Original Article

ASSESSMENT OF MACROPROLACTINEMIA AND ITS ROLE IN DIAGNOSIS OF HYPERPROLACTINAEMIA

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ABSTRACT

Objective: This study was conducted with objective to determine the macroprolactin levels after polyethyl glycol (PEG) treatment.

Material and Methods: This was a descriptive cross-sectional study and conducted in Armed Forces Institute of Pathology (AFIP) Rawalpindi for a period of six months from Jan to June 2019. Female patients presenting to tertiary care hospital with prolactin levels more than 1000 mIU/L were included in the study. An inclusion criterion was thus defined as patients within reproductive age and level of prolactin more than 1000mIU/L. While patients with gestational amenorrhea, patients on chronic medication, hyperthyroid or hypothyroid and patients with diagnosed case of any cancer were excluded from this study. Sample was collected in yellow topped gel tube that was centrifuged and analysed before and after PEG treatment.

Results: In current study total 136 samples were included for measuring macro prolactin levels. All these samples were analyzed for prolactin before and after Polyethylene glycol (PEG). Test of normality (Kolmogorov, s and simrnov test) was run to check whether data was parametric or non-parametric and it proved data to be non-parametric. Median age was 35 years. Prolactin in untreated samples ranged from 1089mIU/L to 66000 mIU/L while after treatment with PEG (6000) ranged from 92 mIU/L -4700 MIU/L. Out of 136 patients 79 (58 %) showed macroprolactinemia while 57 (42 %) had true hyperprolactinemia.

Conclusion: This study concluded that macroprolactinemia occur in significant number of cases leading to false over diagnosis of hyperprolactinemia.

Key Words: Hyperprolactinemia, Macroprolactinemia, PEG treatment.

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INTRODUCTION

Prolactin is a hormone released by anterior pituitary gland through lactopher cells and its main function is to stimulate lactation, other than this it has more than 300 functions in the human body. However, the most common one are osmoregulation and immune regulation [1-2] Prolactin has also regulatory reproductive functions [3]. Prolactin level increases during stress as well. As the dopamine level (stress hormone) decreases prolactin level increases [4]. Another important regulatory mechanism of prolactin secretion is oestrogen. That is why there is small increase of prolactin level during reproductive cycle when there is highest level of oestrogen [5]. Studies have shown and it is important to know clinically that hyperprolactinemia not only caused by reproductive problems like polycystic ovary but its level also increases by stress and certain other factors [6]. One such physiological condition is macroprolactinemia; which is accumulation of big prolactin and is caused by binding of globulins with prolactin and it gives false positive result in patient with no clinical features [7]. Macroprolactin has reduced or no biological activity and it can be analysed in immunoassays for prolactin. Macroprolactinemia needs to be screened out to avoid misdiagnosis and hence mismanagement of patients [8]. Macroprolactin can easily be detected by doing immunoassay before and after treatment with polyethylene glycol (PEG) which help to precipitate the macroprolactin. In current study we used PEG 6000 to elucidate the macro prolactin which contributes in inappropriate investigations, misdiagnosis and unnecessary treatment. This study aims to determine the frequency of true hyperprolactinemia and macroprolactinemia in tertiary care hospital settings.

MATERIAL AND METHODS

Study Design was observational cross-sectional. This study was conducted at Armed Forces Institute of Pathology (AFIP) after getting institutional review board (IRB) approval. This study was conducted with objective to determine the macro prolactin after polyethylene glycol (PEG 6000)

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treatment. Female patients presenting to tertiary care hospital with prolactin levels more than 1000 mIU/L were included in the study. An inclusion criterion was Patients of reproductive age referred from gynecology department with prolactin level of 1000 mIU/L were included in the study. While patients with gestational amenorrhea, those on chronic medication, hyperthyroidism or hypothyroidism and patients diagnosed with any cancer were excluded.

Serum samples were taken in yellow topped gel tube and analysis was done on ADVIA Centaur by Siemens on principle of Chemiluminescence. Macroprolactin levels were determined by measuring prolactin before and after treatment of sample with PEG 6000. For screening PEG 6000 was used.

PEG Preparation was made by dissolving 250 g/L PEG in phosphate buffered saline (137 mmol/l sodium chloride and 10 mmol/l sodium phosphate). 250 microliter (µL) of sample was mixed with 250 microliter (µL) of the PEG. Mixture was incubated for 10 min at room temperature and then centrifuged at 1800 rpm for 30 min. Prolactin levels were measured in the supernatant again. Macroprolactin level was measured by difference in two values, which truly reflects the level of macroprolactin.

Recovery of prolactin was calculated by formula of prolactin levels after PEG treatment multiplied by 2(A) which was further divided by total prolactin levels before PEG treatment divided by 100(B) {A/B =Recovery}. Recovery of 50 % or less than 50% considered as no macroprolactinemia while recovery of more than 50 % was considered as macroprolactinemia [9]. Validation of calibration and results of patients was done by internal quality control of both high and normal levels.

Data was entered in SPSS version 21.0. Test of normality (Kolmogorov Simonov) was applied to check whether data was parametric or not. Quantitative variables computed as mean ± SD if data was parametric and in case of non-parametric data median and IQR computed and qualitative variables computed as frequency and percentages. Wilcoxon test was applied to test the study significance and to see the difference between prolactin levels before and after treatment with PEG.

RESULTS

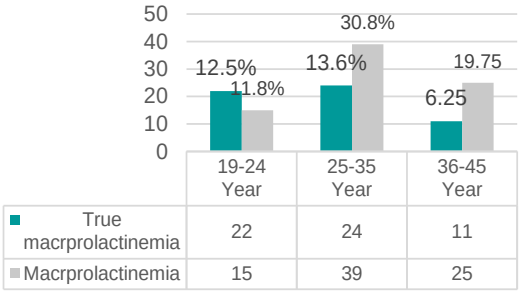
Total 136 samples were analyzed for macroprolactin. Serum samples from healthy women were analysed for prolactin before and after Polyethylene glycol (PEG). Test of normality (Kolmogorov's, simrnov test) was run to check whether data was parametric or non-parametric and data was found to be non-parametric. Median age of patient was 35 years. Patients with age of 25-35 year were 72 (53 %) while 64 (47.0 %) were in the range of 36-45 year.

Descriptive statistics for non-parametric data as median and Interquartile range (IQR) for all quantitative variables is given in (Table-1). Prolactin in untreated samples ranged from 1089mIU/L to 66000 mIU/L while after treatment with PEG (6000) ranged from 92 mIU/L -4700 MIU/L. Out of 136 patients 79 (58 %) showed macroprolactinemia while 57 (42 %) showed true hyperprolactinemia. Frequency of macroprolactinemia and hyperprolactinemia is computed in (Figure-1). Man Whitney U test applied for significance of study and it gave significant difference between prolactin level before and after treatment (P-value <0.05).

Table-1: Descriptive statistics of study parameter.

Parameter	19-25 year Median (IQR)	26-35 year Median (IQR)	36-45 year Median (IQR)
Age	24(18)	29(21)	39(32)
Prolactin Before PEG	1891(2321)	1789(2475)	1389(1987)
Prolactin after PEG	532(429)	554(538)	499(392)

Figure-1: Frequency of macroprolactinemia and true hyperprolactinemia.



DISCUSSION

True hyperprolactinemia is caused by the accumulation of excessive monomeric prolactin in serum [10]. Whereas macroprolactinemia is characterized by a large molecular mass of prolactin (macroprolactin) as the main molecular form of prolactin in sera. Macroprolactin is largely a complex of prolactin with immunoglobulin G (IgG) especially Anti PRL autoantibodies.

In current study recovery of prolactin after PEG (600) treatment was <50 % for macroprolactinemia and >50 % for Hyper prolactinemia while in a study conducted by Suliman *et al*. [11] recovery of 40 percent was considered to

differentiate between monomeric prolactin and macroprolactin but it is suggested in same study that prolactin recovery 30-65 % should be considered and results should be confirmed by gel chromatography for confirmation and final diagnosis [12]. In the current study macro prolactin levels decrease after PEG treatment which correlate with the findings of study conducted by John *et al.* in which prolactin serum samples were treated with the PEG (8000) [13]. A study conducted by Gibney *et al.* suggested that macroprolactin is an eye opener for physicians as macroprolactin may misclassify patients with hyperprolactinemia [14]. In the current study it was observed that patients with macroprolactin showed clinical features in 8 % of the patients while in 62 % patients with true hyper prolactinemia showed clinical features [15]. Study strength is that it will be helpful to physicians in better diagnosis and management of hyperprolactinemia. While current study has some limitations regarding follow up of patients.

CONCLUSION

This study concluded that macroprolactinemia occurs in significant number of cases leading to falsely over diagnosis of hyperprolactinemia.

AUTHORS CONTRIBUTION

Amna Shujaat Ali Naqvi: Literature review
Zujaja Hina Haroon: Final review and approval
Qurat ul Ain: Statistical and result analysis
Muhammad Aamir: Proof reading
Sobia Irum Kirmani: Data collection
Muhammad Tahir Khadim: Overall supervision

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