

Case Report

Macroprolactinemia in a young man and review of the literature

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ABSTRACT

A 21-year old man, complaining of headaches and fatigue, with a negative past medical history and a normal clinical examination, underwent a hormonal investigation which revealed hyperprolactinemia and intact pituitary-gonadal axis. Drug-induced hyperprolactinemia was excluded. Pituitary magnetic resonance imaging indicated a microadenoma in the right part of the gland, with a diameter of 1.5mm. No medical treatment was given as the patient had no symptoms relevant to prolactin excess. The PEG precipitation test was carried out and showed 7% recovery, which was diagnostic of the macroprolactinemia. Relatively few cases of macroprolactinemia have been published in the literature, although the condition is regarded as a fairly common cause of hyperprolactinemia. Macroprolactinemic men represent 10% of published cases.

Key words: macroprolactinemia, PEG precipitation test

INTRODUCTION

Three main forms of prolactin have been identified by gel filtration chromatography in human serum. These are monomeric PRL with a molecular weight 23kDa, big PRL with a molecular weight 50-60kDa and big-big PRL (or macroprolactin) with a molecular weight >100kDa (150-170kDa)¹. Monomeric PRL is the main circulating form of prolactin (approximately 85%) and accounts for the majority of PRL immunoreactivity in serum of normal individuals and of most patients with hyperprolactinemia. However, in some patients with hyperprolactinemia high molecular weight forms of PRL, mainly big-big PRL, pre-

dominate. This condition has been known for many years^{2,3} and termed macroprolactinemia³. Macroprolactin remains reactive in immunoassays for prolactin and is cleared more slowly than monomeric prolactin from the circulation, as shown from clearance studies^{4,5}, leading to apparent hyperprolactinemia.

Confirmation of this situation required, till recently, the use of gel filtration chromatography, a labor-intensive and expensive technique. Consequently, no good routine test for macroprolactinemia existed, so that the frequency and clinical implications of hyperprolactinemia have not been clearly established. Recently, a precipitation test using polyethylene glycol (PEG), a chemical that precipitates large molecular weight proteins, has been used to identify the presence of macroprolactin in serum⁶⁻¹⁰. Macroprolactin, if present in serum, is precipitated by PEG, leaving monomeric PRL in the supernatant.

Relatively few cases of macroprolactinemia have

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been published in the literature and from the existing data, the prevalence among hyperprolactinemic patients ranges from 9%-36%⁷⁻¹². Macroprolactinemic men represent 10% of published cases.

We present a 21-year old man with hyperprolactinemia who was suspected of having macroprolactinemia because of the discrepancy between clinical, hormonal and neuroradiological data. The presence of macroprolactinemia was confirmed via the PEG precipitation test.

CASE REPORT

A 21-year old man complained of headaches and fatigue. High prolactin value was detected and the patient was referred to our Department. A complete history and physical examination were negative for symptoms and signs associated with hyperprolactinemia. Drug-induced hyperprolactinemia was excluded. Initial investigation included general laboratory screening tests, which were normal, and baseline hormone levels, using commercial electrochemiluminescent immunoassay kits (Elecsys, Roche Diagnostics GmbH, Mannheim). T3, free T4, TSH, FSH, LH, testosterone, estradiol and cortisol were within the normal range: T3: 2,1nmol/L (1,2-3,1), T4: 111,2nmol/L (70,8-173,7), free-T4: 18,7pmol/L (11,6-25,7), TSH: 3,0mU/L (0,3-4,2), FSH: 3,3IU/L (1,5-12,4), LH: 3,6IU/L (1,7-8,6), testosterone: 27,7nmol/L (9,7-27,7), E2: 159,7pmol/L (40,4-161,5), F: 469,3nmol/L (190,0-690,0). Thyroid peroxidase and thyroglobulin antibodies were negative. PRL was measured using a sandwich-format electrochemiluminescent assay using two mouse monoclonal antibodies specifically directed against human PRL (Elecsys assay, Roche Diagnostics, GmbH, Mannheim). Intra- and interassay coefficients of variation were less than 5% and sensitivity was 10mIU/L. PRL levels by this method ranged from 2.508-2.562mIU/L (normal range <444mIU/L). A GnRH stimulation test was performed using standard procedures and a normal gonadotropin response was noted. Pituitary magnetic resonance imaging with IV gadolinium suggested a microadenoma in the right part of the gland, with a diameter of 1.5mm. Despite the fact that the possibility of a microprolactinoma could not be excluded, the absence of symptomatology relevant to prolactin excess and the presence of intact pituitary-gonadal axis led us to consider the possibility of macroprolactinemia and no medical

treatment was given.

A new blood sample was obtained and the PEG precipitation test was performed, as it has been previously described^{6,8}. PRL was measured in the untreated serum and in the supernatant, after treating the serum with a 25% solution of PEG 6000 and centrifugation, using the same assay (Elecsys assay, Roche Diagnostics). The result was expressed as prolactin recovery (%R), derived as a percentage of the PRL measured in the supernatant relative to that measured in the untreated serum, using the formula: % recovery: $100 \times [\text{PRL value after PEG precipitation} \times 2 / \text{PRL value before PEG precipitation}]$ (PRL concentration measured in the supernatant is adjusted by a factor of two to correct for dilution in the preparation). A PRL recovery of 7% was found in our patient, identifying the presence of macroprolactinemia, as PRL recoveries of less than 40% are considered diagnostic of macroprolactinemia.

DISCUSSION

Macroprolactinemia is characterized by the predominance of high molecular weight forms of PRL, mainly big-big PRL (macroprolactin), in the circulation. The structure of macroprolactin has not yet been fully defined. Recent studies have shown that macroprolactin is primarily a complex of monomeric PRL with an IgG antibody, thought to be directed against the PRL molecule (antiprolactin autoantibody)^{6,13-15}. In some cases macroprolactin was identified as a heterogeneous complex of covalently and noncovalently bound PRL with increased glycosylation^{3,16}. Most probably, different etiologies result in the formation of macroprolactin, which seems to be a postsecretory event. The peripheral origin of macroprolactin is supported by data showing a normal PRL chromatographic pattern (predominance of monomeric PRL) in the culture media of two pituitary adenomas removed from patients with macroprolactinemia¹⁷.

Clearance studies have shown that macroprolactin is cleared more slowly than monomeric prolactin from the circulation; this may, therefore, lead to apparent hyperprolactinemia, as PRL immunoassays in routine use exhibit variable degrees of reactivity with macroprolactin^{14,15,18}. A possible explanation for the variable detection of macroprolactin is that PRL is a polypeptide with several antigenic sites and therefore,

if the endogenous autoantibody and the antibodies used in the assay react with different epitopes, macroprolactin is detected to a high degree; however, if the endogenous autoantibody and any of the two reagent antibodies react with the same epitope, the assay will not be able to detect macroprolactin^{12,18,19}. The immunoassay used in our laboratory (Elecsys assay) is highly sensitive to the presence of macroprolactin, resulting in high PRL levels, as shown by recent comparative studies of various commercial immunoassays of PRL^{12,20,21}.

The PEG precipitation test is a relatively simple and inexpensive technique, reproducible and sensitive for the detection of macroprolactin⁷⁻⁹, but not specific or quantitative. Results generated by using the PEG precipitation test correlate well with those of gel filtration^{7,9}, although numerically they differ significantly, a finding that can be explained by the non-specific way PEG reduces protein solubility²¹. A prolactin recovery (%R) is derived as a percentage of the PRL measured in the supernatant relative to that measured in the untreated serum. A percentage recovery less than 40% indicates the presence of macroprolactinemia, a percentage recovery greater than 50% indicates the absence of macroprolactinemia, while a percentage recovery between 40% and 50% represents a gray area and further investigation is needed (gel filtration chromatography)^{9,11,12,20}. Wider cut-off limits have been proposed in some studies^{8,21} classifying recoveries between 30% and 65% as a gray area⁸, but these values have not been generally accepted. The PEG precipitation test can be used in routine laboratory practice to detect macroprolactinemia, although it should be borne in mind that PEG interferes with a considerable number of commonly used immunoassay methodologies²¹. It should be noted that PEG does not interfere with the immunoassay used in our laboratory (Elecsys assay)^{12,20,21}.

The frequency of macroprolactinemia has not been established. Studies that used the PEG precipitation test for the detection of macroprolactinemia have reported a prevalence between 9%¹² and 36%⁸ in hyperprolactinemic patients, whereas a very recent study using gel filtration chromatography in a selected group of hyperprolactinemic patients reported a prevalence of 29%¹⁷. Macroprolactinemia is seen in both sexes, although women represent 90% of published cases at all ages, including children^{22,23}. The clinical implica-

tions of macroprolactinemia have not been determined, with some reports (mostly case reports) documenting an asymptomatic condition^{2,3,24,25}, as the case we present and other reports documenting symptomatology in a number of patients. Recently, studies with large series of patients showed that a significant proportion of macroprolactinemic patients present with clinical symptoms of prolactin excess^{9,11,17,20}. In women, galactorrhea and menstrual disorders are the most usual symptoms, while fertility is usually preserved. In men, although sexual dysfunction constitutes the most frequent ground for PRL evaluation, gonadotroph function is not affected^{20,26}.

Recent *in vitro* studies have shown that IgG-bound PRL is fully bioactive using the NB2 lymphoma cell bioassay^{4,13}, although earlier studies have demonstrated no bioactivity²⁷. If IgG-bound PRL is biologically active, the effects may be blunted because of decreased bioavailability. Because of its high molecular mass, the autoimmune complex is confined to the vasculature; thus, given the limited capacity to cross vascular endothelium, the large PRL-IgG complex fails to reach receptors of target cells and hence exhibits limited bioactivity *in vivo*.

Macroprolactinemia may coexist with any other cause of hyperprolactinemia and pituitary lesion¹⁷, indicating that this diagnosis is not limited in the context of idiopathic hyperprolactinemia. However, the majority of macroprolactinemic patients have no abnormalities on pituitary imaging^{9,11,17,28}. Pituitary microadenomas have been described in 3.7 to 21.9% of macroprolactinemic patients, most of them being non-functioning (pituitary incidentalomas)^{9,11,17,28}. In our patient, pituitary imaging was inconclusive of a microadenoma. If the presence of a microadenoma is confirmed in a new pituitary magnetic resonance imaging, this should be considered as a nonfunctioning adenoma.

We did not recommend medical treatment to our patient as he had no symptoms relevant to prolactin excess. Dopamine agonists have been given in symptomatic patients, resulting in alleviations of symptoms in some cases^{9,11,17}. On dopamine agonist therapy, normalization of PRL levels was observed in less than a half of the patients^{3,9,17}. Pseudoresistance to dopamine agonist therapy may be considered in cases where there is a failure of normalization of PRL levels¹⁷.

We intend to follow up our patient since a spontaneous decline in PRL levels has been reported in three patients with macroprolactinemia²⁹. How long PRL levels should be monitored in macroprolactinemic patients remains an open question.

In conclusion, macroprolactinemia, seems to represent a relatively common cause of hyperprolactinemia and should be sought in hyperprolactinemic patients displaying discrepant clinical, biological and/or follow up data, as was the case of our male patient. In macroprolactinemia, PRL levels are usually modestly elevated. The PEG precipitation test is a reproducible and sensitive method, which allows detection of macroprolactin easily and inexpensively. Dopamine agonist therapy may sometimes be beneficial but does not result in normalization of PRL levels in a significant number of patients. Pituitary lesions are rarely found and possibly constitute incidentalomas. By confirming the diagnosis of macroprolactinemia, repeated hormonal and neuroradiological investigations as well as unnecessary treatments can be avoided.

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