

**AMERICAN ASSOCIATION OF CLINICAL ENDOCRINOLOGISTS,
AMERICAN COLLEGE OF ENDOCRINOLOGY
DISEASE STATE CLINICAL REVIEW:
CLINICAL RELEVANCE OF MACROPROLACTIN IN
THE ABSENCE OR PRESENCE OF TRUE HYPERPROLACTINEMIA**

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ABSTRACT

Objective: To review the current literature regarding the prevalence of macroprolactin (macroPRL) in hyperprolactinemic patients and determine recommendations for testing.

Methods: An electronic United States National Library of Medicine PubMed search (through October, 2014) was conducted for search term “macroprolactin.” Only English-language articles were considered.

Results: MacroPRL is an under-recognized cause of elevated prolactin (PRL) and is present in approximately 4% to 40% of hyperprolactinemic patients depending on the referral population. Clinical findings which could be due to hyperprolactinemia are the impetus for testing for PRL. Because of this there is significant overlap in the clinical presentation of patients with true hyperprolactinemia and those with macroPRL, differentiation cannot always be made on the basis of symptoms. A lack of recognition of the presence of macroPRL can lead to unnecessary laboratory investigations, imaging, and pharmacologic or surgical treatment.

Conclusion: Until there is a commercially available PRL assay that is not subject to interference by macroPRL, clinicians should consider the possibility of macroPRL, especially if the clinical presentation, imaging findings, and/or response to therapy reveal inconsistencies. (**Endocr Pract. 2015;21:1427-1435**)

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This review article is a working document that reflects the state of the field at the time of publication. Because rapid changes in this area are expected, periodic revisions are inevitable. We encourage medical professionals to use this information in conjunction with, and not a replacement for, their best clinical judgment. The presented recommendations may not be appropriate in all situations. Any decision by practitioners to apply these guidelines must be made in light of local resources and individual patient circumstances.

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Abbreviations:

DA = dopamine agonist; **GFC** = gel filtration chromatography, **IgG** = immunoglobulin G; **macroPRL** = macroprolactin; **MMP3** = matrix metalloproteinase-3; **NS** = nonsignificant; **PEG** = polyethylene glycol; **PRL** = prolactin; **RA** = rheumatoid arthritis; **SLE** = systemic lupus erythematosus.

INTRODUCTION

The human prolactin (PRL) gene encodes a 227-amino acid (a.a.) polypeptide, which following translational cleavage of its 28-a.a. signal peptide, yields the main PRL 23-kDa monomer (1,2). A proportion may be glycosylated (25 kDa) which can facilitate aggregation of monomers to form “big prolactin” (10-20% of circulating forms), which is 50 to 60 kDa and likely is clinically silent (1-3). Additional larger molecular weight forms have also been reported due to glycosylation, aggregation, covalent,

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and noncovalent bonding. Nevertheless, these represent a small proportion of circulating immunoreactive PRL (4). “Big big prolactin” was first described by Soong et al (5) as a cause of moderate hyperprolactinemia, and Jackson et al (6) first used the term “macroprolactinemia” to describe a predominance of high molecular weight forms of PRL in a patient without a pituitary adenoma. Additional reports followed describing patients with idiopathic hyperprolactinemia in the absence of symptoms or a pituitary lesion (7,8). Lack of recognition of the presence of macroprolactin (macroPRL) can lead to costly laboratory and imaging investigations, as well as inappropriate pharmacologic interventions in patients with hyperprolactinemia. It can be challenging for clinicians to decide which patients should be tested for macroPRL.

Our goal is to discuss the most current clinical data on macroPRL and the evidence for and against the clinical relevance of testing. A U.S. National Library of Medicine PubMed search through October 2014 was conducted with the search term “macroprolactin.” All English language articles were read and vetted for relevance to this document, especially with regard to quality of experimental design and data analysis.

What is the Composition of MacroPRL and How Common is it?

MacroPRL is a large protein complex of 150 kDa or more as established by size-exclusion gel filtration chromatography (GFC). The complex is comprised of an immunoglobulin bound to monomeric PRL as confirmed by affinity chromatography using antihuman immunoglobulin agarose or protein A sepharose (7-9). Hattori et al (10) confirmed that immunoglobulin G (IgG) was the predominant immunoglobulin subclass and that the majority of the antibodies are PRL-specific rather than nonspecific heterophilic antibodies, as only human PRL could displace bound PRL from IgG compared with PRL from other species (4,11,12). MacroPRL has a slower clearance rate consistent with that of IgG, leading to accumulation in the circulation and thus, elevated immunoreactive PRL levels (13). In a longitudinal study, the presence of anti-PRL antibodies and the macroPRL:PRL ratio were stable for up to 17 years (12).

In most of the literature, macroPRL is considered to be significant if <40% of immunoreactive PRL is monomeric (14). The <40% cutoff appears to be 100% sensitive for confirmation of macroPRL, while only 6% of samples with 40 to 60% monomeric PRL truly contain significant amounts of macroPRL by GFC (15). Using these cut-offs, macroPRL has been reported to be present in approximately <1 to 4% of sera in patient populations not selected for elevated PRL or associated symptoms (4,16,17). From 10,737 consecutive samples, Bjoro et al (17) noted that approximately 1.5% had macroPRL. Hattori et al (4) tested sera for hepatitis B from 1,330 presumably healthy

hospital workers and found that 3.68% had macroPRL with normal monomeric PRL levels. In hyperprolactinemic patients, the reported prevalence of macroPRL ranges from 4 to 46% depending on the assay and referral population (11-13,15,18).

Does MacroPRL have Biological Activity?

Although the lack of symptoms in some patients with elevated macroPRL endorse that it is not biologically active in vivo, early cell culture experiments demonstrated bioactivity using the lymphoma Nb2 cell line, which expresses the rat PRL receptor and has a proliferative response to PRL (7,19). One interpretation put forward for this dichotomy is that, in vivo, the large macroPRL complex has poor bioavailability; it is confined to the intravascular compartment and is unable to pass through capillary walls to interact with target tissues, but it can still functionally interact with the cell-surface receptor in culture (7,19). However, it was later demonstrated that the long incubation time and dilution of serum for the Nb2 bioassay may have allowed for dissociation of PRL from the IgG, and the receptor activity was likely secondary to the released monomeric PRL (20). An additional flaw of the Nb2 proliferation assay may be that the rat PRL receptor appears to be more permissive for activation by macroPRL compared with the human receptor (21). Later experiments that employed a breast cancer cell line (T47D) expressing the endogenous human PRL receptor demonstrated that intact macroPRL lacked bioactivity as measured by phosphorylation of downstream signal transducer and activator of transcription (STAT) 5, while dissociated PRL was fully active (22). Therefore, the bound IgG molecule appears to block PRL action at the human receptor. Activation of the cell-surface receptor is initiated by binding of PRL, with subsequent interaction of the bound PRL molecule with a second receptor to form a homodimer with the bound ligand (1). Anti-PRL antibodies from patient sera have been shown to recognize epitopes located at both the carboxy- and amino-termini of PRL, which could interfere with these PRL receptor binding sites and/or receptor dimerization (22). It is thus likely that macroPRL leads to both decreased bioavailability and diminished bioactivity.

Why do Anti-PRL Antibodies Form?

The mechanisms that promote anti-PRL antibody production and macroPRL formation require clarification. We have not found evidence among available cohort studies, which would support that elevation of circulating PRL, such as from drug-induced hyperprolactinemia or a prolactinoma, promotes the formation of antibodies and macroPRL.

One hypothesis is that inappropriately phosphorylated PRL results in antigen stimulation. In the pituitary, PRL is phosphorylated at serines 163 and 195, while circulating PRL is only phosphorylated at serine 195. Some patients

who have macroPRL also have a higher predominance of circulating serine 163 phosphorylated PRL, which is more acidic and may not be recognized as a self-antigen (10). This phosphorylated PRL can cause anti-PRL antibody development in a rat model (23). The mechanism by which inappropriately phosphorylated PRL escapes from the gland is not clear, although it has been speculated to involve pituitary inflammation, such as with a mild hypophysitis, leading to unregulated pituitary release of phosphorylated PRL.

The same research group has examined a role for matrix metalloproteinase-3 (MMP3) in rheumatoid arthritis (RA) patients, although the applicability to the general population is unclear (24). They noted that the presence of macroPRL in RA patients showed an association with elevated serum MMP3, a marker of disease activity. MacroPRL was present in nearly 10% of RA patients with elevated MMP-3 but in only 2.6% of those with normal MMP-3 levels (24). *In vitro*, PRL can be cleaved by MMP-3 into smaller molecular weight vasoinhibins, which inhibit endothelial cell proliferation and angiogenesis, and the investigators speculated that PRL digestion by MMP-3 may expose new epitopes to induce antibody formation (24).

Are Specific Patient Populations More Prone to Forming MacroPRL?

Because anti-PRL antibodies can be responsible for macroPRL, the association with autoimmune disease has been studied. Case reports have described the presence of macroPRL in association with autoimmune thyroid disease (6,25). However, a significant association has not been borne out in larger patient cohorts. In regularly menstruating, euthyroid females with a diagnosis of thyroid disease who were positive for 1 or more antithyroid antibodies, 4 of 96 had macroPRL, similar to females with nontoxic goiter and negative antithyroid antibodies (5 of 86) (26). In a macroprolactinemic cohort, 14% of patients were positive for antithyropoxidase antibodies compared with 15% of patients with true hyperprolactinemia (25). Kavanaugh-Wright et al (27) also found no increased prevalence of macroPRL in the presence of antithyropoxidase or antithyroglobulin antibodies.

The association with other autoimmune diseases has also been examined. As discussed above, macroPRL was studied in a RA population (24). However, the prevalence of macroPRL in RA patients was 6.3%, similar to age- and sex-matched controls (6%) but higher than young controls (2.3%), suggesting that macroPRL prevalence increases with aging (24) as previously reported (4). There is a high prevalence of elevated PRL in patients with systemic lupus erythematosus (SLE) and correlates with disease activity (28). MacroPRL has also been described in hyperprolactinemic SLE patients, but the prevalence has not been directly compared to a hyperprolactinemic population

without SLE and the prevalence (16.6% in one study) (29) is similar to other hyperprolactinemic cohorts. Also, there is no apparent association of macroPRL with the presence of antinuclear antibodies or the levels of 12 inflammatory cytokines including interferon- γ , tumor necrosis factor- α , and interleukin-6 or CD5+ B cells. Thus, the investigators concluded that there is no increased association of macroPRL with an autoimmune milieu (27). In longitudinal 10-year follow-up, patients with macroPRL also did not manifest autoimmune disease (30). The prevalence of macroPRL among male hyperprolactinemic patients is similar or lower than in females (4,31).

Beyond autoimmune conditions, other disease populations have been examined. The kidneys clear 25% of circulating PRL, and 20 to 75% of patients with chronic renal insufficiency have hyperprolactinemia, but this has been shown to be due to elevated monomeric PRL not macroPRL (32). One study described an association of macroPRL with diabetes mellitus: of 174 patients with hyperprolactinemia, 27 had diabetes and a large proportion (18/27, 66.6%) had macroPRL, correlating with higher hemoglobin A1C compared with nondiabetic patients with hyperprolactinemia (39.5%) (33). Additionally, the prevalence of macroPRL is not increased with progression of diabetic nephropathy through micro- and macroalbuminuria (34).

How Do We Assay for macroPRL?

Unfortunately, there is no commercially available immunologically based PRL assay free from interference by macroPRL, although there is wide variation in the degree of reactivity (35-37). Smith et al (36) tested 10 macroprolactinemic patient samples and reported that immunoreactive PRL levels varied from over twofold to nearly eightfold among 9 different commercial assays when compared to the true monomeric PRL level by GFC. All assays currently in use are 2-site immunometric assays that rely on binding PRL by capture and detection antibodies that can be variably affected by the masking of PRL epitopes and steric hindrance caused by anti-PRL antibodies (Fig. 1 A).

GFC is the "gold standard" for separating and quantifying PRL and its complexes in serum, but it is technically demanding and time-consuming and is not widely available for clinical use. However, GFC has been used extensively in the research setting for comparison with more convenient methods to remove the larger macroPRL complex from sera. Immunoglobulin affinity-based methods and ultrafiltration can overestimate the amount of monomeric PRL in serum samples (38). Olukoga and Kane (14) and Leslie et al (39) first validated the use of polyethylene glycol (PEG) precipitation (e.g., treatment of equal parts of serum with PEG followed by centrifugation), to remove both macroPRL and big PRL (Fig. 1 B) (40). Notably, this procedure also results in some loss of monomeric PRL (~25%) in the precipitant (38). Because

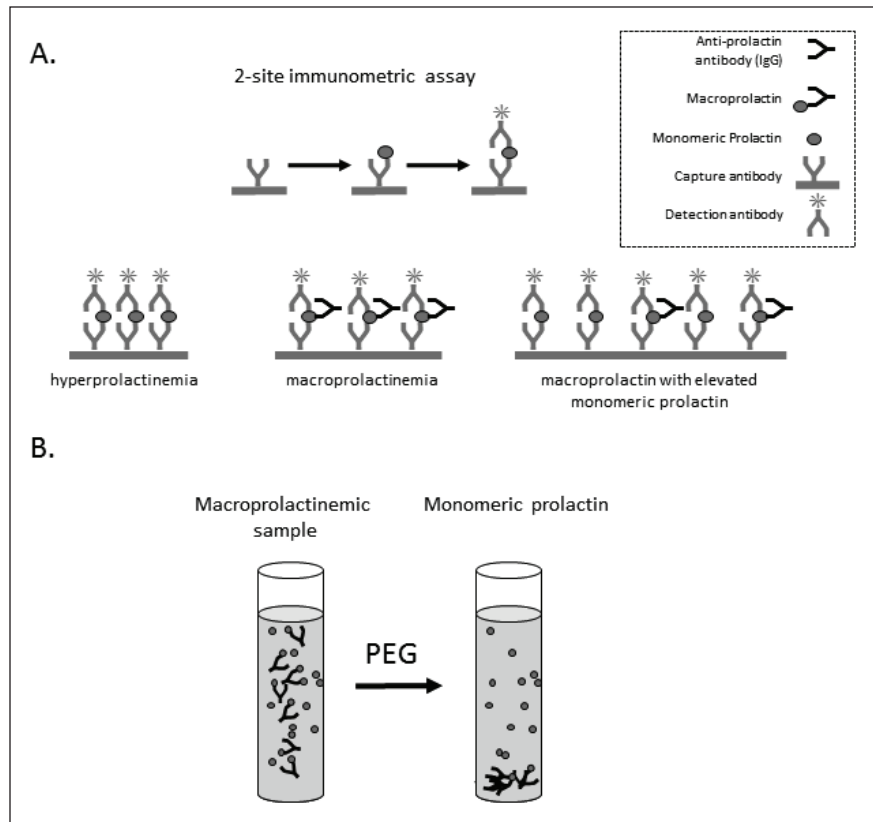


Fig. 1. A, Prolactin complexed as macroprolactin is still able to interact with the capture antibody of immunometric assays and is detected by the detection antibody. There is wide variability among assays for detection of complexed prolactin dependent on steric hindrance of and/or obscuration of the target epitope for the detection antibody. B, Illustration of PEG precipitation of macroprolactin forms. *PEG* = polyethylene glycol.

losses of monomeric PRL standard outside of serum are minimal with PEG, this suggests that the precipitation of monomeric PRL in patient samples is due to the “matrix effect” of serum. It is important to take this into consideration when determining the normal range for PRL in patients with macroPRL, as the manufacturer’s usual reference range for PRL will not account for such losses (41). To our knowledge, many labs do report a post-PEG reference range for PRL.

Although PEG precipitation usually does not cause an overestimation of monomeric PRL levels (38), it was reported by one group (42) and was likely attributable to the particular PRL assay used. The precipitation procedure is not automated, lending itself to variability (14). There are also rare examples where PEG has led to false-negative or false-positive tests. For example, elevated gamma globulin levels can cause increased precipitation of monomeric PRL and thus a false-positive result for macroPRL (2). Also, IgA containing macroPRL (rather than IgG) may not be precipitated as efficiently, leading to a false-negative result (43). Despite these shortcomings, PEG precipitation is currently the most cost-effective and practical method

for removing macroPRL and quantifying monomeric PRL after PEG precipitation and correlates best with GFC data (38). For this reason, it has been widely adopted by many reference clinical laboratories.

Can MacroPRL Coexist with True Hyperprolactinemia?

There can be coexistence of macroPRL with elevated monomeric PRL post-PEG precipitation, and these patients may have a pathologic reason (e.g., pituitary lesion) for elevated PRL. In females with menstrual disturbance and documented macroPRL, 59% had concomitant elevation of monomeric PRL after precipitation (44). Importantly, 36% of such patients had a pituitary abnormality on magnetic resonance imaging (MRI) compared with only 4% in the normoprolactinemic group after PEG precipitation (44). Therefore, to meet the criteria of benign macroprolactinemia, the clinician must take into account more than 60% precipitation of immunoreactive PRL and normalization of the remaining monomeric PRL to the reference range.

Can We Predict Which Patients are More Likely to Have MacroPRL on a Clinical Basis?

The justification for testing macroPRL is more clear if a hyperprolactinemic patient is asymptomatic and the results will affect management. However, difficulty arises when the initial PRL measurement is spurred by a presentation that could be consistent with hyperprolactinemia, such as galactorrhea, oligo- or amenorrhea, and infertility in females and decreased testosterone, decreased libido, and sexual dysfunction in males. Certainly, a proportion of these patients will have macroPRL. With nonspecific clinical features than can be ascribed to other causes, it can be challenging to predict which patients may have macroPRL based on their symptoms. This is borne out by studies of

macroPRL cohorts where there can be significant overlap in the prevalence of hyperprolactinemic symptoms and macroPRL (Table 1). However, one caveat is that many studies did not confirm that patients with macroPRL also had normal monomeric PRL.

In 1992, Leite et al (19) first noted the high prevalence of galactorrhea and/or menstrual irregularity in patients (7 of 11) with macroPRL. On a larger scale, 2 key publications from investigators in Ireland, where universal macroPRL testing is performed, compared the frequency of hyperprolactinemic symptoms or signs in patients with macroPRL (31,45). Oligo- or amenorrhea and galactorrhea were the most frequently reported symptoms for both PRL and macroPRL groups. Infertility and headache were

Table 1
Clinical Features of Patients with Macroprolactinemia Compared to True Hyperprolactinemia

	N, Hyperprolactinemia	% with Macroprolactin	Normalized monomeric prolactin noted?	Menstrual disturbance	Infertility	Galactorrhea	Headache	Other
				% Prevalence for macroprolactinemia (vs. true hyperprolactinemia)				
Leslie (39), 2001 (Northern Ireland)	1,225	26 ^e	NR	24 ^f	13 ^f	2 ^f	11 ^f	Microadenoma in 7.2% ^f
Hauache (18), 2002 (Brazil)	113	46	NR	36 ^f	7 ^f	25 ^f	11 ^f	Asymptomatic 46% (vs. 10%) Pituitary adenoma 11% (vs. 75%)
Strachan (42), 2003 (Scotland)	227	52 ^e	NR	20 ^f	11 ^f	14 ^f	NR	Pituitary adenoma in 17% ^f
Vallette-Kasic (53), 2002 (France)	1,106	10	NR	39 (vs. 49) ^a	32 (vs. 30)	46 (vs. 66) ^a	NR	Pituitary lesion 22% (vs. 31%)
Suliman (45), 2003 (Ireland) ^d	110	21	Y	57 (vs. 89) ^a	28 (vs. 8)	29 (vs. 63) ^a	10 (vs. 8)	Higher estradiol ^a , LH ^a Pituitary lesion 15% (vs. 34%)
Gibney (31), 2005 (Ireland) ^d	2,089	22	Y	59 (vs. 73) ^a	22 (vs. 7)	22 (vs. 54) ^a	5 (vs. 7)	Higher estradiol ^a , LH ^a Microadenoma 18% (vs. 41%)
Isik (47), 2012 (Turkey)	337	26	NR	12 (vs. 29) ^b 36 (vs. 54) ^c	5 (vs. 11)	39 (vs. 57) ^a	10 (vs. 20)	Asymptomatic females ^a 32% (vs. 11%) Higher LH ^a Abnormal imaging ^a 66% (vs. 81%) Erectile dysfunction 50% (vs. 72%) Male infertility 0% (vs. 16%)

Abbreviations: LH = luteinizing hormone; NR = not reported.

^a Statistically different between macroPRL vs true hyperprolactinemia, $P < 0.05$

^b Amenorrhea only

^c Oligomenorrhea/irregular menses

^d Possible overlapping study populations

^e Analyzed symptoms of a subset of macroPRL patients with available clinical records

^f No comparison to a true hyperprolactinemia cohort

prominent complaints in both groups. From hormonal analysis, follicle-stimulating hormone (FSH) levels were similar between the 2 groups, but luteinizing hormone (LH) and estradiol were higher in the macroPRL group, consistent with diminished bioactivity of macroPRL. Also, 87% of macroPRL patients who were treated with dopamine agonists (DAs) displayed a mean PRL reduction into the normal range. Nevertheless, only 1 of 8 macroPRL patients with menstrual irregularity responded to DAs with restoration of menstrual frequency compared to 19 of 23 in the true hyperprolactinemic cohort. As some patients with macroPRL report improvement of galactorrhea, the authors reason that normoprolactinemic galactorrhea may also respond to DAs (45).

Even with abnormal pituitary imaging, measurement of macroPRL may be considered if the patient is hyperprolactinemic. Although the prevalence of macroPRL has not been reported in cohorts of consecutive patients referred for assessment due to an incidental pituitary lesion, the prevalence of macroPRL in this population can likely be extrapolated from our current knowledge of the prevalence of pituitary adenomas and macroPRL in hyperprolactinemic patients. In the cohort from Ireland, 15% had an abnormality noted (microadenomas) compared to 34% (micro- and macroadenomas) of hyperprolactinemic subjects (45). Strachan et al (42) reported abnormal imaging in 19% of patients with macroPRL. For both of these studies, the microadenoma prevalence in the macroPRL patients could be considered congruent with what has been reported in the general population (46). However, other investigators have reported a much higher prevalence. Isik et al (47) examined a cohort of 337 consecutive hyperprolactinemia patients and noted that macroPRL was present in 26.1%. Notably, 65.9% of macroPRL patients still had abnormal MRI findings (54.5% had microadenomas, 6.8% macroadenomas, and 4.5% empty sella), although this was lower than the frequency of abnormal imaging in the PRL cohort (81.1%, $P = .02$). They do not indicate if all patients with macroPRL had normal monomeric PRL but the mean value for the cohort (including males and females) post-PEG precipitation was 17.0 ± 15.6 ng/mL.

Is there a level of PRL above which we can confidently rule out the presence of macroPRL? McCudden et al (48) noted that there were no macroprolactinemic patients in their study with a pre-PEG PRL >85 ng/mL, and they suggested that there is a low likelihood of macroPRL above this threshold. In most reports, the levels of PRL pre-PEG in macroPRL patients are moderate, in the 50 to 100 ng/mL range, with some approaching 150 ng/mL (18,31,42,49). In the study by Beda-Maluga et al (41), the majority of macroprolactinemic patients with total PRL levels >100 ng/mL also had hyperprolactinemia post-PEG recovery and would have been investigated and treated as such. However, in most cohorts of hyperprolactinemic patients, the median and mean immunoreactive PRL values are not significantly

different when comparing those with true hyperprolactinemia and macroprolactinemia (18,31,49). It is also important to remember that in one of the earliest reports of macroPRL by Jackson et al (6), the immunoreactive PRL levels were 350 to 400 ng/mL, with only 2% monomeric PRL as determined by GFC.

Overall, from the available published data, it can be challenging to determine who should be tested for macroPRL. Certainly, the lack of symptoms and/or imaging findings should alert the clinician to the possibility, which should prompt testing for macroPRL. However, as indicated earlier, quantifying the remaining monomeric PRL fraction should also be considered to be confident that the 2 conditions do not coexist.

Does Universal Testing for MacroPRL Reduce Overall Cost?

Unfortunately, there is a paucity of published data on the effects of health care utilization with universal testing for macroPRL in hyperprolactinemic patients. In the study from Ireland (31), there were increased laboratory costs associated with universal macroPRL screening (+27%), but this was offset by decreased costs of imaging (by -15%) and DA treatment (by -17%). De Soarez et al (50) from Brazil also examined the direct medical costs of testing, imaging, and treatment when macroprolactinemia is diagnosed. From a database of 1,793 patients with elevated PRL tested over a 3-year period, 36.5% were identified as having macroPRL. There were similar frequencies of imaging (6.5% PRL versus 5.6% macroPRL) and prescription of DAs (4.0% vs. 2.6%), although these values seem unexpectedly low for the hyperprolactinemia cohort. The incremental cost was 25% higher for the hyperprolactinemia group, but overall the authors were surprised by the number and costs of investigations and the frequency of treatment in the macroPRL group. The factors driving these interventions were unclear. Thus, even with early evidence of macroPRL, clinical practice did not differ and health care utilization and costs were not decreased by the expected margin.

Where Does This Leave Us?

Most current guidelines advocate for a case-finding approach, rather than universal testing for macroPRL. Asymptomatic, eugonadal patients without a macroadenoma are often observed without DA therapy, and demonstration of macroprolactinemia may not influence clinical management. However, the Endocrine Society (51) suggests that testing should be undertaken in all asymptomatic patients. The Pituitary Society (52) recommends assaying for macroPRL in a hyperprolactinemic patient who has moderately elevated levels (25 to 150 ng/mL) and "atypical" symptoms, an example of which is "headaches and diminished libido in the presence of regular menses." However, investigators who have reported on macroPRL

prevalence and the overlap in clinical presentation with hyperprolactinemia tend to advocate for routine testing in all hyperprolactinemic patients (2,13,15,45). Universal screening for macroPRL in hyperprolactinemic patients has been more widely adopted in the United Kingdom, Ireland, and Europe compared to North America (2).

Barring universal testing, there can be additional clinical clues to improve case-finding for macroPRL beyond an asymptomatic patient; some of these atypical presentations are suggested in Figure 2. Because PRL inhibits the pulsatile secretion of gonadotropin-releasing hormone, the presence of appropriate gonadotropin (e.g., LH) and/or sex hormone levels can be an important clue to the diagnosis of macroPRL (31,45,47). In the studies summarized in Table 1, the prevalence of galactorrhea was lower for macroPRL (31,45,47,53), so that a female presenting with menstrual irregularity but without galactorrhea may also be a candidate for testing (Fig. 2). Although a true association with autoimmune disease has been difficult to confirm,

macroPRL may be considered when combined with an atypical clinical picture, as discussed above. Although a subset of macroPRL patients has been shown by some to respond to DAs, apparent DA resistance, such as lack of reduction or normalization of PRL, could also be an indication for macroPRL testing.

CONCLUSION

A significant proportion of patients with immunoreactive hyperprolactinemia may have elevated macroPRL. It can sometimes be challenging to identify patients who are candidates for macroPRL testing. However, it is important to appreciate the implications of the diagnosis of macroPRL on health care utilization, laboratory testing, imaging, and unnecessary medical or surgical treatment. In some practices and health care systems, universal testing of all hyperprolactinemic patients is the norm. Until PRL assays can reliably exclude macroPRL, clinicians should

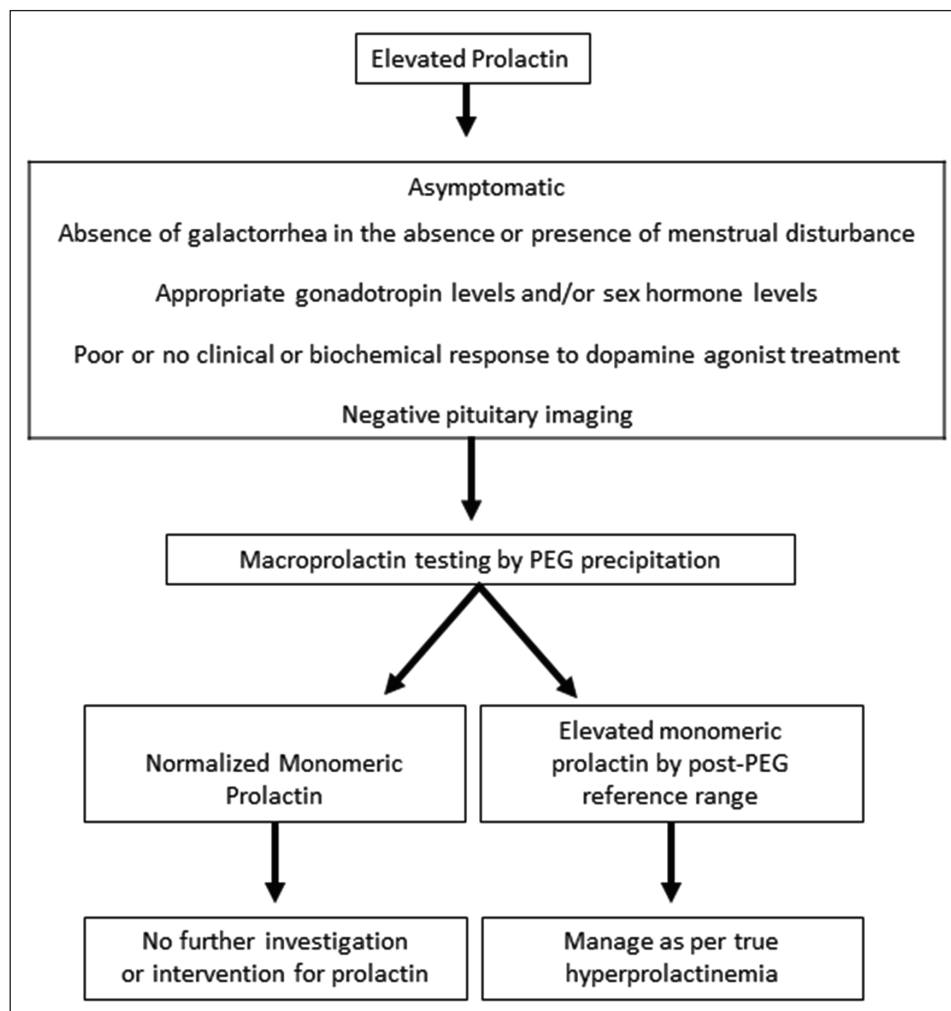


Fig. 2. Algorithm for consideration of macroprolactin testing in hyperprolactinemic patients. Prior to PEG precipitation, the majority of patients with macroprolactinemia will have prolactin values in the 25 to 150 ng/mL range (18,31,42,49). PEG = polyethylene glycol.

be aware of the potential for its interference. Also, we must emphasize the need to exclude monomeric PRL elevation in patients with macroPRL as the former may require additional investigations and treatment.

DISCLOSURE

The authors have no multiplicity of interest to disclose.

REFERENCES

- Freeman ME, Kanyicska B, Lerant A, Nagy G. Prolactin: structure, function, and regulation of secretion. *Physiol Rev.* 2000;80:1523-1631.
- Fahie-Wilson M, Smith TP. Determination of prolactin: the macroprolactin problem. *Best Pract Res Clin Endocrinol Metab.* 2013;27:725-742.
- Tritos NA, Guay AT, Malarkey WB. Asymptomatic 'big' hyperprolactinemia in two men with pituitary adenomas. *Eur J Endocrinol.* 1998;138:82-85.
- Hattori N, Ishihara T, Saiki Y. Macroprolactinaemia: prevalence and aetiologies in a large group of hospital workers. *Clin Endocrinol (Oxf).* 2009;71:702-708.
- Soong YK, Ferguson KM, McGarrick G, Jeffcoate SL. Size heterogeneity of immunoreactive prolactin in hyperprolactinaemic serum. *Clin Endocrinol (Oxf).* 1982;16:259-265.
- Jackson RD, Wortsman J, Malarkey WB. Macroprolactinemia presenting like a pituitary tumor. *Am J Med.* 1985;78:346-350.
- Bonhoff A, Vuille JC, Gomez F, Gellersen B. Identification of macroprolactin in a patient with asymptomatic hyperprolactinemia as a stable PRL-IgG complex. *Exp Clin Endocrinol Diabetes.* 1995;103:252-255.
- Hattori N, Ikekubo K, Ishihara T, Moridera K, Hino M, Kurahachi H. A normal ovulatory woman with hyperprolactinemia: presence of anti-prolactin autoantibody and the regulation of prolactin secretion. *Acta Endocrinol (Copenh).* 1992;126:497-500.
- Cavaco B, Leite V, Santos MA, Arranhado E, Sobrinho LG. Some forms of big big prolactin behave as a complex of monomeric prolactin with an immunoglobulin G in patients with macroprolactinemia or prolactinoma. *J Clin Endocrinol Metab.* 1995;80:2342-2346.
- Hattori N, Ikekubo K, Nakaya Y, Kitagawa K, Inagaki C. Immunoglobulin G subclasses and prolactin (PRL) isoforms in macroprolactinemia due to anti-PRL autoantibodies. *J Clin Endocrinol Metab.* 2005;90:3036-3044.
- Hattori N, Nakayama Y, Kitagawa K, Ishihara T, Saiki Y, Inagaki C. Anti-prolactin (PRL) autoantibody-binding sites (epitopes) on PRL molecule in macroprolactinemia. *J Endocrinol.* 2006;190:287-293.
- Hattori N, Ishihara T, Saiki Y, Shimatsu A. Macroprolactinaemia in patients with hyperprolactinaemia: composition of macroprolactin and stability during long-term follow-up. *Clin Endocrinol (Oxf).* 2010;73:792-797.
- McKenna TJ. Should macroprolactin be measured in all hyperprolactinaemic sera? *Clin Endocrinol (Oxf).* 2009;71:466-469.
- Olukoga AO, Kane JW. Macroprolactinaemia: validation and application of the polyethylene glycol precipitation test and clinical characterization of the condition. *Clin Endocrinol (Oxf).* 1999;51:119-126.
- Jamaluddin FA, Sthaneshwar P, Hussein Z, Othman N, Chan SP. Importance of screening for macroprolactin in all hyperprolactinaemic sera. *Malays J Pathol.* 2013;35:59-63.
- Vieira JG, Tachibana TT, Obara LH, Maciel RM. Extensive experience and validation of polyethylene glycol precipitation as a screening method for macroprolactinemia. *Clin Chem.* 1998;44:1758-1759.
- Bjøro T, Mørkrid L, Wergeland R, et al. Frequency of hyperprolactinaemia due to large molecular weight prolactin (150-170 kD PRL). *Scand J Clin Lab Invest.* 1995;55:139-147.
- Hauache OM, Rocha AJ, Maia AC Jr, Maciel RM, Vieira JG. Screening for macroprolactinaemia and pituitary imaging studies. *Clin Endocrinol (Oxf).* 2002;57:327-331.
- Leite V, Cosby H, Sobrinho LG, Fresnoza MA, Santos MA, Friesen HG. Characterization of big, big prolactin in patients with hyperprolactinaemia. *Clin Endocrinol (Oxf).* 1992;37:365-372.
- Kavanagh L, Smith TP, McKenna TJ. Bioactivity of macroprolactin in the Nb2 bioassay may be explained by dissociation yielding bioactive monomeric prolactin. *Clin Endocrinol (Oxf).* 2007;67:954.
- Glezer A, Soares CR, Vieira JG, et al. Human macroprolactin displays low biological activity via its homologous receptor in a new sensitive bioassay. *J Clin Endocrinol Metab.* 2006;91:1048-1055.
- Hattori N, Nakayama Y, Kitagawa K, Ishihara T, Saiki Y, Inagaki C. Anti-prolactin (PRL) autoantibodies suppress PRL bioactivity in patients with macroprolactinaemia. *Clin Endocrinol (Oxf).* 2008;68:72-76.
- Hattori N, Nakayama Y, Kitagawa K, Li T, Inagaki C. Development of anti-PRL (prolactin) autoantibodies by homologous PRL in rats: a model for macroprolactinemia. *Endocrinology.* 2007;148:2465-2470.
- Adachi T, Hattori N, Ishihara T, et al. Possible involvement of matrix metalloproteinase-3 in the pathogenesis of macroprolactinaemia in some patients with rheumatoid arthritis. *Eur J Endocrinol.* 2013;169:203-209.
- Malarkey WB, Jackson R, Wortsman J. Long-term assessment of patients with macroprolactinemia. *Fertil Steril.* 1988;50:413-418.
- Lewandowski KC, Gasiór-Perczak D, Kowalska A, Lewiński A. Prevalence of macroprolactinaemia in regularly menstruating women with non-toxic goitre or autoimmune thyroid disease. *Thyroid Res.* 2012;5:20.
- Kavanagh-Wright L, Smith TP, Gibney J, McKenna TJ. Characterization of macroprolactin and assessment of markers of autoimmunity in macroprolactinaemic patients. *Clin Endocrinol (Oxf).* 2009;70:599-605.
- Cárdenas-Mondragón G, Ulloa-Aguirre A, Isordia-Salas I, Goffin V, Leños-Miranda A. Elevated serum bioactive prolactin concentrations in patients with systemic lupus erythematosus are associated with disease activity as disclosed by homologous receptor bioassays. *J Rheumatol.* 2007;34:1514-1521.
- Leños-Miranda A, Pascoe-Lira D, Chávez-Rueda KA, Blanco-Favela F. Detection of macroprolactinemia with the polyethylene glycol precipitation test in systemic lupus erythematosus patients with hyperprolactinemia. *Lupus.* 2001;10:340-345.
- Wallace IR, Satti N, Courtney CH, et al. Ten-year clinical follow-up of a cohort of 51 patients with macroprolactinemia establishes it as a benign variant. *J Clin Endocrinol Metab.* 2010;95:3268-3271.

31. **Gibney J, Smith TP, McKenna TJ.** The impact on clinical practice of routine screening for macroprolactin. *J Clin Endocrinol Metab.* 2005;90:3927-3932.
32. **Yavuz D, Topçu G, Ozener C, Akalin S, Sirikçi O.** Macroprolactin does not contribute to elevated levels of prolactin in patients on renal replacement therapy. *Clin Endocrinol (Oxf).* 2005;63:520-524.
33. **Gulcelik NE, Usman A.** Macroprolactinaemia in diabetic patients. *Neuro Endocrinol Lett.* 2010;31:270-274.
34. **Sari F, Sari R, Ozdem S, Sarikaya M, Cetinkaya R.** Serum prolactin and macroprolactin levels in diabetic nephropathy. *Clin Nephrol.* 2012;78:33-39.
35. **Schneider W, Marcovitz S, Al-Shammari S, Yago S, Chevalier S.** Reactivity of macroprolactin in common automated immunoassays. *Clin Biochem.* 2001;34:469-473.
36. **Smith TP, Suliman AM, Fahie-Wilson MN, McKenna TJ.** Gross variability in the detection of prolactin in sera containing big big prolactin (macroprolactin) by commercial immunoassays. *J Clin Endocrinol Metab.* 2002;87:5410-5415.
37. **Ellis MJ, Reed MR, Livesey JH.** Cross-reactivities of macroprolactin and big-prolactin in three commercial immunoassays for prolactin: a chromatographic analysis. *Clin Biochem.* 2007;40:1285-1290.
38. **Kavanagh L, McKenna TJ, Fahie-Wilson MN, Gibney J, Smith TP.** Specificity and clinical utility of methods for the detection of macroprolactin. *Clin Chem.* 2006;52:1366-1372.
39. **Leslie H, Courtney CH, Bell PM, et al.** Laboratory and clinical experience in 55 patients with macroprolactinemia identified by a simple polyethylene glycol precipitation method. *J Clin Endocrinol Metab.* 2001;86:2743-2746.
40. **Ellis MJ, Livesey JH, Soule SG.** Macroprolactin, big-prolactin and potential effects on the misdiagnosis of hyperprolactinemia using the Beckman Coulter Access Prolactin assay. *Clin Biochem.* 2006;39:1028-1034.
41. **Beda-Maluga K, Pisarek H, Komorowski J, Swietoslowski J, Fuss-Chmielewska J, Winczyk K.** Evaluation of hyperprolactinaemia with the use of the intervals for prolactin after macroforms separation. *J Physiol Pharmacol.* 2014;65:359-364.
42. **Strachan MW, Teoh WL, Don-Wauchope AC, Seth J, Stoddart M, Beckett GJ.** Clinical and radiological features of patients with macroprolactinaemia. *Clin Endocrinol (Oxf).* 2003;59:339-346.
43. **Ram S, Harris B, Fernando JJ, Gama R, Fahie-Wilson M.** False-positive polyethylene glycol precipitation tests for macroprolactin due to increased serum globulins. *Ann Clin Biochem.* 2008;45:256-259.
44. **Lewandowski KC, Gasiior-Perczak D, Kowalska A, Lewinski A.** Coexistence of macroprolactinaemia and hyperprolactinaemia in women with oligo-/amenorrhoea is associated with high risk of pituitary adenomas. *Gynecol Endocrinol.* 2014;30:385-387.
45. **Suliman AM, Smith TP, Gibney J, McKenna TJ.** Frequent misdiagnosis and mismanagement of hyperprolactinemic patients before the introduction of macroprolactin screening: application of a new strict laboratory definition of macroprolactinemia. *Clin Chem.* 2003;49:1504-1509.
46. **Molitch ME, Russell EJ.** The pituitary "incidentaloma". *Ann Intern Med.* 1990;112:925-931.
47. **Isik S, Berker D, Tutuncu YA, et al.** Clinical and radiological findings in macroprolactinemia. *Endocrine.* 2012;41:327-333.
48. **McCudden CR, Sharpless JL, Grenache DG.** Comparison of multiple methods for identification of hyperprolactinemia in the presence of macroprolactin. *Clin Chim Acta.* 2010;411:155-160.
49. **Tamer G, Telci A, Mert M, et al.** Prevalence of pituitary adenomas in macroprolactinemic patients may be higher than it is presumed. *Endocrine.* 2012;41:138-143.
50. **de Soárez PC, Souza SC, Vieira JG, Ferraz MB.** The effect of identifying macroprolactinemia on health-care utilization and costs in patients with elevated serum prolactin levels. *Value Health.* 2009;12:930-934.
51. **Melmed S, Casanueva FF, Hoffman AR, et al.** Diagnosis and treatment of hyperprolactinemia: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab.* 2011;96:273-288.
52. **Casanueva FF, Molitch ME, Schlechte JA, et al.** Guidelines of the Pituitary Society for the diagnosis and management of prolactinomas. *Clin Endocrinol (Oxf).* 2006;65:265-273.
53. **Vallette-Kasic S, Morange-Ramos I, Selim A, et al.** Macroprolactinemia revisited: a study on 106 patients. *J Clin Endocrinol Metab.* 2002;87:581-588.