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ORIGINAL ARTICLE

Prevalence and reproductive manifestations of macroprolactinemia

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Abstract

Purpose Macroprolactinemia is characterized by predominance of macroprolactin molecules in circulation and generally has extra-pituitary origin. Macroprolactin is viewed as biologically inactive, therefore asymptomatic, and thus may not require any treatment or prolonged follow-up. In addition, data on prevalence of macroprolactinemia and its clinical manifestation are also rare. Therefore, the present study was aimed to find out prevalence of macroprolactinemia and its association, if any, with reproductive manifestations.

Material and methods Macroprolactin was measured in 102 hyperprolactinemia cases (>100 ng/ml prolactin level), 135 physiological hyperprolactinemia cases (50 pregnant and 85 lactating females; >100 ng/ml prolactin level) and 24 controls. Poly ethylene glycol (PEG) precipitation method was carried out to screen macroprolactin. Prolactin recovery of <25% was considered overt macroprolactinemia. Detailed clinical data was recorded which included complete medical history, physical examination and hormone measurements besides CT/MRI for pituitary abnormalities.

Results Prevalence of macroprolactinemia was 21.57% (22/102) in hyperprolactinemia (prolactin >100 ng/ml). There was no case of macroprolactinemia in physiological hyperprolactinemia, or healthy control females. Reproductive manifestations were present in 72.73% (16/22) macroprolactinemia cases, out of which macroprolactinemia was the sole cause of associated reproductive manifestations in 68.7% (11/16) cases. Reversal of reproductive dysfunction/s was observed in five cases with appropriate treatment for high macroprolactin.

Conclusion Macroprolactinemia prevalence was found to be 21.5%, out of which 72.73% cases had associated reproductive dysfunctions.

Keywords Hyperprolactinemia · Macroprolactin · Macroprolactinemia · Polyethylene Glycol Precipitation · Prolactin

Introduction

Macroprolactin is a complex formed randomly in circulation predominantly by the binding of monomeric prolactin (~22 kDa), secreted from pituitary, to IgG antibodies and less likely to non IgG antibodies or by the formation of monomeric prolactin aggregates [1]. Macroprolactin is also known as 'big-big/large prolactin' due to its big/large size (>150 kDa). It accounts for less than 10% of total circulating prolactin in healthy individuals [1, 2]. If macroprolactin level increases more than 60% (or >50% in some studies), the condition is called macroprolactinemia. Prevalence of macroprolactinemia in hyperprolactinemia ranges from 8–66% [3–5]. However, little information is available on prevalence of macroprolactinemia from India [6]. The cut-off value of prolactin for considering hyperprolactinemia in various studies varies from >20 ng/ml to >45.1 ng/ml [7, 8].

Macroprolactinemia is believed to be asymptomatic [9] as macroprolactin molecule remains confined to circulation due to its high molecular mass that prevents it to reach extravascular spaces in target cells, resulting in limited bioavailability, thus bioactivity in vivo [10, 11]. In addition, anti-prolactin autoantibody molecules compete with prolactin receptor (PRL-R) for binding prolactin molecule resulting in reduced bioactivity of macroprolactin [12]. Reduced bioactivity of macroprolactin observed even in

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in vitro studies using human prolactin receptor mediated assays, without any influence of autoantibodies [13, 14], suggest that some other mechanisms might also be responsible for reduced bioactivity of macroprolactin. In contrast, some studies reported reproductive manifestations, such as galactorrhoea and oligomenorrhoea, for which no other cause could be found except macroprolactinemia [15, 16]. Both similar as well as lower bioactivity and immunoreactivity of macroprolactin and monomeric prolactin have been reported [17, 18]. The objective was to study the prevalence of macroprolactinemia and reproductive manifestations in undisputed cases of hyperprolactinemia (>100 ng/ml) and macroprolactinemia (>75% macroprolactin).

Patients and methods

This study was carried out in the Department of Reproductive Biology, AIIMS, New Delhi between 2014 and 2017. Ethical clearance for the study was obtained from Institute Ethics Committee, AIIMS, New Delhi, India. Patients having prolactin level > 100 ng/ml in 2 occasions of >1 month interval were included in the study. There were 102 patients with prolactin level > 100 ng/ml who were prospectively enrolled in the study after obtaining written consent. Detailed clinical data was recorded as per predetermined proforma.

The minimum initial evaluation consisted of a complete medical history, physical examination and hormone measurements besides CT/MRI to evaluate pituitary fossa for prolactinoma and/or other pituitary adenoma. Prolactin assays were performed using highly specific Chemiluminescence Microparticle Immunoassay (CMIA) (7K76 G6-5314/R06 B7K760) using ARCHITECT PLUS i2000SR automated immunoassay system (Abbott Laboratories, USA). About 5 ml peripheral blood was collected in serum vial under aseptic conditions and allowed to coagulate at room temperature for 30 min, followed by centrifugation at $3000 \times g$ for 10 min. Serum was collected in 1.5 mL eppendorf tubes in multiple aliquots of 500 µl per vial and stored at −80 °C till further analysis.

Hyperprolactinemia cases were classified according to the cause of hyperprolactinemia into- pituitary adenoma, drug induced, secondary causes and idiopathic. Twenty-four normal healthy women and 135 physiological hyperprolactinemia cases (50 pregnant and 85 lactating women) were recruited for comparison. Healthy women of age group 20 to 35 years, having regular menstrual cycle were recruited as controls. Pregnant women were in late third trimester of pregnancy and lactating women were within first 2 weeks of delivery and lactating. Women with known medical disorder, on medication or high-risk pregnancy were excluded from the study.

Reproductive manifestations

Reproductive manifestations due to high prolactin level included in the study were anovulatory infertility, menstrual irregularity (amenorrhoea, oligomenorrhoea, hypomenorrhoea or polymenorrhoea/short luteal phase), early abortions/implantation failure and galactorrhoea in females, and erectile dysfunction, loss of libido and infertility in males. Other reproductive manifestations unrelated to high prolactin levels such as dysmenorrhoea, pelvic inflammatory disease, infertility due to PCOS, endometriosis, fibroid, tubal block, etc. were not considered for analysis. Overt hypothyroidism cases were excluded, if associated.

Poly ethylene glycol (PEG) precipitation method

Screening for macroprolactinemia was done by PEG precipitation method. The method is based on differential precipitation of proteins according to their molecular weight and solubility in defined concentrations of aqueous PEG solution. Thus, prolactin-IgG complex of macroprolactin gets precipitated and supernatant gives value of free prolactin by PEG (F-4, 22). The PEG 6000 ($H(OCH_2CH_2)_nOH$, F.W.6000, CAS No. 25322-68-3) was purchased from ChemCruz, Santa Cruz Biotechnology, Inc (10410 Finnell Street, Dallas TX, 75220; Catalogue# SC- 302016 Lot# E0515). About 250 µL serum was mixed with an equal volume of PBS (phosphate buffer saline) (8 g Sodium chloride of 137 mmol/L) and 1.42 g sodium hydrogen phosphate (Na_2HPO_4: 10 mmol/L) in 1 L milliQ water; pH adjusted to 7.4 with hydrochloric acid (HCl) in 1.5 mL eppendorf tube (marked as 'pre PEG'). In another eppendorf tube 250 µL of same serum sample was mixed with an equal volume of 250 g/L PEG 6000 (marked as 'post PEG'). Both the tubes were incubated for 10 min at room temperature. Suspension was clarified by centrifugation at $14000 \times g$ for 5 min. Supernatant of each tube was transferred to separate 2 ml storage vials and was used for repeat prolactin assay. Macroprolactinemia was considered when prolactin recovery was <25% of initial prolactin value.

Statistical analysis

Descriptive statistics were expressed as *n* (%), mean ± SD and median (interquartile range). Continuous variables were compared by Wilcoxon rank-sum (Mann–Whitney) test. Categorical variables were compared using Chi-square test. Statistics was performed using StataCorp. 2015.Stata Statistical Software: Release 14.2. College Station, TX: StataCorp LP.

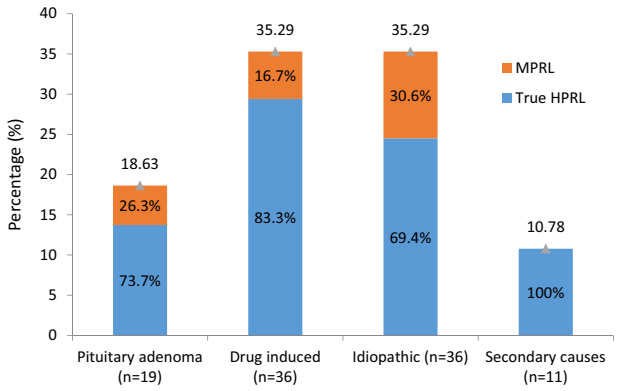

Fig. 1 Percentage of hyperprolactinemia cases belonging to pituitary adenoma, drug induced, idiopathic and secondary cause categories out of total 102 cases and percentage of macroprolactinemia and true hyperprolactinemia cases in each category (MPRL macroprolactinemia, HPRL hyperprolactinemia)

Results

Out of 102 hyperprolactinemia cases, 98 (96.08%) were females and 4 (3.92%) were males. Mean age of hyperprolactinemia cases was 30.61 ± 6.6 (range 19–48) years. Prolactin levels varied greatly among hyperprolactinemia cases, ranging from 100 to 8484 ng/ml, with median prolactin level 159.17 ng/ml (IQR: 128.43, 245.36). Most cases of hyperprolactinemia belonged to drug induced (36/102) and idiopathic (36/102) categories (35.29% each), followed by pituitary adenoma cases (19/102; 18.63%) and other secondary hyperprolactinemia cases (11/102; 10.78%) (Fig. 1).

Prevalence of macroprolactinemia

PEG precipitation results showed 22 cases (21.57%) with post PEG recovery of prolactin < 25% and were therefore, considered overt macroprolactinemia cases. No case of macroprolactinemia was observed in pregnant or lactating cases (physiological hyperprolactinemia cases) or controls. Post PEG prolactin recovery percentage ranged between 51.68 and 83.68% in pregnant women; 39.88 and 89.43% in lactating women; and 61.27 and 75.39% in controls. Thus, prevalence of macroprolactinemia was 21.57% (22/102) in our study. Prevalence of macroprolactinemia was highest in idiopathic (30.56%), followed by pituitary adenoma (26.32%), and drug-induced (16.67%) categories (Fig. 1).

Macroprolactinemia cases

All macroprolactinemia cases were females. Age of macroprolactinemia cases was between 22 and 48 years with mean age 28.68 years. Prolactin levels in macroprolactinemia cases varied from 100 to 566.3 ng/ml with median prolactin level 137.6 ng/ml (IQR: 115.43, 189.88). Post PEG prolactin recovery percentage in these macroprolactinemia cases varied from 2.4 to 22.4%. Post PEG prolactin levels were normal in 20 (90.9%) macroprolactinemia subjects, remaining two cases having post PEG prolactin 43.26 and 65.04 ng/ml (Fig. 2). Details of two macroprolactinemia patients with raised levels of monomeric prolactin were as follows:

Case no.3 was 32 years old female, having hyperprolactinemia associated with pituitary adenoma with history of psychiatric disorder, prolactin level 566.3 ng/ml (post PEG free PRL 43.26 ng/ml, % recovery of prolactin 7.89%, macroprolactin level 504.34 ng/ml) and presented with primary infertility and oligomenorrhoea, but no galactorrhoea.

Case no.9 was 23 years old female, having hyperprolactinemia associated with drugs (on antipsychotic drugs: Risperidone, Quetiapine and Pacitane), prolactin level of 363.3 ng/ml (post PEG prolactin 65.04 ng/ml, % recovery of prolactin 19.6%, macroprolactin level 266.8 ng/ml) and presented with oligomenorrhoea, but no galactorrhoea.

Pre PEG total prolactin, post PEG free prolactin and PEG precipitated macroprolactin levels of cases and controls are summarized in Table 1.

Reproductive manifestations

Reproductive manifestations were observed in 72.73% (16/22) macroprolactinemia cases. Remaining six cases had reproductive manifestations (four cases) more likely due to tubal blockage and/or PCOS or were without any reproductive manifestation (two cases). Of the 16 cases presenting high macroprolactin associated reproductive manifestations, 11 (68.75%) cases had only macroprolactinemia associated with presenting reproductive manifestation/s, while 5 (31.25%) cases had other associated causes like bilateral tube blockage in two cases, ovarian cyst (possibly endometriotic) in two cases and uterine hypoplasia with ovarian pathology (left ovarian cyst and right ovary not visualized) in one case (Table 2). Galactorrhoea was observed in eight cases (36.36%), menstrual irregularity in seven cases (31.82%; amenorrhea in 1, oligomenorrhoea in 4, and hypomenorrhoea in 2 cases). Infertility was observed in 11 macroprolactinemia cases (primary infertility in ten cases and secondary infertility in one case; Table 3). Isolated reproductive manifestations were observed in seven of these macroprolactinemia cases (primary infertility (2), hypomenorrhoea (2), recurrent abortions (1), galactorrhoea (1), and oligomenorrhoea (1)) and combinations of reproductive manifestations were observed in nine cases.

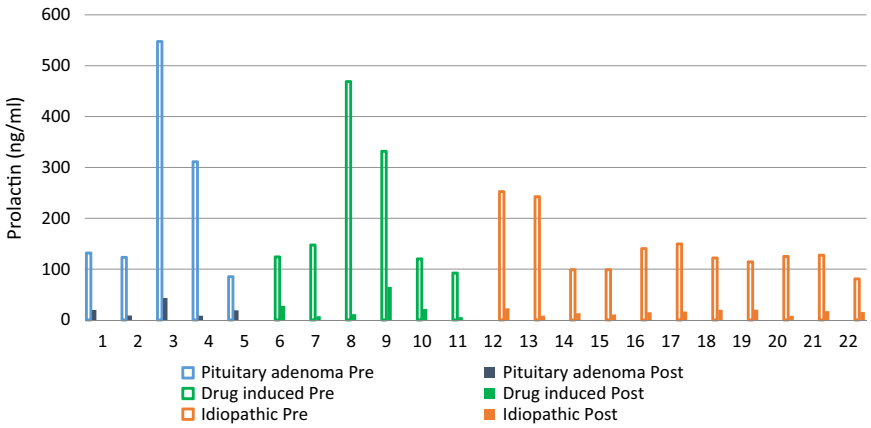

Fig. 2 Levels of Pre PEG (hollow bars) and post PEG (filled bars) prolactin in 22 macroprolactinemia cases

Follow up (telephonic and during study period) of 11 macroprolactinemia cases having no other associated cause of the presenting reproductive manifestation/s revealed resolution of symptoms (infertility and menstrual irregularity) following appropriate treatment in five cases (Table 4), further strengthening the point that macroprolactin could be the cause of presenting reproductive manifestation/s. Remaining six cases either could not be contacted or refused Cabergoline or continued antipsychotics.

Comparison between macroprolactinemia and true hyperprolactinemia

The mean age of macroprolactinemia (28.68 years) and true hyperprolactinemia cases (31.14 years) did not show any significant difference (p value 0.1210). There was no significant difference (p value 0.0543) in median prolactin level between macroprolactinemia and true hyperprolactinemia cases (137.6 ng/ml (IQR: 115.43, 189.88) and 164.02 ng/ml (IQR: 130.91, 257.58) respectively. Reproductive manifestations were observed in 72.73% macroprolactinemia cases and 90% (72/80) true hyperprolactinemia cases. Galactorrhoea was observed in 36.36% (8/22) macroprolactinemia cases and 52.55% (42/80) true hyperprolactinemia cases. Menstrual irregularities were seen in 31.82% (7/22) macroprolactinemia cases and 52.5% (42/80) true hyperprolactinemia cases. Infertility cases were more in macroprolactinemia group than true hyperprolactinemia group; 50% (11/22) and 31.25% (25/80) respectively. Although, hypomenorrhoea and primary infertility cases were significantly more in macroprolactinemia than in true hyperprolactinemia group (p value 0.006 and 0.015 respectively), there was no significant difference in total number of cases with associated reproductive manifestations between the two groups (p value 0.191). More so, there were cases without hyperprolactinemia related reproductive manifestation in both the groups; 6 (27.27%) in macroprolactinemia and 8 (10%) in true hyperprolactinemia. Thus, both macroprolactinemia and true hyperprolactinemia groups had cases with and without associated reproductive manifestations, and the differences were not significant. Table 3 shows comparison of number of cases with reproductive manifestation/s related to macroprolactinemia and true hyperprolactinemia groups.

Discussion

In this study we have evaluated 102 cases of hyperprolactinemia for prevalence of macroprolactinemia and reproductive manifestations. The study is unique because of high cut off value for hyperprolactinemia (>100 ng/ml prolactin level) and macroprolactinemia (>75% macroprolactin). The cut off prolactin level for selecting hyperprolactinemia cases varies greatly between studies and no study has taken cut-off ≥ 100 ng/ml. Most studies have taken cut-off value between 20 and 45 ng/ml with about 8–22% cases having prolactin level ≥ 100 ng/ml [7, 8, 19]. Our selection criteria of taking cases with ≥100 ng/ml prolactin took into account overt pathological hyperprolactinemia cases unlike other studies with lower cut off. Prolactin level below 100 ng/ml is often transient and may be associated with non-pathological conditions like intercourse, breast/chest wall stimulation, pain, stress, prolonged sleep, etc. Recovery prolactin value taken as cut off for considering macroprolactinemia also varies from <50% to < 30%, with majority studies taking <40% as cut-off for selecting macroprolactinemia [7, 20, 21]. We considered <25% recovery prolactin percentage as cut off for selecting macroprolactinemia cases. More so, there was no case with post PEG recovery prolactin between 25 and 40% in our study.

Table 1 is showing median (inter quartile range) of total prolactin (ng/ml), post PEG or free prolactin (ng/ml), macroprolactin (ng/ml) levels and percent recovery of prolactin after PEG precipitation in different categories of hyperprolactinemia and controls

Category of patients (*N*)	PRL (ng/ml)	Post PEG PRL/ Free PRL (ng/ml)	Macroprolactin (ng/ml)	% Recovery
Total HPRL cases (102)	159.17 (128.43, 245.36)	103.07 (59.75, 139.93)	45.39 (31.1, 102.15)	72.88 (61.87, 78.47)
MPRL cases (22)	137.6 (115.43, 189.88)	16.08 (9.26, 20.2)	113.2 (94.61, 207.22)	10.99 (6.65, 17.39)
True HPRL cases (80)	164.02 (130.91, 257.58)	110.85 (89.42, 167.28)	40.56 (26.55, 61.24)	75.11 (70.78, 80.23)
Controls (24)	10.42 (8.74, 12.92)	7.33 (6.05, 9.08)	2.95 (2.57, 4.11)	71.35 (69.41, 72.98)
Lactating females (85)	258.7 (208.4, 311.56)	194 (154.94, 249.02)	56.16 (42.14, 68.72)	78.75 (75.43, 81.12)
Pregnant females (50)	219.75 (171.57, 269.83)	165.53 (124.56, 207.43)	50.53 (43.47, 63.24)	75.97 (72.84, 78.51)

PRL prolactin, *MPRL* Macroprolactinemia, *HPRL* hyperprolactinemia

Prevalence of macroprolactinemia

Our study found 21.57% (22/102) prevalence of macroprolactinemia in cases with ≥100 ng/ml prolactin. Other studies have reported prevalence of macroprolactinemia from as low as 8% to as high as 66% [4, 5]. Studies also have shown 17–23% prevalence of macroprolactinemia in cases with serum prolactin > 32.9 ng/ml [21–24]. These results are consistent with our finding where prevalence of macroprolactinemia was 21.57% in 102 cases having serum prolactin level ≥ 100 ng/ml. However, lower or higher prevalence of macroprolactinemia has also been reported. As low as 8–10% prevalence of macroprolactinemia has been reported [5]. Higher prevalence of macroprolactinemia was reported in cases with infertility (35%) as well as in asymptomatic hyperprolactinemia (66.6%) [3, 4]. Lower prevalence of macroprolactinemia in various studies could be due to low cut-off value for hyperprolactinemia (>26 ng/ml) which will have less chances of having macroprolactin molecules [5]. Study by Gibney et al. [25] showed that the prevalence of macroprolactinemia was similar (17–21%) across all levels of total prolactin above 700 mU/l, as in our finding. Thus, the observed variation in prevalence of macroprolactinemia in some studies compared to ours may be attributed to differences in the selection criteria, prolactin level and cut-off value of post PEG prolactin.

Our study showed highest prevalence of macroprolactinemia in idiopathic group of patients (30.56%), followed by pituitary adenoma (26.32%) and drug induced (16.67%) hyperprolactinemia. Other studies have also shown highest prevalence of macroprolactinemia in idiopathic group (16%), compared to prolactinoma cases (2.7%) or drug induced cases (4.8%) [26, 27].

No case of macroprolactinemia was detected in physiological hyperprolactinemia (advanced pregnancy and lactation), or healthy controls in our study i.e., post PEG prolactin recovery value was >25%. However, some studies have found very low prevalence of macroprolactinemia in pregnancy. A prevalence of 1.4% (2/138) to 3.8% (8/209) of macroprolactinemia was reported in pregnancy [28–30]. However, the three macroprolactinemia cases in the study of Hattori et al. [30]. had history of idiopathic hyperprolactinemia before pregnancy without any clinical symptoms, and so cannot be considered macroprolactinemia in physiological hyperprolactinemia. No data has been found on prevalence of macroprolactinemia in lactating women. Studies have reported macroprolactin in healthy controls also, although, the prevalence is very low. Between 1.3 and 3.68% of general population with normal prolactin levels may have macroprolactinemia [24, 29].

Reproductive dysfunctions associated with macroprolactinemia

In this study we observed reproductive dysfunctions associated with high prolactin levels in 72.73% (16/22) macroprolactinemia cases, possibly attributed to either macroprolactin alone (68.75%) or macroprolactin in association with other factors (31.25%). Reversal of reproductive dysfunctions with appropriate treatment for high prolactin in five macroprolactinemia cases further strengthened the likelihood of macroprolactin being the cause of presenting reproductive dysfunction. Contrary to our finding, several studies observed rare presentation of macroprolactinemia with classical symptoms of hyperprolactinemia [31–34]. Absence of clinical symptoms in macroprolactinemia was said to be due to their large size making them unable to reach PRL-receptors in extravascular spaces, and competitive binding of anti-prolactin antibodies and prolactin-receptors to epitopes on prolactin molecule [35, 36]. In vitro bioassays using human prolactin receptors also show reduced bioactivity of macroprolactin, irrespective of the presence or absence of autoantibodies, suggesting yet another mechanism for reduced bioactivity of macroprolactin [13, 14]. However, there are other studies where clinical symptoms have been found associated with macroprolactinemia, although to a lesser extent [7, 19, 37]. In a study, 37% macroprolactinemia cases had symptoms associated with reproductive disorders [7]. In another study, 53.6% macroprolactinemia cases were symptomatic [37],

Table 2 is showing details of macroprolactinemia cases including the cause of hyperprolactinemia, presenting reproductive manifestation/s and probable associated cause

Cause of macroprolactinemia (*N*)	Reproductive dysfunction/clinical manifestation (*N*)	Associated cause (*N*)
Pituitary microadenoma (5)	Primary infertility for 2–13 yrs (3)	Unrelated to HPRL (1)
	Primary infertility + Galatorrhoea (1)	B/L tube blockage (1)
	Hypomenorrhoea (1)	Cyst in left ovary (1)
Idiopathic (11)	Primary infertility (2)	Unrelated to HPRL (2)
	Primary infertility + Oligomenorrhoea (1)	–
	Primary infertility + Galactorrhoea (3)	B/L tube blockage (1); Multiple cysts (1)
	Primary infertility + Galactorrhoea + Oligomenorrhoea (1)	–
	Secondary infertility (2)	Unrelated to HPRL (1)
	Galactorrhoea + Oligomenorrhoea (1)	–
	Hypomenorrhoea (1)	–
Drug induced (6)	No reproductive dysfunction (2)	–
	Primary infertility + Amenorrhoea (1)	Uterine hypoplasia, LT OV-CYST, RT OV-not visualised (1)
	Primary infertility + Galactorhoea (1)	–
	Galactorrhoea (1)	–
	Oligomenorrhoea (1)	–

Table 3 shows comparison of cases with reproductive manifestations related to macroprolactinemia ($n = 22$) and true hyperprolactinemia ($n = 80$)

Clinical symptom	MPRL ($n = 22$)	True HPRL ($n = 80$)	p Value
Reproductive manifestation/s related to high PRL level	72.73% (16)	90% (72)	0.191
Galactorrhoea	36.36% (8)	52.5% (42)	0.180
Menstrual irregularity	31.82% (7)	52.5% (42)	0.214
Oligomenorrhoea	18.18% (4)	27.5% (22)	0.374
Amenorrhoea	4.55% (1)	21.25% (17)	0.069
Polymenorrhoea	0	3.75% (3)	0.357
Hypomenorrhoea	9.09% (2)	0	**0.006***
Infertility	50% (11)	31.25% (25)	0.103
Primary infertility	45.45% (10)	20% (16)	**0.015***
Secondary infertility	4.55% (1)	11.25% (9)	0.349
Erectile dysfunction/reduced libido	0	5% (4)	0.285

MPRL macroprolactinemia, *HPRL* hyperprolactinemia, *p value <0.05 considered significant

similar to our study. In our study, galactorrhoea was observed in 36.36%, menstrual irregularity in 22.73%, and infertility was observed in 50% cases. Other studies have shown galactorrhoea in 4–33.3% and menstrual irregularities in 14–57% cases [6, 8, 38–40].

Comparison between macroprolactinemia and true hyperprolactinemia

Our study found reproductive manifestations in 72.73% macroprolactinemia cases and 90% true hyperprolactinemia cases and the difference was not significant (p value 0.191). The study by Thirunavakkarasu et al. [6] showed incidence of oligomenorrhoea and galactorrhoea more in true hyperprolactinemia (46 and 30%) than in macroprolactinemia (14 and 5%). In yet another study of 106 patients with macroprolactinemia, menstrual abnormality was seen in 37% true hyperprolactinemia and 25% macroprolactinemia patients, while galactorrhoea was associated with 34% true hyperprolactinemia and 12% macroprolactinemia cases (35). These studies correlate with our study which also shows that galactorrhoea and menstrual irregularity are less frequent in macroprolactinemia compared to true hyperprolactinemia (36.36% v/s 52.55% cases with galactorrhoea and 22.73% v/s 52.5% cases with menstrual irregularity), however, the differences are not statistically significant (Table 3). Thus, both macroprolactinemia and true hyperprolactinemia groups have comparable number of cases with associated reproductive manifestations.

Despite 72.73% macroprolactinemia cases in our study having symptoms associated with reproductive manifestations (menstrual irregularity, galactorrhoea, infertility, etc.) free/true prolactin levels were normal (<28 ng/ml) in 90.9% cases. Hence, we propose that macroprolactin itself may also be biologically active variant responsible for clinical symptoms; however, it needs further verification. Reduction

Table 4 showing details of five macroprolactinemia cases showing resolution of reproductive manifestation/s after treatment for high prolactin

Case No.	PRL ng/ml	Post PEG recovery PRL/ Pre PEG Total PRL ng/ ml (%)	Cause of HPRL	Reproductive Manifestation/s	Treatment	Outcome	Remarks
2	114.46	8.76/123.16 (7.11)	Pituitary adenoma	Primary infertility	Cabergoline	Prolactin level post treatment - 0.73 ng/ml. Conceived, delivered, lactating normally	Macro-prolactinemia seems responsible for reproductive manifestation/s
3	566.3	43.26/547.60 (7.89)	Pituitary adenoma	Primary Infertility, Oligomenorrhoea	Cabergoline	Prolactin level post treatment - 1.14 ng/ml. Regular menstrual cycle. Infertility continued due to additional male factor	
9	363.3	65.04/331.84 (19.6)	Antipsychotic drugs	Oligomenorrhoea	Stopped all medication after marriage	Prolactin level post treatment – NA. Conceived, delivered, lactating normally	
15	101.88	10.76/98.92 (10.87)	Idiopathic	Primary Infertility, Oligomenorrhoea	Cabergoline	Prolactin level post treatment - 0.48 ng/ml. Conceived, delivered, lactating normally	
21	123.2	17.38/127.44 (13.63)	Idiopathic	Oligomenorrhoea, Galactorrhoea	Treatment from private hospital (exact composition not known)	Prolactin level post treatment – NA. Regular menstrual cycle, conceived, delivered, lactating normally	

PRL prolactin, *PEG* poly ethylene glycol, *HPRL* hyperprolactinemia, *NA* not available

in levels of macroprolactin in cases treated with dopamine agonist along with recovery of clinical symptoms also suggest preserved bioactivity of macroprolactin molecule [41, 42].

Conclusion

The prevalence of macroprolactinemia was 21.57% in pathological hyperprolactinemia cases and highest prevalence was observed in idiopathic hyperprolactinemia (35.29%). Macroprolactinemia was seen in all age groups and was associated with reproductive manifestations in 72.73% cases. The reproductive manifestations resolved in patients appropriately treated for high prolactin. We conclude that macroprolactin was possibly responsible for clinical symptoms of hyperprolactinemia in 72.73% cases. Therefore, probably there is no clinical benefit in distinguishing macroprolactinemia from true hyperprolactinemia.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All procedures performed in the study involving human participants were in accordance with the ethical standards of the institutional ethical committee.

Informed consent Informed consent was obtained from all individual participants included in the study.

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