A 12-cm Mass with No Symptoms and Unremarkable Laboratory Results

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CASE DESCRIPTION

A 60-year-old man presented to his general practitioner with complaints of indigestion and flu-like symptoms. The results of a clinical examination were unremarkable. The only abnormality found in routine hematology and biochemistry investigations was an increased serum alkaline phosphatase concentration (131 U/L; reference interval, 35–120 U/L). Given his abdominal discomfort, the patient was referred for an abdominal ultrasound evaluation, which revealed a 12-cm cystic mass in the area of the left kidney. Further imaging with computed tomography scanning suggested that the mass arose from the left adrenal gland (Fig. 1). The patient’s medical history was notable for hypertension, which was controlled with 8 mg candesartan once daily. The patient reported no headaches, palpitations, or diaphoresis.

The patient was referred for further investigation. Plasma free metanephrines (PMets)6 were measured. Initial results showed borderline increases in plasma normetanephrine (NMN) and metanephrine (MN) that were not diagnostic of pheochromocytoma (PCC). PMet measurements were repeated with a separate plasma sample, and total fractionated urine MNs (UMets) were measured in a sample of an acidified 24-h urine collection. Urine and plasma analysis results were concordant, with borderline increases in NMN (Table 1). The patient was not on any medications known to cause a physiological increase in PMets.

CASE FOLLOW-UP

On the basis these findings, the size of the mass, and the lack of interfering medications, the patient underwent α-blockade with 10 mg phenoxybenzamine 3 times per day and surgical resection of the adrenal mass. A subsequent histologic analysis revealed a cystic PCC with evidence of intratumoral hemorrhage and degeneration. PMet concentrations returned to normal after tumor resection (Table 1). Subsequent testing identified no germline mutations in PCC-predisposing genes, a result consistent with sporadic disease. During follow-up the patient remained normotensive and off antihypertensive medication.

DISCUSSION

PCC and paraganglioma (PGL) are rare neuroendocrine tumors arising from chromaffin cells. PCCs are typically found in the adrenal medulla, accounting for 80%–85% of cases. PGLs are closely related extraadrenal tumors that can arise in sympathetic (potentially catecholamine-producing) or parasympathetic (non–catecholamine-producing) ganglia.

Estimates of the prevalence of PCC in hypertensive populations vary between 0.1% and 0.6% (1). Advances in imaging and screening for familial disease have led to an increased frequency of diagnosis in normotensive and asymptomatic patients. It is estimated that 1.5%–23% of all incidentally detected adrenal masses (incidentalomas) are PCCs (2).

PCC has long been regarded as a difficult pathology to diagnose; thus, sensitive and specific diagnostic tests are required. The interpretation of results is also critical to identifying cases. As the number of diagnosed PCCs has increased, a subgroup of tumors, termed “cystic PCC and PGL,” has been identified. These cystic tumors are characterized by a thick wall with no internal septa on imaging, with the enhance-

QUESTIONS TO CONSIDER

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<td>1. How should borderline increases in MNs be interpreted and followed up?</td>
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<td>2. How best should one integrate clinical, radiologic, and biochemical features of adrenal masses?</td>
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<td>3. How do sampling conditions and medications affect MN and NMN concentrations?</td>
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6 Nonstandard abbreviations: PMets, plasma metanephrines; NMN, normetanephrine; MN, metanephrine; UMets, urine MNs; PCC, pheochromocytoma.
ment persisting after the administration of contrast medium. The presence of a thick wall may reduce the passage of catecholamine metabolites into the plasma and urine, making an early diagnosis difficult if only biochemical investigations are performed.

**BIOCHEMICAL EVALUATION OF PCC**

The first-line biochemical investigation for PCC/PGL is the measurement of urine or plasma NMN and MN (3). Measurement of 3-methoxytyramine is not recommended, but it reportedly is useful for distinguishing patients with and without metastases. 3-Methoxytyramine has been shown to be a diagnostically more sensitive biomarker of tumoral dopamine production than plasma or urinary dopamine (4).

The most common reasons for biochemical testing are: (a) presence of both hypertension and episodic symptoms of catecholamine excess (e.g., palpitations,

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**Fig. 1. Computed tomography images.**

Coronal (A) and axial (B) images showing a 12-cm mass in the left adrenal gland.
headache, periods of diaphoresis); (b) treatment of resistant hypertension; (c) assessment of functionality of an adrenal incidentaloma; and (d) predisposition to hereditary PCC/PGL.

Hereditary PCC/PGL is of increasing interest because 10 genes are now known to play an important role in the pathogenesis of PCC. These genes include the RET (ret proto-oncogene), VHL (von Hippel-Lindau tumor suppressor, E3 ubiquitin protein ligase), and NF1 (neurofibromin 1) tumor suppressor genes. Additional genes encoding the SDHD (succinate dehydrogenase complex, subunit D, integral membrane protein) gene and the newly described TMEM127 (transmembrane protein 127) and MAX (MYC associated factor X) tumor suppressor genes are also known to contribute to the pathogenesis of PCC (5).

**MN MEASUREMENT**

The most widely used method for measuring urine catecholamines and their methylated metabolites has been HPLC with electrochemical detection. Although this method is analytically sensitive, the instrument requires frequent maintenance and is prone to analytical interference. Laboratories are increasingly measuring PMets and UMets with liquid chromatography–tandem mass spectrometry. This method is subject to less analytical interference and provides greater analytical sensitivity and precision. Immunoassays are also available for PMet measurement. Differences in calibration and matrix effects are believed to be responsible for the observed differences between results produced by chromatographic and immunoassay methods (6). Some laboratories prefer to use immunoassays, because they are simple to perform and do not require a large capital investment. Results should be interpreted with method-derived reference intervals.

**INTERPRETATION OF BORDERLINE MN INCREASES**

Because of the high diagnostic sensitivity of MNs, a “negative” result (i.e., within the reference interval) excludes the presence of a PCC/PGL in the majority of cases. False negatives (normal MN in the presence of a tumor) are rare. Most diagnostic pitfalls center around the interpretation of results in the “borderline-positive interval” in patient populations with a low prevalence of PCC/PGL. In such cases, a false positive (increase of MNs in the absence of a tumor) due to a confounding factor such as concomitant drug therapy or a physiological increase in catecholamine release is more likely than a true positive (increase in MNs due to the presence of a tumor).

Borderline increases in PMets or UMets have been defined as anything between 1 and 3 times the upper limit of the reference interval (7).

Defining the pretest probability of harboring a PCC/PGL in the individual patient is key to an appropriate interpretation of results, which should not be based entirely on the magnitude of the increase above the reference interval. One should also consider the reason for investigation, as illustrated by the present case.

Assurance when one evaluates the importance of a borderline result can be gained by collecting a repeat plasma or urine sample under standardized low-stress conditions (i.e., collecting a fasting plasma sample in the supine position and avoiding urine collections after exercise) and by demonstrating similar patterns of MNs in the plasma and urine. In addition, patients should have potentially interfering medication stopped and/or any underlying conditions managed optimally before retesting.

Further clarification can be achieved by performing a clonidine suppression test. Därr et al. (8) proposed that this test in combination with measurements of plasma NMN can be used to confirm or exclude PCC/PGL in patients with borderline-increased test results.

In a small case series of adrenal incidentaloma, 100% of patients with normal PMets were found not to

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**Table 1. Plasma free MN and urine total fractionated MN results.**

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<thead>
<tr>
<th>Analyte</th>
<th>Initial</th>
<th>Follow-up</th>
<th>Postsurgery</th>
<th>Reference interval</th>
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<tbody>
<tr>
<td>Plasma NMN, pmol/L</td>
<td>1348</td>
<td>1487</td>
<td>211</td>
<td>120–1180</td>
</tr>
<tr>
<td>Plasma MN, pmol/L</td>
<td>598</td>
<td>638</td>
<td>106</td>
<td>80–510</td>
</tr>
<tr>
<td>Urine NMN, µmol/24 h</td>
<td>6.6</td>
<td>3.8</td>
<td>2.2</td>
<td>&gt;3.8</td>
</tr>
<tr>
<td>Urine MN, µmol/24 h</td>
<td>0.9</td>
<td></td>
<td></td>
<td>&lt;2.2</td>
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*a Plasma MNs were measured with liquid chromatography–tandem mass spectrometry. Samples were collected with the patient in a seated position after a 20-min rest. Reference intervals are based on a seated reference population.

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7 Human genes: RET, ret proto-oncogene; VHL, von Hippel-Lindau tumor suppressor, E3 ubiquitin protein ligase; NF1, neurofibromin 1; SDHD, succinate dehydrogenase complex, subunit D, integral membrane protein; TMEM127, transmembrane protein 127; MAX, MYC associated factor X.
have PCC/PGL, and all patients with PMet concentrations greater than twice the upper limit of normal had a PCC/PGL (9). This leaves a gray area where PMet concentrations are between 1 and 2 times the reference interval.

The population prevalence of adrenal incidentaloma is higher than PCC. A thickened wall, lack of internal septa, and persistent enhancement after the use of contrast media may indicate the possibility of a cystic PCC. This rare pathology may be associated with minimally increased or even normal MN values.

**DRUGS THAT CAUSE FALSE-POSITIVE RESULTS**

Drugs are often implicated in the production of increased PMet concentrations. The majority of drug effects occur through alteration of catecholamine production in vivo. Tricyclic antidepressants, selective serotonin reuptake inhibitors, and tetracyclines (e.g., venlafaxine) inhibit the reuptake of norepinephrine at the nerve terminal.

The nonspecific α-blocker phenoxybenzamine produces a high false-positive rate (10). Calcium channel blockers can alter norepinephrine concentrations and consequently cause increased NMN concentrations. Beta-blockers can also cause false-positive results; however, such findings are relatively infrequent. Thus, there is no need to stop treatment before initial screening. All other antihypertensive medications can be continued during testing and need only be withdrawn if confirmation of a borderline-positive result is required.

This case provides a clear illustration that for this increasingly recognized subtype of tumor, current assays may not provide unequivocal results of increased concentrations, and the concentrations of catecholamine metabolites should be interpreted in conjunction with the results of imaging when possible. This case also highlights the importance of considering a borderline increase in catecholamine metabolites in a clinical context. Further investigation may be appropriate in patients without previous imaging if symptoms are ongoing and other causes have been excluded.

**POINTS TO REMEMBER**

- Measurements of urine or plasma MNs are the recommended biochemical screening test for PCC/PGL. These biomarkers are not prone to the fluctuations seen for catecholamines and can be measured in the form of plasma free MNs and urinary fractionated MNs.

- There are different methods for analyzing and detecting both urine and plasma MNs, including HPLC with electrochemical detection, liquid chromatography–tandem mass spectrometry, and immunoassay. Reference intervals are method dependent and are not interchangeable between methods.

- The pretest probability of a patient having a PCC/PGL should be carefully considered before testing, because that will help in the interpretation of results and the decision on further action.

- Radiologic features, such as a thickened wall, lack of internal septa, and enhancement persisting after the use of contrast media may indicate the possibility of an unusual pathology, such as a patient with a cystic PCC who may have very minimally increased or even normal MN concentrations.

**References**