

## Pheochromocytoma

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Pheochromocytomas (PCCs)<sup>10</sup> are catecholamine-secreting tumors derived from chromaffin tissue of the adrenal medulla. Closely related tumors, called extraadrenal paragangliomas (PGL), can arise at extraadrenal sites. Catecholamine secretion from these tumors is often episodic, causing headache, perspiration, palpitations, and hypertension. If not recognized and treated, PCC and PGL (PPGL) can lead to arrhythmia, myocardial infarction, stroke, and death.

The diagnosis of PPGL relies on biochemical evidence of excess catecholamine secretion and confirmation of tumor presence by imaging studies. Although many different biochemical tests have historically been used in screening for PPGL, measurements of the catecholamine breakdown products metanephrine and normetanephrine in plasma and urine are now regarded as the first-line tests. Florid increases of either of these metabolites are associated with a nearly 100% probability of PPGL. However, it can be challenging to differentiate between true-positive and false-positive results when metanephrine or normetanephrine concentrations are only slightly above the upper limit of the respective reference interval.

Not long ago, approximately 90% of PPGLs were believed to occur sporadically. However, germline mutations in 10 different genes have been shown to cause PPGLs, and at least 30% of these tumors are now known to be hereditary. Importantly, genotype–phenotype correlations have been elucidated: different mutations are associated with specific clinical features and sites of disease, the production of cer-

tain catecholamines, and varying frequency of malignancy.

In this Q&A article, 5 experts discuss the state of the art in the diagnosis, localization, and treatment of PPGL. They also provide their opinions on the role of genetic testing in the diagnosis and management of patients with these tumors.

**What is your estimate of the prevalence of PPGL? Are certain populations at increased risk for developing these tumors?**



**Graeme Eisenhofer:** Early autopsy series indicated prevalences of PCC of 1 per 1000, with more recent series indicating a lower prevalence of 1 per 2000, suggesting that detection rates in living individuals have improved. Nevertheless, at reported annual detection rates of 2–5 per million, corre-

sponding to prevalences of 1.5–4 per 10 000, it seems that most cases remain undiagnosed during life. This probably also holds true for extraadrenal paragangliomas, which have a prevalence of about 15% that of the adrenal tumors.

Populations at increased risk for PPGL are those with germline mutations of the now identified 10 tumor-susceptibility genes. Other populations at increased risk that should be screened for the tumors include individuals with a previous history of the disease or with adrenal lesions found incidentally on imaging studies.

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<sup>10</sup> Nonstandard abbreviations: PCC, pheochromocytoma; PGL, extraadrenal paraganglioma; PPGL, PCC and PGL; LC-MS/MS, liquid chromatography–tandem mass spectrometry; CT, computed tomography; MIGB, metaiodobenzylguanidine; FDG, fluorodeoxyglucose; PET, positron emission tomography; CVD, cyclophosphamide, vincristine, and dacarbazine.



**Karel Pacak:** PPGLs are very rare neuroendocrine tumors; their prevalence is estimated to be around 0.05% in the general population. Since about 50% of these tumors are diagnosed only at autopsy, the prevalence of these tumors could be higher, perhaps even reaching 0.1%. The prevalence is higher in the population of patients with hypertension and in those families with a risk for developing these tumors (e.g., carriers of a particular gene mutation).

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**Eamonn R. Maher:** We don't have any specific prevalence data for our local population. However, in terms of the prevalence of inherited susceptibility to PPGL in individual populations, it is important to consider that the presence of founder mutations, e.g., the "Black Forest" mutation in the von Hippel-Lindau tumor suppressor, E3 ubiquitin protein ligase (*VHL*)<sup>11</sup> gene (p.Tyr98His) that is common in southwestern Germany and is associated with a high risk of PCC, may cause geographic variations in the frequency of specific inherited forms of PPGL.

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**William F. Young:** Catecholamine-secreting tumors are rare, with an annual incidence of 2–8 cases per million people. Based on screening studies for secondary causes of hypertension in outpatients, the prevalence of PCC has been estimated at 0.1% to 0.6%. Nevertheless, it is impor-

tant to suspect, confirm, localize, and resect these tumors because (a) the associated hypertension is curable with surgical removal of the tumor, (b) a risk of lethal paroxysm exists, (c) at least 10% of the tumors are malignant, and (d) approximately 20% are familial, and detection of this tumor in the proband may result in early diagnosis in other family members.

Case detection testing for these rare neoplasms is indicated in clinical settings where the prevalence is increased, and these include: hyperadrenergic spells (e.g., episodes of palpitations, diaphoresis, headache, tremor, pallor); treatment-resistant hypertension; a familial syndrome that predisposes to PCC or paraganglioma (e.g., multiple endocrine neoplasia type 2, neurofibromatosis type 1, von Hippel Lindau syndrome, or succinate dehydrogenase mutations); a family history of PCC; an incidentally discovered adrenal mass; pressor response to anesthesia, surgery, or angiography; onset of hypertension at a young age (<20 years); idiopathic dilated cardiomyopathy; and a history of gastrointestinal stromal tumors or pulmonary chondromas.



**Ronald R. de Krijger:** I am not aware of the prevalence of PPGL in the Dutch population (17 million inhabitants) or worldwide. In the Netherlands I estimate that there is an annual incidence of 0.5–1.0 per 100 000 for PCC, and about one-tenth of this is for abdominal PGL. For

head and neck PGL, the annual incidence is probably in the order of 0.2–0.3 per 100 000. In populations with founder mutations in certain genes, there is an increased risk. This is the case for head and neck PGL in the Netherlands, given the founder mutations in succinate dehydrogenase complex, subunit D, integral membrane protein (*SDHD*).

***Plasma free and urinary fractionated metanephrines are regarded as the first-line tests in screening for PPGL. Do you feel that one of these tests is superior? Are there specific situations where one should be used over the other?***

**Graeme Eisenhofer:** To date there have been 4 studies directly comparing the diagnostic performance of

<sup>11</sup> Human genes: *VHL*, von Hippel-Lindau tumor suppressor, E3 ubiquitin protein ligase; *SDHD*, succinate dehydrogenase complex, subunit D, integral membrane protein; *MAX*, MYC associated factor X; *TMEM127*, transmembrane

protein 127; *SDHA*, succinate dehydrogenase complex, subunit A, flavo-protein (Fp).

plasma free vs urinary fractionated metanephrines, all consistently indicating higher diagnostic sensitivity and specificity of the plasma over the urine test. Nevertheless, all had limitations and the reported differences were small relative to those of each test compared to other tests of catecholamine excess. Therefore, until proven otherwise, either test remains suitable for first-line screening.

The plasma test is more suitable than the urine test in children and in patients with renal insufficiency. Some studies have suggested the same for populations at increased risk of PPGLs, but this is really a matter of reference intervals. At the upper cutoffs suitable for optimal diagnostic sensitivity for detecting tumors, diagnostic specificity is higher for the plasma than the urine test; therefore the plasma test may also be preferable in low-risk populations.

Of more importance to the choice of test is the method of measurement and the experience and expertise of clinicians and laboratory staff with each test. Urinary metanephrines measured by liquid chromatography–tandem mass spectrometry (LC-MS/MS) are, for example, preferable over measurements of plasma metanephrines by immunoassays, particularly when personnel are not experienced in the correct preparation of patients for blood collection.

**Karel Pacak:** Our experience at the NIH, based on a very large number of patients, suggests that plasma metanephrines are superior to urine metanephrines as the first biochemical test. It should be noted that these tumors produce catecholamines that are metabolized inside the tumor into free metanephrines, which are continuously released from the tumor tissue into the circulation. The assessment of metanephrines in urine includes the measurement of conjugated metanephrines (measured as free after their deconjugation). Conjugated metanephrines are also produced in different organs. Therefore, measurement of plasma free metanephrines provides a better diagnostic marker than urine-conjugated metanephrines in the biochemical diagnosis of these tumors. However, the proof of this in terms of practical utility has not yet been established.

**William F. Young:** At the Mayo Clinic, the most reliable case-detection strategy is measuring fractionated metanephrines and catecholamines in a 24-h urine collection. If clinical suspicion is high, then plasma fractionated metanephrines should also be measured. Some groups have advocated that plasma fractionated metanephrines should be a first-line test for PCC; the predictive value of a negative test is extremely high, and a normal plasma fractionated metanephrines result excludes PCC except in patients with early preclinical dis-

ease and those with strictly dopamine-secreting neoplasms. A plasma test is also attractive because of simplicity. Although measurement of plasma fractionated metanephrines has a diagnostic sensitivity of 96% to 100%, the diagnostic specificity is suboptimal at 85% to 89%; the diagnostic specificity falls to 77% in patients older than 60 years. It has been estimated that 97% of patients with hypertension seen in a tertiary care clinic who have plasma fractionated metanephrine measurements above the reference range will not have a PCC, resulting in excessive healthcare expenditures because of subsequent imaging and potentially inappropriate surgery. Thus, plasma fractionated metanephrines lack the necessary diagnostic specificity to be recommended as a first-line test, and this measurement should be reserved for cases for which the index of suspicion is high.

***In instances where plasma free or urinary fractionated metanephrines are slightly or modestly increased, what additional tests are useful in the biochemical workup of a potential case of PPGL?***

**Graeme Eisenhofer:** For borderline test results, it is important that follow-up tests have at least equal diagnostic sensitivity and, ideally, better diagnostic specificity than the initial screening test. In cases of borderline increases of urinary fractionated metanephrines, it is therefore appropriate to follow up with measurements of plasma metanephrines. For borderline increases of plasma normetanephrine, the clonidine suppression test, with measurements of normetanephrine before and 3 h after administration of the drug, provides an accurate method for distinguishing true- from false-positive results.

Usually, however, most false-positive results for plasma metanephrines simply reflect inadequate preparation of patients, easily resolved by repeating the blood collection after at least 30 min of supine rest, with the patients as comfortable as possible. A wait and retest approach to assess for further increases in values 6 months or more after initial testing provides another approach in more difficult-to-resolve cases.

**Karel Pacak:** About 20%–30% of patients with these tumors present with values that are equivocal (e.g., for plasma metanephrines below 4× above the upper reference limit), and an additional test is needed to confirm or rule out the presence of PPGL before any localization is initiated. The clonidine suppression test coupled with the measurement of plasma normetanephrine is the best test to use. The diagnostic sensitivity of this test is about 97%, with 100% diagnostic specificity. However, it should be noted that this test cannot be used for tumors secreting only metanephrine, but



because almost 99% of metanephrine is derived from the adrenal gland, the diagnosis and localization of these epinephrine-producing tumors is usually not a difficult task. I should also mention that in many patients increased metanephrine concentrations can be due to various drugs (including antihypertensives); therefore, any drug interference with biochemical results must be considered first.

**William F. Young:** The answer to this question is dependent on clinical context. If the clinical context is an incidentally discovered 2-cm vascular adrenal mass, PCC should be suspected even if measurements of fractionated metanephrines and catecholamines are normal. All PPGLs are “prebiochemical” in their early stages. Whereas, if the clinical context is a patient with marked paroxysms, then if a PCC is responsible for the paroxysms, the increases in the fractionated metanephrines and catecholamines should be similarly impressive; in this clinical setting, minimal increases in fractionated metanephrines and catecholamines are not consistent with PCC.

**What imaging modalities are used in your institution to localize PPGL?**

**Graeme Eisenhofer:** At Dresden, as at most centers, we primarily use computed tomography (CT) for initial localization, with MRI also available as called for. In most cases, we also employ  $^{123}\text{I}$ -metaiodobenzylguanidine (MIBG) scintigraphy, but have additional availability of  $^{18}\text{F}$ -fluorodeoxyglucose (FDG) and  $^{68}\text{Ga}$ -DOTATATE for positron emission tomography (PET)/CT.

**Karel Pacak:** Anatomic imaging studies, either CT or MRI, are used for the initial attempts to locate a PPGL. The preference of using anatomical imaging studies over functional imaging is that surgical procedures are rarely performed without good anatomical localization of a tumor. At the NIH, we prefer CT over MRI. However, MRI should be used in pregnant women, children and those with an allergy to contrast dye, and in situations in which radiation exposure needs to be minimized. A CT of the abdomen should be performed first, since most of these tumors are located in that area. Functional imaging studies, including FDG PET,  $^{18}\text{F}$ -fluorodopamine, or  $^{18}\text{F}$ -fluorodopa PET, or  $^{123}\text{I}$ -MIBG scintigraphy, are also used in the localization of these tumors, for several reasons. First,  $^{18}\text{F}$ -fluorodopamine PET and  $^{123}\text{I}$ -MIBG scintigraphy, for example, are PPGL-specific imaging modalities that confirm a tumor with almost 100% diagnostic specificity in a patient with biochemical proof of a tumor. Second, there is a genotype-specific imaging phenotype

that is very valuable in the assessment of these tumors, including their metastatic lesions. Thus, for *SDHB*-related PPGLs, FDG PET is the most sensitive, for head and neck PPGLs  $^{18}\text{F}$ -fluorodopa PET should be used, and for primary or metastatic tumors in general (e.g., when the genetics of these tumors is unknown),  $^{18}\text{F}$ -fluorodopamine is the preferred functional imaging modality. In the future, a cost-effective approach for tumor-specific functional imaging modalities needs to be further established, especially the role of  $^{18}\text{F}$ -fluorodopa in the evaluation of metastatic PPGLs or the use of functional imaging in newly discovered PPGLs associated with the MYC associated factor X (*MAX*) and transmembrane protein 127 (*TMEM127*).

**Eamonn R. Maher:** Our standard screening modality to detect PPGL in individuals at increased genetic risk is MRI scanning. For MRI-detected abnormalities that require further investigation, CT scanning or MIBG might be performed.

**William F. Young:** Localization studies should not be initiated until biochemical studies have confirmed the diagnosis of a catecholamine-secreting tumor. Computer-assisted imaging of the adrenal glands and abdomen with CT or MRI should be the first localization test. Approximately 85% of these tumors are found in the adrenal glands, and 95% are found in the abdomen and pelvis. The most common locations of catecholamine-secreting paragangliomas (in order of prevalence) include the superior abdominal paraaortic region, inferior abdominal paraaortic region, urinary bladder, thorax, skull base and neck, and pelvis.

CT with contrast of the abdomen and pelvis is our first localization test. If a PPGL is not detected with this study, the clinician should reassess the diagnosis. For example, did the clinician overlook treatment with a tricyclic antidepressant (the most common cause of false-positive biochemical testing)? If the biochemical diagnosis is secure and the CT of the abdomen and pelvis is negative, we would proceed to  $^{123}\text{I}$ -MIBG scintigraphy, which has a diagnostic sensitivity of approximately 80% and a diagnostic specificity of 99%. Localizing procedures that also can be used, but are rarely required, include computer-assisted imaging of the chest, neck, and skull base. The mean size of a symptomatic PCC or paraganglioma is 4.5 cm; they are not hard to find.

**Are there effective means to differentiate between benign and malignant PPGLs?**

**Graeme Eisenhofer:** As yet there is no reliable histopathological method to distinguish benign from malignant PPGLs. The only accepted method to diagnose

malignancy remains demonstration of metastatic lesions; this, however, does not mean that the absence of metastases denotes a benign classification since such lesions often only become apparent many years after surgical resection. Thus, until there is a reliable method for predicting malignancy, no PPGL should ever be classified as benign.

Despite the above shortcomings in the identification of malignant PPGLs, there are numerous known risk factors for metastatic disease. Tumors with an extraadrenal location have a 3.4-fold higher risk of malignancy than those with an adrenal location. Large size is also a risk factor and together with extraadrenal location accounts for the high risk of malignancy associated with mutations of the *SDHB* gene.

High concentrations of plasma free methoxytyramine, the metabolite of dopamine, also look to provide a promising new biomarker of metastatic PPGLs, with recent evidence suggesting that when accurately determined by LC-MS/MS these measurements can detect over 80% of patients with metastatic disease at a diagnostic specificity of over 90%.

**Karel Pacak:** At present there are no effective methods, including histopathological examination, to differentiate between benign and malignant PPGLs. Carboxypeptidase E is a promising marker, but its role must be established on a large series of PPGLs, and it may be useful only in particular PPGLs (e.g., *SDH*-related PPGLs). However, on the basis of previous and recent observations and large clinical studies, it is clear that patients presenting with *SDHB*-related PPGLs, with primary tumors over 5 cm in size, and with increased plasma methoxytyramine, have a much higher risk for developing metastatic disease.

**Eamonn R. Maher:** In the absence of distant metastases, the presence of a germline *SDHB* mutation significantly increases the prior risk of malignancy but cannot definitively inform whether an individual PPGL is malignant or not. As Dr. Eisenhofer has stated above, plasma methoxytyramine may be a useful predictor of the likelihood of metastatic spread.

**William F. Young:** Distinguishing between benign and malignant catecholamine-secreting tumors is difficult on the basis of clinical, biochemical, or histopathologic characteristics. The diagnosis of malignant PPGL requires finding this tumor in sites that do not normally contain chromaffin tissues (e.g., liver, bone, lung, omentum, or lymph nodes). Malignancy is rare in patients with multiple endocrine neoplasia type 2 or von Hippel Lindau syndrome but is common in those with familial paraganglioma caused by mutations in *SDHB*. Patients with *SDHB* mutations are more likely to de-

velop malignant disease and nonparaganglioma neoplasms (e.g., renal cell carcinoma). Although the 5-year survival rate for patients with malignant PCC is <50%, the prognosis is variable; approximately 50% of patients have an indolent form of the disease, with a life expectancy of more than 20 years, and the other 50% of patients have rapidly progressive disease, with death occurring within 1–3 years after diagnosis.

**Ronald R. de Krijger:** This is a very difficult issue. The short answer is still no. When there is obvious metastasis or ingrowth in surrounding structures detected with radiology or nuclear imaging or at surgery, one can confidently make a diagnosis of malignancy, especially if these findings are further supported by histological results. However, this is rarely the case. The vast majority of PPGL present as a single lesion in an organ with no further evidence of disease. Histological criteria have been shown to be of little help in assessing the future behavior of endocrine tumors in general. Likewise, MIB-1 labeling for identification of the proliferative fraction could not sufficiently discriminate non-metastasizing from metastasizing PCC and PGL. The PASS (Pheochromocytoma of the Adrenal Gland Scaled Score) was proposed in 2002 but was subsequently shown to suffer from high interobserver variability. No immunohistochemical or molecular markers with sufficient diagnostic sensitivity and specificity have been proposed so far.

#### **What are the treatment options for an individual with a diagnosis of PPGL?**

**Graeme Eisenhofer:** Surgical resection after appropriate preoperative preparation provides an effective cure for most patients, but in up to a quarter of patients there may be subsequent recurrent or metastatic disease. Therefore, postsurgical periodic screening is called for in all patients.

In patients with metastatic disease there is as yet no effective cure. Radiotherapy with  $^{131}\text{I}$ -MIBG is most commonly used, but is effective only in occasional patients. There are several other palliative or experimental treatment options. As yet, none have demonstrated effectiveness. Combination or personalized therapies that target specific pathways according to the PPGL genetic subtype offer the best hope.

**Karel Pacak:** In all patients, a surgical approach, if feasible, is the first choice. For patients with metastatic disease, the options are limited. If patients have slowly progressing but extensive disease and are positive on  $^{123}\text{I}$ -MIBG scintigraphy, radiotherapy using  $^{131}\text{I}$ -MIBG is usually recommended. In patients with rapidly progressing disease, cyclophosphamide, vincris-

tine, and dacarbazine (CVD) chemotherapy is usually used. However, neither  $^{131}\text{I}$ -MIBG nor CVD chemotherapy result in a cure in most patients (rarely some cured patients were described), and only about one-third of patients will respond. Nevertheless, new results show that about 70%–80% of *SDHB*-related metastatic PPGLs respond to CVD chemotherapy. For *SDHB*-related metastatic PPGLs I do not recommend the use of Sunitinib or Affinitor; our experience did not show any response to these chemotherapeutics in these patients. Combined mTOR1 and 2 (mammalian target of rapamycin 1 and 2) inhibitors, hypoxia-inducible factor (HIF), heat shock protein 90 (HSP90), AKT, or other inhibitors (or their combinations) are most likely to become somewhat successful treatments in these patients in the near future.

**William F. Young:** The treatment of choice for PCC and PGL is complete surgical resection. Surgical survival rates are 98% to 100% and are highly dependent on the skill of the endocrinologist–endocrine surgeon–anesthesiologist team. The most common adverse event following surgery is sustained hypertension. Careful preoperative pharmacologic preparation is crucial for successful treatment. Most catecholamine-secreting tumors are benign and can be totally excised. Tumor excision usually cures hypertension.

**What role should genetic testing play in the diagnosis/management of PPGL? Would you recommend widespread genetic testing for all patients with PPGL?**

**Graeme Eisenhofer:** Genetic testing is already having a substantial impact in the diagnosis and management of patients with PPGLs. Such patients and family members with identified mutations represent important groups who must be periodically screened for PPGLs and in whom the choice of specific tests, test interpretation, and management of disease, including other manifestations, should be individualized according to the affected gene.

As an example, periodic biochemical testing in patients with mutations of the *SDHB* gene should include measurements of plasma methoxytyramine, with test interpretation concentrating on this analyte and normetanephrine. The high risk of malignancy in these patients mandates careful management, with the anticipation that early detection and resection of tumors, when small, will reduce risk and rates of malignancy.

Despite the importance of genetics I do not recommend widespread testing until there are less expensive methods available for accurately testing panels of tumor susceptibility genes. In the meantime, genetic testing is best offered for specific genes in selected patients

for whom the family history or clinical presentation is consistent with a risk of a mutation for those genes.

**Karel Pacak:** As I described above, genetic testing is very crucial for the proper diagnosis, management, and therapeutic options of each patient. Gene-specific biochemical and imaging phenotypes have already been well described. Gene-specific therapeutic phenotypes are likely to be introduced in the very near future. However, this does not justify offering genetic screening to every patient. Those patients who do not have any family history of PPGLs, have a very small epinephrine-secreting PCC, and are 50 years old or older most likely have a sporadic tumor and do not need genetic testing, at least not initially. Any specific genetic testing must be guided by the presence of family history, the biochemical phenotype, the location of the tumor, the presence of metastatic disease or multiplicity, and the age at first diagnosis. When genetic testing becomes less expensive through the use of high-throughput methods, there is a good chance that genetic testing will be offered to all patients.

**Eamonn R. Maher:** I do think that, in the future, all PPGL patients will be offered genetic diagnosis to inform the risk, for them and their families, of further primary tumors and malignancy. In the past few years, the expanding number of PPGL genes and the high cost of gene testing by conventional (Sanger) sequencing has caused a shift from universal to targeted testing. However, though consideration of clinical features (family history, age at diagnosis, tumor location) and immunohistochemistry [e.g., *SDHB*, succinate dehydrogenase complex, subunit A, flavoprotein (Fp) (*SDHA*)] can provide more cost-effective targeting, I believe that universal testing is required to detect all patients harboring germline mutations. The advent of high-throughput second generation sequencing technologies enables multiple genes to be tested inexpensively, and we have developed a PPGL gene panel test that sequences 9 PPGL genes simultaneously and thus provides comprehensive and more rapid genetic testing for the approximate cost of analyzing a single gene by conventional testing methods. I anticipate that the availability of this and similar genetic testing strategies will result in an expansion of genetic testing.

**William F. Young:** Genetic testing should be considered if a patient has one or more of the following: (a) PGL; (b) bilateral adrenal PCC; (c) unilateral adrenal PCC and a family history of PCC/PGL; (d) unilateral adrenal PCC onset at a young age (<45 years); or (e) other clinical findings suggestive of one of the associated syndromic disorders. Clinicians can obtain a list of clinically approved molecular genetic diagnostic labo-

ratories (www.genetests.org). Given the considerable cost of genetic testing, the use of a stepwise approach based on each patient's clinical scenario is prudent.

An asymptomatic person known to be at risk for disease on the basis of family history of PCC/PGL should have genetic testing only if an affected family member has a known mutation. Genetic testing can be complex; testing one family member has implications for related individuals. Genetic counseling is recommended to help families understand the implications of genetic test results, to coordinate testing of at-risk individuals, and to help families work through the psychosocial issues that may arise before, during, and after the testing process. If mutation testing in a patient is positive, first-degree relatives (patient's parents, siblings, and children) should be offered genetic testing.

In addition, because some genetic causes of PPGLs have not yet been identified, all first-degree relatives of a patient with PCC or PGL should have biochemical testing (e.g., 24-h urine for fractionated metanephrines and catecholamines).

**Ronald R. de Krijger:** The current figures for the accumulated rate of germline mutations and other germline genetic abnormalities in PPGLs are between 30% and 40%. This is high enough to recommend genetic testing in any individual with PPGL. This testing should preferably be done in a stepwise manner, on the basis of the further clinical picture (location of the tumor, the presence of other lesions), a biochemical profile, and immunohistochemical testing for *SDHB* (and *SDHA*) if tumor tissue is available. Identification of a germline genetic abnormality will direct future follow-up in the index patient and allows further screening of family members.

***Do you anticipate that additional susceptibility genes for PPGL will be identified?***

**Graeme Eisenhofer:** Without doubt there are other PPGL susceptibility genes that will be identified. I also anticipate that the day will come when it will be both technically and economically feasible to offer efficient and accurate testing of all tumor susceptibility genes in all patients with PPGLs.

**Karel Pacak:** Yes, I do, especially genes that may be involved in the pathogenesis of malignant/metastatic or multiple PPGLs.

**Eamonn R. Maher:** Yes I do. There are a significant proportion (up to 30%) of familial cases and multiple tumor cases in which we cannot detect a germline in a known inherited PPGL gene. Although some of these cases might ultimately prove to have a mutation in a currently known gene that cannot be detected by standard mutation detection methods, I strongly suspect that further inherited PPGL genes will be identified in the next few years.

**William F. Young:** Yes. We have families at the Mayo Clinic with familial PCC and PGL who do not have germline mutations in any of currently known susceptibility genes.

**Ronald R. de Krijger:** Given the number of genes identified so far, and the fact that 2 further susceptibility genes have recently been identified, I anticipate that additional genes will be found. There are still familial cases in which there seem to be no abnormalities in the known genes. Thus, there appears to be room for other genes, potentially in pathways in which known genes play a role.

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