Comparison of Prolactin serum levels between the remission and relapse phases of Multiple Sclerosis and healthy individuals

Mehdi Maghbooli[1], Zahra Hosseinian[1], Nazanin Azizi Shalbaf[2], Arezoo Jafarzadeh[2]

[1.] Department of Neurology, Zanjan University of Medical Sciences, Zanjan, Iran.

[2.] School of Medicine, Zanjan University of Medical Sciences, Zanjan, Iran.

Corresponding author: Associate professor of Neurology, Department of Neurology, Zanjan University of Medical Sciences, Zanjan, Iran.

E-mail: m.maghbooli@zums.ac.ir

Abstract

Background: The association between serum Prolactin (PRL) levels and disease activity in Multiple Sclerosis (MS) remains debated. Studies regarding the role of PRL in the immunology of MS (regardless of gender) have had conflicting results.

Objective: This study aimed to compare the serum levels of PRL between the remission and relapse phases of MS and also between MS patients and healthy individuals.

Methods: This study was conducted on 60 patients with a confirmed diagnosis of MS, 30 of which were in remission while the other 30 were in relapse, in addition to 30 sex-matched and age-matched healthy controls. Those with underlying conditions affecting serum PRL levels were excluded from the study. Serum PRL levels were measured in fasting blood samples. Duration of disease and the existence of enhancing/non-enhancing gadolinium plaques in brain MRIs were also recorded.

Results: Serum PRL levels did not significantly differ in the MS group based on disease phase (relapse or remission phases), gender, the existence of enhancing/non-enhancing plaques, disease duration and also between MS patients in relapse and remission phases with the control group (both men and women).

Conclusions: There were no significant differences in serum PRL levels between the case and control groups (both genders). Also, no significant relationship between serum PRL levels and disease duration or the existence of active MRI lesions**.**

Keywords: Multiple sclerosis; Prolactin; Remission; Relapse.

1. Introduction

Researches performed in previous decades have demonstrated a relationship between the immune system and prolactin, subsequently opening new doors in immunoendocrinology. Prolactin plays a significant part in innate and adaptive immune responses. Based on pathological findings, response to immunomodulatory therapy and the association of immune genes with disease susceptibility, MS is understood to be an immune-mediated disease, even though its exact triggers remain unclear. MS more commonly affects women, especially those of childbearing ages [1]. MS is a chronic condition clinically characterized by episodes of focal disorders affecting the optic nerves, brain, and spinal cord, which remit to a varying extent and later recur over a period of many years. The inflicted lesions are separated into four histological subgroups: inflammatory lesions made up of T cells and macrophages (pattern I), an autoantibody lesion mediated by immunoglobulins and the complement system (pattern II), lesions characterized by the apoptosis of oligodendrocytes and the absence of immunoglobulins, the complement system and remyelination (pattern III), and lesions with just oligodendrocyte dystrophy and no remyelination (pattern IV). The last two histopathological subtypes were considered to represent primary oligodendroglial cell degeneration [2]. Hyperprolactinemia may be associated with clinical relapses in MS, especially among patients with hypothalamic lesions and/or optic neuritis. Although it is currently unknown if this is a cause or consequence of relapse, and the impact of PRL on MS outcomes still remains unclear [3]. Hyperprolactinemia **(**HRPL) is seen in numerous autoimmune diseases such as multiple sclerosis, SLE, systemic sclerosis and Sjogren's syndrome. Data regarding the association between PRL levels and disease activity are inconsistent. PRL has immunomodulatory effects by interfering with B cell tolerance induction,

inhibiting cytokine production and increasing antibody secretion. The role of dopamine agonists in the treatment of autoimmune diseases is yet to be determined [4]. Some studies do not support the hypothesis that PRL plays a role in the immunopathology of MS while the others do [5, 6]. Some studies have declared that PRL has a positive association with MS in both sexes [7, 8].

This study aimed to compare the serum levels of PRL between the remission and relapse phases of MS and also between MS patients and healthy individuals.

2. Material and Methods

2.1 Trial design:

This was a cross-sectional study. Sixty Multiple Sclerosis patients were divided into two groups, 30 were in a remission phase and 30 were in an attack phase and 30 healthy people were chosen as controls. This study was approved by the ethical committee of Zanjan University of Medical Sciences and written informed consents were obtained from all patients before entering the study.

2.2 Participants:

The patients enrolled in the study had all referred to Vali-e-Asr hospital in Zanjan. In order to participate in the study, the MS patients had to be exempt from conditions leading to elevated PRL levels, which include pregnancy, lactation, recent delivery/abortion, hypothalamic disease, pituitary adenoma, primary hypothyroidism, seizures, renal failure and cirrhosis, in addition an extensive list of drugs, including phenothiazines, butyrophenones, benzamides, reserpine, methyldopa, opiates, estrogens, cimetidine and ranitidine. TCAs and SSRIs were considered to be exclusive as well. The control group consisted of healthy individuals selected to match the patients to age and gender. The controls were not

pregnant or breastfeeding at the time of study and did not take any regular medication.

2.3 Variables:

The clinical characteristics of MS patients such as disease onset, subtype, and current clinical manifestation were recorded. We assessed the participants' serum prolactin levels as the study outcome. A definite diagnosis of multiple sclerosis was achieved using revised McDonald criteria.

2.4 Data sources & measurement:

Venous blood samples were collected in the morning (8-10 am) on an empty stomach. PRL levels were determined using an immunoradiometric assay test (Kavoshyar Iran Co. Test Kit). Patient demographic information and disease history were collected via questionnaires.

2.5 Study size:

Sixty Multiple Sclerosis patients, divided into two groups, 30 in a remission phase and 30 in an attack phase, in addition to 30 healthy controls. The study size was calculated via the following equation:

$$n=\frac{[Z_1-\frac{\alpha}{2}+Z_1-\beta]^2[S_1^2+S_2^2]}{(\mu_1-\mu_2)^2}$$

μ_1=25: Mean prolactin level in remission phase of MS.

SD_1=8: Standard deviation of prolactin level in remission phase of MS.

μ_2=20: Mean prolactin level in control group.

SD_2=6: Standard deviation of prolactin level in control group.

N=30: Number of participants in each group.

2.6 Statistical analysis

In order to compare mean PRL levels between MS patients and the control group, independent sample t-test and in order to compare PRL levels between control, attack and remission groups One-Way ANOVA was utilized. Statistical significance of $P<0.05$ was assumed. The results are presented as mean ± standard errors mean (SEM). All computations were performed with Prism software for Windows, version 6.

3. Results

3.1 Participants:

A total of 90 men and women were enrolled in this case-control study. Thirty cases were studied during an attack phase of MS, thirty cases were studied during a remission phase of MS and thirty healthy individuals were studied in a control group.

3.2 Descriptive data:

We divided M.S. patients into two groups of 30 consisting of 6 men and 24 women in the relapse group and 9 men and 21 women in the remission group. The control group contained 9 men and 21 women. 23 out of 60 patients had gadolinium-enhancing lesions in their Brain MRI, of which 17 belonged to the relapse group and 6 belonged to the remission group.

3.3 Outcome data:

Prolactin level was 23.9±1.7 ng/dL in women from the control group, 22.5±2 ng/dL in women in remission phase and 19±1.4 ng/dL in women in the attack phase. Prolactin level was 18.3±3.3 ng/dL in men from the control group, 16±1.6 ng/dL in men in remission phase and 17.2±2.7 ng/dL in men in relapse phase. Prolactin level was 20.7±1.25 ng/dL in women with MS (both remission and attack) vs. 16.5±1.4 ng/dL in men with MS. Prolactin level was 23.8±1.6 ng/dL in women with a gadolinium-enhanced plaque and 20±1.5 ng/dL in women with a non-enhancing plaque, compared with 16.5±2 ng/dL in men with a gadolinium-enhanced plaque and 16.4±2 ng/dL in men with a non-enhancing plaque. Prolactin level was 22.6±2.2 ng/dL in women with a history of 1-5 years of MS, 19.1±2.1 ng/dL in women with a history of 6-10 years of MS and 21.1±2.6 ng/dL in women with a history of more than 11 years of MS. In men, these numbers were 16.6±1.8, 17.8±2.6 and 14±2 ng/dL, respectively. Prolactin level was 22.2±1.6 ng/dL in the control group (both men and women) and 19.1±1 ng/dL in MS patients (both phases and both genders). Prolactin level was 22.2±1.6 ng/dL in the control group, 20.5±1.6 ng/dL in both men and women in a remission phase and 18.7±1.2 ng/dL in both men and women in a relapse phase.

3.4 Main results

Results were assumed statistically significant if P-value<0.05. There was no significant difference in serum PRL levels in women between the case and control groups in both remission and relapse phases and between remission and relapse phases together (P=0.123). There was no significant difference in serum PRL levels of men between the case and control groups in both, and between remission and relapse phases together (P=0.8). There were no significant differences in serum PRL levels between the control group and MS patients (both remission and relapse

phases) in women and men. There were no significant differences in serum PRL levels between the case and control groups in both Gadolinium enhancing plaque, non-enhancing plaque, and between Gadolinium enhancing plaque, non-enhancing plaque together in women and men. There was no significant relationship between serum PRL levels and duration of disease in women and men. There were no significant differences in serum PRL levels between the control group and MS patients (both remission and relapse phases) in both men and women. There were no significant differences in serum PRL levels between the case and control groups in both remission and relapse phases, and between remission and relapse phases together in both men and women.

4. Discussion

This case-control study was carried out on 60 multiple sclerosis patients who were hospitalized in the neurology department or were referred to the neurology clinic of Vali-e-Asr hospital in Zanjan, Iran. The purpose of this study was to measure the serum levels of prolactin in the different phases of multiple sclerosis whilst comparing the results with a control group. There weren't any significant differences in prolactin levels between the case group (both relapse and remission phase) and the control group even regarding gender, nor within the case group based on MS duration or MRI activity in both genders.

De Giglio et al. studied the association between serum prolactin levels and brain injury extensively in 2015. They measured prolactin levels in 106 multiple sclerosis patients, there were no significant differences between the prolactin levels of patients with gadolinium absorbing lesions and those with non-absorbing lesions in brain MRIs, which was concordant with our study [6]. However, in our study,

we also evaluated patients in various subgroups such as gender and current phase of the disease.

In a case-control study in 2012, Moshirzade et al. measured in 58 patients with relapsing-remitting MS in a relapse phase and compared it to the prolactin levels of 58 people in a control group. Mean serum prolactin levels were significantly higher in the group of M.S. patients compared to the control group which was not consistent with our study [7]. It seems that the difference between the results of our study and the aforementioned one is owing to the fact that we also assessed patients in a relapse phase in addition to patients in a remission phase, which makes our study more reliable.`

Coreale et al. studied the role of prolactin in the pathogenesis of M.S. in 2014 and concluded that prolactin levels of multiple sclerosis patients are higher in both remission and relapse phases when compared to controls, which was not concordant with our study [8]. A noteworthy advantage of our study compared to theirs is that we assessed patients separately according to their disease duration.

In 2015, Belal et al. measured prolactin levels in 34 patients with multiple sclerosis and compared them with 30 controls, concluding that there were no significant differences between the two groups. There were also no correlations between prolactin levels and age, type of disease, EDSS, and duration of disease which was concordant with our study [9]. The advantage of our study compared to the aforementioned one is that our study size is larger.

In a cohort study conducted by Turkuglu et al. in 2016, they assessed the serum prolactin levels of 255 MS patients, 19 neuromyelitis optica (NMO) patients, 15 clinically isolated syndrome (CIS) patients, and 240 healthy controls. They concluded that prolactin may have a role in the immunopathogenesis of MS, NMO and the conversion of CIS to MS [10]. This is not concordant with our study. The

strength of our study is that we evaluated patients separately based on their current phase of the disease.

In a meta-analysis by Wei et al in 2017, they utilized 8 studies with 426 MS patients and 296 healthy controls and concluded that there were significantly higher prolactin levels in MS patients in comparison to healthy controls, also being influenced by region, age and disease duration [11]. This study is not consistent with our findings, the advantage of our study is that we studied patients with a similar race in a specific region and also we incorporated the duration of disease in our analysis, but a large difference in study size between our study and the aforementioned one does exist.

In 2015, Etemadifar et al. assessed twenty-two MS patients with hyperprolactinemia concomitant with a pituitary adenoma and 66 MS patients without hyperprolactinemia. They concluded that a correlation between the duration of disease and duration of hyperprolactinemia exists, but no statistical significance was found between prolactin and duration of disease onset [12]. The mentioned study is not concordant with our study regarding prolactin levels but the advantage of said study is that they have included the duration of high PRL in their analysis, unlike our study.

Limitations:

This study had some limitations including confined number of participants in each group, restrictions in exact time for taking blood samples in almost all of the subjects, determining the accurate point of the MS course, full consideration of all factors confound the prolactin measurement and the undetermined effects of MS disease modifying drugs on prolactin levels.

5. Conclusion

The findings of the current study report no differences in the serum levels of prolactin in MS patients during remission and relapse phases, and also when compared to the control group. Furthermore, no differences in serum prolactin levels were seen among patients with gadolinium-enhancing lesions in their Brain MRI and those without. No significant correlation was found between prolactin levels and the duration of MS. Our findings do not support prolactin playing a role in the immunopathology of multiple sclerosis.

We offer these recommendations for future studies on this matter:

1. Studying more samples and controlling factors affecting prolactin levels such as patients with hypothalamic lesions and those presenting with optic neuritis.
2. Performing interventional trials on animals and cell lines.
3. Performing multi-group clinical trials with many samples and the use of drugs that increase or inhibit prolactin secretion.
4. Performing a meta-analysis in order to combine the results of various studies.

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Figure1. Mean prolactin levels in Control, Attack, Remission and Case groups.