disorder in chronic fatigue syndrome

Slavica TOMIC[1], Snezana BRKIC[1], Dajana LENDAK[1,*], Daniela MARIC[1], Milica MEDIC STOJANOSKA[2], Aleksandra NOVAKOV MIKIC[3]

[1]Clinic for Infectious Diseases, Clinical Center of Vojvodina, Faculty of Medicine, University of Novi Sad, Novi Sad, Serbia
[2]Clinic of Endocrinology, Diabetes and Metabolic Disease, Clinical Center of Vojvodina, Faculty of Medicine, University of Novi Sad, Novi Sad, Serbia
[3]Faculty of Medicine, University of Novi Sad, Novi Sad, Serbia

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Background/aim: Neuroendocrine disorders are considered a possible pathogenetic mechanism in chronic fatigue syndrome (CFS). The aim of our study was to determine the function of the hypothalamic–pituitary–adrenal axis (HPA) and thyroid function in women of reproductive age suffering from CFS.

Materials and methods: The study included 40 women suffering from CFS and 40 healthy women (15–45 years old). Serum levels of cortisol (0800 and 1800 hours), ACTH, total T4, total T3, and TSH were measured in all subjects. The Fibro Fatigue Scale was used for determination of fatigue level.

Results: Cortisol serum levels were normal in both groups. The distinctively positive moderate correlation of morning and afternoon cortisol levels that was observed in healthy women was absent in the CFS group. This may indicate a disturbed physiological rhythm of cortisol secretion. Although basal serum T4, T3, and TSH levels were normal in all subjects, concentrations of T3 were significantly lower in the CFS group.

Conclusion: One-time hormone measurement is not sufficient to detect hormonal imbalance in women suffering from CFS. Absence of a correlation between afternoon and morning cortisol level could be a more representative factor for detecting HPA axis disturbance.

Key words: Chronic fatigue syndrome, hypothalamic–pituitary–adrenal axis, cortisol, thyroid hormones

1. Introduction

According to the most recent definition given by the Center for Disease Control (CDC) in 2003, chronic fatigue syndrome (CFS) is a condition of relatively rapidly developed, medically unexplainable, persistent, or relapsing fatigue that lasts for at least 6 months, does not occur after physical exertion, remains after the rest period, and always results in final reduction of life activities—work, social, and personal (1,2). The introduction of a single definition of CFS had the purpose to create clearly defined diagnostic criteria and to standardize the methodological approaches in future research. The CDC has established clear criteria for the diagnosis of CSF (3,4).

Despite numerous studies and large interest among the scientific community, the etiology of this illness is still unknown and most studies indicate multifactorial etiology (5,6). It is assumed that the underlying pathophysiological mechanism of a complex interaction between neuroendocrine, humoral, immunological, and disorders of the autonomic nervous system, with a certain psychological predisposition leads to the emergence and persistence of CFS. There is also evidence of oxidative stress (OS) occurrence, but it is not known whether it is a cause or a consequence of this syndrome (7,8).

Most of the research efforts in the last 20 years have been dealing with the research functions of the hypothalamic–pituitary–adrenal axis (HPA axis). The original idea that the CFS is associated with dysfunction of the HPA axis resulted from the similarity of its symptoms with those of glucocorticoid deficiency, as well as from research showing that the chronically fatigued person shows reduced adrenocortical activity (9,10). The available written sources show no data on a single uniform HPA axis disturbance in the CFS. There are different disorders at different levels of the HPA axis disturbances. Activation of the HPA axis during infection or stress leads to activation and release of corticotropin—corticotropin-releasing hormone (CRH)—in the central nervous system

* Correspondence: dajana.lendak@mf.uns.ac.rs

(CNS) and the peripheral tissues. The CRH system is the main component that responds to stress and the HPA axis acts as the effector organ. This release contributes to the induction of behavioral responses, autonomic reactivity, and hormonal responses via the HPA axis (11).

It is assumed that the organism switches from hypoactivity to hyperactivity of the stress system, leading to typical symptoms of fatigue, pain, and low mood through changes over self-regulatory feedback, after a period of chronic stress (11). Several mechanisms could lead to a response of decreased adreno-corticotropic hormone (ACTH) to stressors, and thus to CFS: hypersecretion of CRH without the resulting consequential regulation of target receptors, increased sensitivity to negative glucocorticoid feedback, reduced availability of free cortisol, and decreased sensitivity of target tissues to cortisol (12,13). Mediators of inflammation such as interleukin-1 (IL-1) trigger CRH neurons in the hypothalamus in terms of negative feedback, resulting in immunosuppressive effects of glucocorticoids. If hypothalamic neurons do not respond adequately to cytokine stimulation, there will be no suppression, which will result in hyperimmune state (14). Some tests that link the activity of cells of the immune system with the status of the HPA axis, thyroid, and sex hormones, point to the mechanism of chronic inflammation and confirm the possible loss of thyroid function in CFS, which is exacerbated by the inaction of the HPA axis (15).

The objectives of our research were to determine the function of the HPA axis and thyroid function in women with CFS.

2. Materials and methods

The research was conducted as a cross-sectional study at the Infectious Diseases Clinic, Clinical Center of Vojvodina in Novi Sad. The study included 40 women of generative age (15–45 years) who met the CDC criteria for the diagnosis of CFS. The youngest subjects were 24 (control group) and 25 (CFS group) years old. The mean duration of CFS was 2.29 ± 2.1 years (mean ± SD), with a minimum of 6 months and a maximum of 10 years. Criteria for noninclusion in the study were as follows: current acute or chronic illness (documentation of acute or chronic infective disease, autoimmune disease, malignancies, endocrine, neurological disease, cardiovascular and other chronic disease), psychotic disorders, bipolar affective disorders (depressive phase), dementia, anorexia, bulimia, drug and alcohol abuse, and extreme obesity (BMI ≥ 40 kg/m^2). The control group consisted of 40 healthy female volunteers (age and BMI matched). The health condition of the control group subjects was assessed by detailed anamnestic, laboratory, and clinical examination that included analysis of urine, total blood count with differential leukocyte count, sedimentation, concentration of urea, creatinine, electrolytes, bilirubin, aspartate aminotransferase (AST), alanine aminotransferase (ALT), gamma-glutamyl transpeptidase (GGT), alkaline phosphatase (ALP), lactate dehydrogenase (LDH), results of lipid status test, complement components C3 and C4, antinuclear antibodies (ANA), antimitochondrial antibodies (AMA), basal serum levels total T4, total T3, thyroid-stimulating hormone (TSH), basal serum cortisol levels at 0800 and 1800 hours, and basal plasma ACTH levels at 0800. Serological investigations included ELISA tests for IgM and IgG antibodies against adenoviruses, EBV, Coxsackie B, Parvo B19, Lyme borreliosis, HBs Ag, anti HCV, and anti HIV.

The study was approved by the Ethics Committee of the Clinical Center as well as the Ethics Committee of the Faculty of Medicine in Novi Sad. All the patients in both groups signed an informed consent for participating in the examination. In the diagnosis of CFS the diagnostic algorithm recommended by the CDC was used. As part of the endocrine tests we analyzed the following values: basal plasma ACTH and serum cortisol levels at 0800 and 1800, and basal serum levels total T4, total T3, and thyroid-stimulating hormone (TSH). T3, T4, and TSH were determined on Abbott Architect i 2000SR (CMIA methodology). ACTH concentration was determined Elecsys 2010, Roche Diagnostics machine (ECLIA methodology). Basal serum cortisol level was determined by CLIA immunometric methodology on an Advia Centaur XP machine. Quantification of fatigue, i.e. the objectification of fatigue and other symptoms related to CFS, was conducted using the Fibro Fatigue Scale (FFS). The scale is not intended to diagnose CFS. It represents a proven instrument that assesses the severity of symptoms as well as changes in symptoms that occur over time and is suitable for use in clinical testing (16,17). The FFS is a visual six-degree self-assessment scale containing 12 questions that the patient gives answers to with the help of examiners (16,17). A total sum on the FFS (summative score) represents a degree of expressed fatigue as well as the degree of all the tested difficulties within CFS.

The results were introduced and presented using standard statistical variables: mean ± standard deviation (SD), or median (minimum–maximum), according to the distribution. The examination of statistical significance of the parametric data was conducted by ANOVA or Mann–Whitney test, according to the normality of distribution. Correlations between investigated parameters were assessed by Pearson's correlation test. Significance level was set at $P < 0.05$.

3. Results

CFS group patients were 35.43 ± 6.06 (minimum 24, maximum 45) years old, while control group subjects

were 35.85 ± 5.89 (minimum 25, maximum 45) years old. There was no statistically significant difference between investigated groups in terms of age (P = 0.751). There was also no significant difference between the investigated groups in terms of BMI (CFS: mean ± SD = 21.5 ± 1.8 vs. control group (21.8 ± 2.4), P = 0.758).

Although the mean values of serum cortisol levels at 0800 in the group of patients with CFS were slightly higher than those of the control group of healthy women, the difference was not statistically significant (Table 1).

A similar relationship was observed with the levels of serum cortisol at 1800. Values were higher in CFS patients than in healthy women, but the difference was not statistically significant (Table 1).

There was no significant correlation between morning and afternoon cortisol values in patients with CFS (r = 0.082, P = 0.072) (Figure 1A). In the group of healthy women, we found a statistically significant correlation of moderate degree (r = 0.562, P < 0.001) (Figure 1B).

The basal plasma ACTH levels were slightly higher in the group of women with CFS than in the control group, but the difference was not statistically significant (Table 1).

In all subjects, basal serum values of TSH, total T3, and total T4 were within normal ranges. However, the mean value of T3 in the CFS group was significantly lower than in the group of healthy women (Table 1).

Analysis of basal serum values of T4 showed higher values in the group of patients with CFS than in the group of healthy women, but the difference was not statistically significant (Table 1). The mean values of TSH were slightly lower in patients with CFS than in healthy women, but the difference was not statistically significant (Mann–Whitney U value = 729.500, P = 0.497).

The mean values of total FFS scores were significantly higher in affected subjects (mean = 21.90, SD = 5.23 vs. mean = 2.93, SD = 1.87, P < 0.0001) than in the control group of healthy subjects.

We examined whether there was a correlation between the degree of fatigue, i.e. total score FFS and basal serum levels of T3, in women in the CFS group, since we found lower values of T3 in women with CFS. The total FFS score correlated with the mean basal values of T3 in the CFS group. There was a statistically significant negative correlation between low levels of expression (r = –0.355, P < 0.05) (Table 2; Figure 2).

The analysis of total FFS score value with T4 and TSH basal serum levels in the affected group did not show a statistically significant correlation (T4 r = 0.211, P > 0.05, TSH r = 0.032, P > 0.05) (Table 2).

4. Discussion

Endocrine dysfunction is one of the etiological factors of CFS. HPA dysfunction and hypocorticism are also considered possible factors in CFS development. Sophisticated neuroendocrine mechanisms are most certainly disturbed in CFS, which is probably triggered by infection, stress, or oxidative stress. In our study we found no statistically significant differences in mean values of morning and afternoon cortisol levels as well as ACTH among the tested groups. This was also confirmed by other researchers (18,19). However, Wyller et al. found significantly lower values of cortisol/creatinine in patients with CFS (20). This can be explained by methodological differences in cortisol determination. The literature data suggest that 20%–25% of patients with CFS and similar conditions have hypocorticism.

In women with CFS there was no correlation between the values of afternoon and morning cortisol levels, while in the group of healthy women there was a strong positive correlation. The results might be explained by physiological differences in the rhythm of cortisol secretion. The basal plasma ACTH levels were within the normal range in both groups. The first study done on this subject indicated reduced morning or even evening cortisolemia and increased basal values of the evening ACTH. Other studies that investigated this in the past included relatively small numbers of patients (21,22). On the other hand, several recent studies, for example the ones done by Altemus et al.,

Table 1. Hormone levels in CFS and control groups.

	CFS group	Control group	ANOVA F*/Mann–Whitney U** value	P
	Mean ± SD or median (min–max)	Mean ± SD or median (min–max)		
Cortisol 0800 (nmol/L)	522.82 ± 153.27	500.49 ± 227.37	0.265*	0.608
Cortisol 1800 (nmol/mL)	247.20± 139.22	216.80± 132.80	0.999*	0.321
ACTH (pg/mL)	22.37± 11.97	20.84± 12.98	0.301*	0.585
T3 (nmol/L)	1.46 ± 0.22	1.64 ± 0.33	8.077*	0.006
T4 (nmol/L)	91.00 ± 12.93	87.66 ± 19.49	0.817*	0.369
TSH (mIU/L)	1.85 (<0.01–13.30)	1.62 (0.60–19.5)	729.50**	0.497

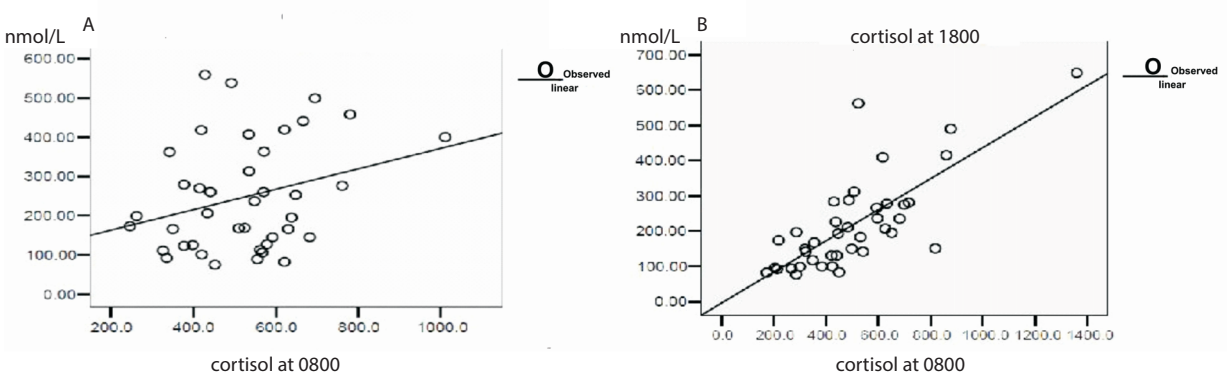

Figure 1. Correlations of 8-h and 18-h cortisol levels in the patient group (A) and control group (B).

Table 2. Correlations of FFS with T3, T4, and TSH concentrations

		T3	T4	TSH
FFS	Correlation coefficient (r)	–0.355	0.211	0.032
	p	<0.05	>0.05	>0.05

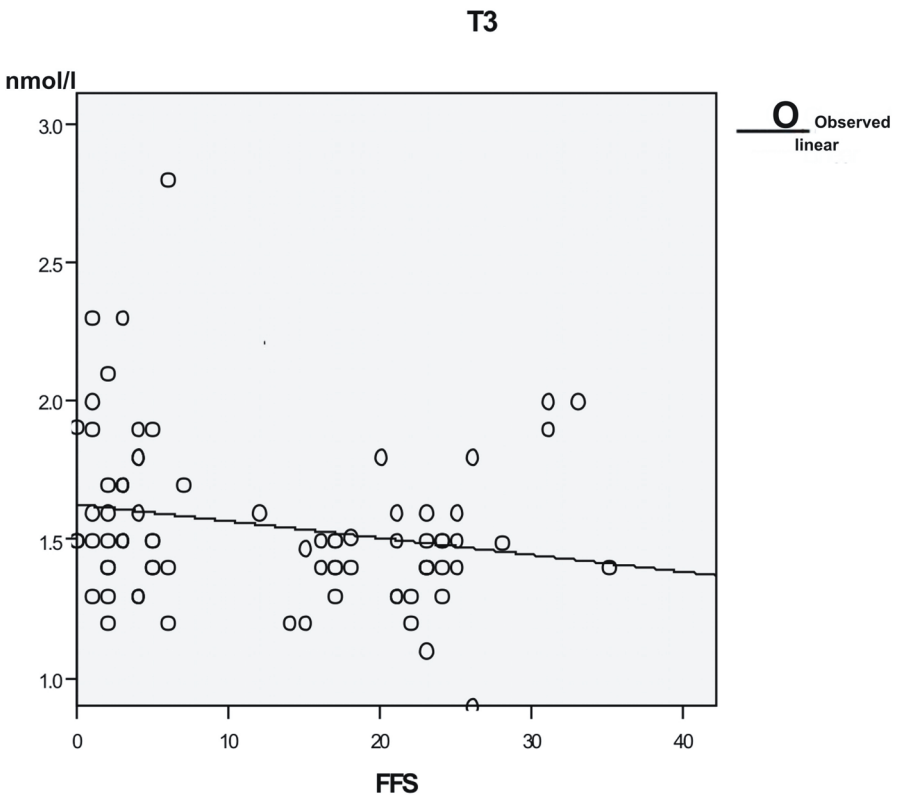

Figure 2. Correlations of FFS and total T3 concentrations.

have not found decreased levels of cortisolemia (23). Two other studies also have not found reduced serum cortisol levels in CFS patients in comparison to the control group (12,24).

There were also a number of studies that dealt with determining the values of free cortisol levels in 24-h urine and in saliva. In the study performed by Clear et al. in 121 patients and 64 healthy controls the duration of CFS was

5.4 years and the study included patients with psychiatric comorbidity. Decreased level of cortisol in the urine in the CFS group compared to the control group has been verified and there was no difference detected in cortisol levels between patients with or without psychiatric comorbidity. We highlight these results as it is known that depression often results in high levels of circulating cortisol. Clear et al. conducted research in a smaller sample of CFS patients, this time in accordance with the Oxford version of the CDC and without psychiatric comorbidity and they also detected lower levels of cortisol in 24-h urine (14).

The available research studies on disorders in the HPA axis in CFS are somewhat contradictory. Some studies measuring cortisol levels in urine and saliva confirmed that levels are lowered while others did not (21,23,24). Even the precise biological studies that measured free cortisol levels in 24-h urine or saliva did not yield different results. There are studies that show a weaker ACTH and cortisol response in the dynamic tests with human CRH in CFS, which implies a generally weakened HPA axis in CFS verified by computed tomography (CT) results for adrenal glands in CFS (6). The study by Torres-Harding et al. found a significant relationship between levels of cortisol in saliva and CFS (fatigue and pain), which also confirms that cortisol and HPA dysfunction have important roles in the development of CFS (25).

Although the HPA axis has been extensively tested, we would like to emphasize that it is still unclear whether hyporcortisolemia causes fatigue. Recent studies that investigated polymorphism in glucocorticoid receptor in CFS suggest that the altered sensitivity to cortisol may be the cause of the relative hyporcortisolemia. This is supported by the different degree of fatigue in investigated patients and this theory rules in favor of genetic predisposition. On the other hand, immune reaction releases IL-6, which increases the quantity of free circulating cortisol by stimulating the hypothalamus CRH neurons and inhibiting the corticosteroid-building globulin gene (CBG) (26). All of this links CFS with possible hereditary disorders, including cortisol transport and CBG gene mutation as well as glucocorticoid resistance.

In our research, we did not detect decreased morning and evening values of cortisolemia, but we found "slower" cortisol response. Once again, we think that it is necessary to point out that all studies concerning HPA axis disorder give very contradictory results. Some of them found flattened cortisol diurnal rhythm (10), but the fact is that a large number of studies found only weakened HPA axis function. Torres-Harding et al. measured cortisol in saliva several times a day in 108 patients with CFS. They found abnormal daily baseline cortisol pattern using clinical judgement in 47.2% patients (25). This was present more in men (62.5%) than in women (40%). Considering the results of our study and the results of other research, we suggest that the duration of CFS should be taken into account, as it eventually leads to exhaustion of the HPA axis. In our study, mean duration of fatigue was 2.23 years, and this is not sufficient to cause total exhaustion of the HPA axis. Moreover, the sample of 40 women is not large enough to make any definitive conclusions.

In all the examinees, basic serum values of TSH, T3, and T4 were within physiological limits, but when the values were compared we noted significantly lower values of T3 in patients who had CFS.

In the literature available to us, there are no data about testing of thyroid gland function in CFS, since, as defined, hyper- or hypothyroidism exclude CFS. Considering this, our results can be interesting. Fatigue is one of the main symptoms of hypothyreosis, which, narrowed down to lower values of T3, suggests that this could be important from the clinical point of view. What implies the same is the negative correlation between T3 and FFS. This means that the patients with lower values of T3 have higher scores on FFS, i.e. more difficulties typical for CFS. Although T4 makes 90% of the total amount of hormones produced by the thyroid gland and T3 only 10%, functionally speaking, both hormones are equally important, because on the periphery, apart from the rest, T4 converts to T3.

Perhaps the lower values of T3 in people suffering from CFS could be related to the changes in the level of thyroid hormones in cases of so-called nonthyroidal illness syndrome (NTIS). This syndrome is present in chronic nonthyroid diseases (cardiovascular disease, polycystic ovary syndrome, chronic renal disease, etc. in which low T3 and normal T4 levels are observed). This could imply that CFS is a chronic disease (27,28).

There is a question whether the decrease in T3 level during any acute or chronic disease (without thyroid function disorder) represents a pathological result or an adaptive response to stress, by which the metabolic activeness is lowered and the loss of energy that is beneficial for these patients (who are actually euthyroid) is prevented. The lower level of T3 in people suffering from CFS might also be referred to as the only possible adaptive response to stress. Although pituitary–thyroid dysfunction is considered a neuroendocrine disorder that is important in the development of CFS, the literature data are scarce and conflicting. Morkens et al. (29) found high levels of TSH and normal levels of T4 in patients with CFS, while results from Wyller at al. (2016) show significantly higher free T4 concentration in patients with CFS (20). Our results also show higher T4 levels in patients with CFS compared to controls, but this difference was not statistically significant. However, these results may imply pituitary–thyroid axis dysfunction.

In conclusion, we can only suggest that subtle alteration of neuroendocrine control mechanisms plays a role in

the development of CFS, with confirmed dysfunction of the HPA axis and pituitary thyroid axis. Further research should investigate the underlying neuroendocrine mechanisms in the development of CFS. Although it is still difficult to answer the question whether these hormonal imbalances are only adaptive mechanisms or pathological disorders, the fact is they correlate with the level of fatigue, which is the central clinical symptom of this disorder.

References

1. Prins JB, van der Meer JWM, Bleijenberg G. Chronic fatigue syndrome. Lancet 2006; 367: 346-355.
2. Nijhof LS, Rutten JMTM, Uiterwaal CSPM, Bleijenberg G, Kimpen LLJ, van de Putte EM. The role of hypocortisolism in chronic fatigue syndrome. Psychoneuroendocrinology 2014; 42: 199-206.
3. Fukuda K, Straus SE, Hickie I, Sharpe MC, Dobbins JG, Komaroff A. The chronic fatigue syndrome: a comprehensive approach to its definition and study. Ann Intern Med 1994; 121: 953-959.
4. Jason LA, Porter N, Herrington J, Sorenson M, Kubow S. Kindling and oxidative stress as contributors to myalgic encephalomyelitis/chronic fatigue syndrome. J Behav Neurosci Res 2009; 7: 1-17.
5. Bains W. Treating chronic fatigue states as a disease of the regulation of energy metabolism. Medical Hypotheses 2008; 71: 481-484.
6. Cleare JA. The neuroendocrinology of chronic fatigue syndrome. Endocrine Reviews 2003; 24: 236-252.
7. Brkic S, Tomic S, Maric D, Novakov-Mikic A, Turkulov V. Lipid peroxidation is elevated in female patients with chronic fatigue syndrome. Med Sci Monit 2010; 16: CR628-632.
8. Smirnova IV, Pall LM. Elevated levels of protein carbonyl in sera of chronic fatigue syndrome patients. Mol Cell Biochem 2003; 248: 93-95.
9. Cleare AJ. The HPA axis and the genesis of chronic fatigue syndrome. Endocrinol Metab 2004; 2: 55-59.
10. Papadopoulos AS, Cleare AJ. Hypothalamic–pituitary–adrenal axis dysfunction in chronic fatigue syndrome. Nature Rev Endocrinol 2012; 8: 22-32.
11. Jones FJ. An extended concept of altered self: chronic fatigue and post-infection syndromes. Psychoneuroendocrinol 2008; 33: 119-29.
12. Jerjes WK, Peters TJ, Taylor NF, Wood PJ, Wessely S, Cleare AJ. Diurnal excretion of urinary cortisol, cortisone andcortisol metabolites in chronic fatigue syndrome. J of Psych Res 2006; 60: 145-153.
13. Lorusso L, Mikhaylova SV, Capelli E, Ferrari D, Ngonga GK, Ricevuti G. Immunological aspects of chronic fatigue syndrome. Autoimm Rev 2009; 8: 287-291.
14. Cleare AJ, Blair D, Chambers S, Wessely S. Urinary free cortisol in chronic fatigue syndrome. Am J Psychiatry 2001; 158: 641-643.
15. Maes M, Mihaylova I, Kubera M, Leunis JC. An IgM-mediated immune response directed against nitro-bovine serum albumin (nitro-BSA) in chronic fatigue syndrome (CFS) and major depression: evidence that nitrosative stress another factor underpinning the comorbidity between major depression and CFS. Neuroendocrinol Lett 2008; 29: 1-5.
16. García-Campayoa J, Pascuala A, Aldaa M, Marzob J, Magallonc R, Fortes S. The Spanish version of the fibro fatigue scale: validation of a questionnaire for the observer's assessment of fibromyalgia and chronic fatigue syndrome. Gen Hosp Psych 2006; 28: 154-160.
17. Ablin JN, Odes L, Neumann L, Buskila D. The Hebrew version of the fibro fatigue scale: validation of a questionnaire for assessment of fibromyalgia and chronic fatigue syndrome. Rheumatol Int 2009; 30: 1173-1176.
18. Gabb J, Engert V, Hetz V, Schard T, Schurmeyer TH, Elhert U. Associations between neuroendocrine responses to the Insulin Tolerance test and patient characteristics in chronic fatigue syndrome. J Psychosom Res 2004; 56: 419-414.
19. ter Wolbeek M, von Doornen Lj, Coffeng LE, Kavellars A, Heijnen CJ. Cortisol and severe fatigue: a longitudinal study in adolescent girls. Psychoneuroendocrinology 2007; 32: 171-182.
20. Wyller VB, Vitelli V, Sulheim D, Fagermoen E, Winger A, Godang K, Bollerslev J. Altered neuroendocrine control and association to clinical symptoms in adolescent chronic fatigue syndrome: a cross-sectional study. J Transl Med 2016; 14: 121.
21. Christley Y, Duffy T, Everall IP, Colin RM. The neuropsychiatris and neuropsychological features of chronic fatigue syndrome: revisiting the enigma. Curr Psychiatry Rep 2013; 15: 353.
22. Cleare AJ, Miell J, Heap E, Sookdeo S, Young L, Malhi GS, O'Keane V. Hypothalamo-pituitary-adrenal axis function in chronic fatigue syndrome, and the effects of low-dose hydrocortisone therapy. J Clin Endocrinol Metab 2001; 86: 3545-3554.
23. Altemus M, Dale JK, Michelson D, Demitrack MA, Gold PW, Straus SE. Abnormalities in response to vasopressin infusion in chronic fatigue syndrome. Psychoneuroendocrinology 2001; 26: 175-188.
24. Mommersteeg PM, Heijnen CJ, Kavelaars A, van Doornen LJ. The HPA axis and immune function in burnout. Prog Brain Res 2007; 167: 281-285.
25. Torres-Harding S, Sorenson M, Jason L, Maher K, Fletcher MA, Braun M. The associations between basal salivary cortisol and illness symptomatology in chronic fatigue syndrome. J Appl Biobehav Res 2008; 13: 157-180.

26. Rajeevan MS, Smith AK, Dimulescu I, Unger ER, Vernon SD, Heim C, Reeves WC. Glucocorticoid receptor polymorphisms and haplotypes associated with chronic fatigue syndrome. Genes Brain Behav 2007; 6: 167-176.

27. Xu G, Wenjun Y, Li J. An update for the controversies and hypotheses of regulating nonthyroidal illness syndrome in chronic kidney diseases. Clin Exp Nephrol 2014; 18: 837-843.

28. Wang B, Liu S, Li L, Yao Q, Song R, Shao X, Li Q, Shi X, Zhang JA. Non-thyroidal illness syndrome in patients with cardiovascular diseases: a systematic review and meta-analysis. Int J Cardiol 2017; 226: 1-10.

29. Moorkens G, Berwaerts J, Wynats H, Abs R. Characterisation of pituitary function with emphasis on GH secretion in the fatigue chronic syndrome. Clin Endocrinol 2000; 53: 99-106.