Predictors and Prevalence of Paraganglioma Syndrome Associated With Mutations of the SDHC Gene

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Context Paraganglioma syndrome includes inherited head and neck paragangliomas (HNPs) and adrenal or extra-adrenal pheochromocytomas and are classified according to the susceptibility genes SDHB, SDHC, and SDHD. In contrast with those with germline mutations of the SDHB and SDHD genes, clinical and genetic data on patients with mutations of SDHC are scarce.

Objective To determine the prevalence and clinical characteristics of SDHC mutation carriers compared with patients with SDHB and SDHD mutations and with sporadic cases.

Design, Setting, and Patients Genetic screening for SDHC mutations in an international HNP registry of 121 unrelated index cases and in 371 sporadic cases from a pheochromocytoma registry, conducted January 1, 2001, until December 31, 2004. Identified index cases and affected relatives were clinically evaluated.

Main Outcome Measures Prevalence of and clinical findings for SDHC mutation–associated HNPs vs those with SDHB and SDHD mutations.

Results The prevalence of SDHC carriers was 4% in HNP but 0% in pheochromocytoma index cases. None of the SDHC mutation carriers had signs of pheochromocytoma. We compared HNPs in 22 SDHC mutation carriers with the HNPs of SDHB (n=15) and SDHD (n=42) mutation carriers and with 90 patients with sporadic HNPs. Location, number of tumors, malignancy, and age were different: more carotid body tumors were found in SDHC (13/22 [59%]) than in sporadic HNPs (29/90 [32%], P=.03), as well as fewer instances of multiple tumors in SDHC (2/22; P<.001), 0 malignant tumors in SDHC vs 6/15 in SDHB (P=.002), and younger age at diagnosis in SDHC than in sporadic HNPs (45 vs 52 years; P=.03).

Conclusions Patients with HNP, but not those with pheochromocytoma, harbor SDHC mutations in addition to those in SDHB and SDHD. In total, more than one quarter of HNP patients carry a mutation in 1 of these 3 genes. Head and neck paragangliomas associated with SDHC mutations are virtually exclusively benign and seldom multifocal. Analysis for germline mutations of SDHC is recommended in apparently sporadic HNP to identify risk of inheritance.

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PARAGANGLIOMA SYNDROME (PGL) is a clinical term that has been introduced to describe a group of diseases in which patients may have neoplasias of several paraganglia. For at least 4 decades, it has been known that such conditions may be heritable. In addition, thoracic, retroperitoneal, and adrenal locations (eg, extra-adrenal or adrenal pheochromocytomas) are also well-recognized components of PGL. Thus,
Table 1. Primers Used in the Study

<table>
<thead>
<tr>
<th>Exon</th>
<th>Primer Sequence*</th>
<th>PCR Conditions†</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>F: CAC ATG ACA GCC CCA ACC CC&lt;br&gt; R: CTG CCC AGG CAC AGG ATA AAC A</td>
<td>65°C/15 s</td>
</tr>
<tr>
<td>2</td>
<td>F: TAG TTT TAA TCT ATC CCT TCA C&lt;br&gt; R: TCT CCA GAC TTA GAA ACT TA</td>
<td>55°C/30 s</td>
</tr>
<tr>
<td>3</td>
<td>F: AGC TTA TGC AAA ATA TTA AAC CAA GT&lt;br&gt; R: AGG TTC TGG TGT GGC TCC A</td>
<td>55°C/15 s</td>
</tr>
<tr>
<td>4</td>
<td>F: GGT TAT ATT TTT GGC AAG ATA GAC TC&lt;br&gt; R: CCA AGT TTT TCA AAG AAC AGT A</td>
<td>60°C/30 s</td>
</tr>
<tr>
<td>5</td>
<td>F: TCA TAT TAG TTT TAA CTT ATG AGC AGC&lt;br&gt; R: CTG CCC ACT CCG TTC ACA G</td>
<td>58°C/15 s</td>
</tr>
<tr>
<td>6</td>
<td>F: TGT TAA TGT COT ATT TAG TCA A&lt;br&gt; R: TAA ACA AAT AAC GAG AAC TTC</td>
<td>55°C/30 s</td>
</tr>
</tbody>
</table>

*Description of the 6 forward (F) and the 6 corresponding reverse (R) primer sequences of the SDHC gene.
†The polymerase chain reaction (PCR) amplification was performed for 30 cycles. Different annealing temperatures (65°C, 60°C, 58°C, and 55°C) and different annealing periods (15 or 30 seconds) were used.

patients may initially present with a local mass of the neck or other paraganglial sites with or without general symptoms of hormone release demonstrating functional or nonfunctional tumors.

Paraganglioma syndrome has been classified genetically into 4 entities, PGL1, PGL2, PGL3, and PGL4. To date, 3 of these 4 entities have been associated with germline mutations in the genes encoding 3 of the subunits of succinate dehydrogenase (SDH), which has a key function in the Krebs cycle and the respiratory chain. Germline mutations in SDHB, SDHc, and SDHD have been found in PGL4, PGL3, and PGL1, respectively. In contrast, mutations of SDHA have not, to date, been associated with paragangliomas (Sarah McWhinney, BA, and C.E., unpublished data, December 2004); instead, germline homozygous mutation in this gene is associated with Leigh syndrome, characterized by severe neurodegeneration. So far, the gene for PGL2 has not been identified.

When germline SDHC and SDHD mutations were first identified in families with several members who had head and neck paragangliomas (HNPs), it was initially believed that SDH mutations were perhaps associated primarily with PGL syndromes presenting with HNPs. Subsequently, however, germline SDHD mutations were identified in individuals and families with pheochromocytoma as well. SDHB mutations were then found in patients with HNPs and pheochromocytoma. Rarely, HNPs have also been observed as an inherited condition in multiple endocrine neoplasia type 2 and von Hippel–Lindau disease, for which the susceptibility genes are RET and VHL, respectively.

It is of great clinical relevance to determine if any particular features of PGL are associated with a particular gene mutation and if the presence of a mutation in a particular gene could predict the characteristics of PGL neoplasia.

METHODS

Registries

The Head and Neck Paraganglioma Registry was founded in 2000 based on ascertainment of patients with HNP through Germany; all 150 German departments of otorhinolaryngology were contacted and most participated. Subsequently, we extended the registry to cases in Italy, Poland, Spain, France, Finland, and Switzerland; unlike in Germany, these registra-

sis.) To determine if the characteristics of the population-based German registry were different from those from the non–population-based cases outside of Germany, we compared their demographic and tumor characteristics and their mutation frequencies. There was no statistical difference between demographic and tumor features between the 2 groups (P = .10-.99), nor was there any difference in mutation frequencies (2/88 vs 2/33; P = .13 by 2-tailed Fisher exact test).

Only index cases (ie, patients not related to one another) are listed in the registries. No patient entered both registries, since those with both HNPs and pheochromocytomas were counted only for the first symptomatic lesion. Regarding tumor biology, we defined a malignant HNP as a tumor with distant metastases. One important inclusion criterion for this study was that blood DNA had to be available. Patients with clinical signs of neurofibromatosis type 1 were excluded.

Molecular Genetic Analyses

For each patient, genomic DNA was extracted from 10 mL of EDTA-anticoagulated blood using standard methods. Polymerase chain reaction amplification was performed for all 6 exons of the SDHC gene using primers and conditions as shown in Table 1 and as reported by others. All samples were analyzed by single-strand conformational polymorphism and denaturing high-performance liquid chromatography. Exons showing single-strand conformational polymorphism or denaturing high-performance liquid chromatography variations were sequenced. Purified polymerase chain reaction products were directly sequenced in both directions using the amplification primers and the dyeoxy chain termination method (DYEnamic ET Kit, Amersham Biosciences, Freiburg, Germany) on a MegaBACE 500 sequencer (Amersham Biosciences). In parallel, all patients underwent germline analysis for the genes RET, VHL, SDHB, and SDHD using methods as previously published.
As controls, we used 100 unpaid healthy blood donor volunteers of both sexes from Freiburg and Warsaw. Samples were stripped of identifiers and made anonymous.

Investigation Program for Carriers of an SDHC Mutation and Their Relatives
Once a germline SDHC mutation was detected, we invited the patient for clinical reevaluation. The investigation program included magnetic resonance imaging of the neck and skull base, the thorax, and the abdomen as well as 24-hour urine assay for norepinephrine, epinephrine, and vanillylmandelic acid levels. In addition, we offered all first-degree relatives testing for the mutation found in the index case. Relatives underwent testing on a volunteer basis. If positive results were found, further clinical evaluation was offered. This strategy of molecular and clinical investigation is similar to that described previously.16

Literature Review
To increase our sample size of individuals with an SDHC mutation for clinical correlates, we performed a literature review using PubMed and the Medical Subject Heading terms SDHC and paraganglioma and noted the presence or absence of description of features such as exact tumor location, age at diagnosis, number of tumors, and tumor malignancy. We also contacted the principal authors of these articles, several of whom are authors on the present article. Head and neck paraganglioma–positive relatives of index cases reported by others in the literature were included in our clinical-molecular correlative studies.

Clinical-Molecular Correlative Studies and Statistical Analysis
We sought to determine the similarities and differences of clinical features among individuals carrying SDHC mutations, those with SDHB and SDHD mutations, and those without germline mutations. For these purposes, the series of SDHC mutation carriers comprised those detected after screening the 2 registries for mutations in this gene, mutation-positive relatives of these index cases, and SDHC mutation carriers found in the literature and their mutation-positive relatives. The relevant features, such as exact tumor location, age at diagnosis, number of tumors, and malignancy, were compared with the features of those with SDHB and SDHD mutations2,16 and those with sporadic tumors, the latter of which was defined as those without SDHC, SDHB, SDHD, RET, or VHL mutations.

For statistical analyses, a 2-tailed Fisher exact test was used for 2 × 2 contingency tables. Based on an α level of .05 and the respective numbers of cases given in Table 2, statistical power for detection of a 25% vs 50% relative prevalence in tumor location was roughly 60% between the SDHC and sporadic groups and was roughly 50% and 30% when SDHC was tested vs SDHD and SDHB, respectively. For larger contingency tables, a generalized Fisher test17 was used. Age differences were tested for significance with the Mann-Whitney U test (pairwise) and the Kruskal-Wallis test (comparison of all groups). All tests for statistical significance were implemented with Mathematica, version 5 (Wolfram Research Inc, Champaign, Ill); statistical power was calculated with Sample Size Web-based software (Massachusetts General Hospital Biostatistics Center, Boston; available at http://hedwig.mgh.harvard.edu/biostatistics).

The project was approved by the ethics committees of the respective institutions. All participants provided oral (17%) or written (83%) consent in accordance with the accepted standards for each respective country. The study was conducted from January 1, 2001, until December 31, 2004.

RESULTS
As of December 31, 2004, the Head and Neck Paraganglioma Registry consisted of 121 unrelated index cases who presented with HNP (Table 3). At this time, 2 (2%) of 93 German and 3 (11%);
P(=.81) of 28 non-German HNP registrants were found to have a family history of autonomic nervous system tumors. Fifteen percent of the German and 18% of the non-German patients had multiple tumors (P = .77).

None of the 121 index cases with HNP showed germline mutations of the genes RET or VHL and none had signs of neurofibromatosis type 1. Five cases had a mutation of the SDHC gene (TABLE 4). Eight (7%) had mutations of the SDHB gene and 20 (17%) had mutations of the SDHD gene. Finally, 88 cases without any mutations in these 5 genes were classified as having sporadic HNP. Thus, prevalence of SDHC was 4% (5/121 cases). The SDHC mutations are located in exons 1, 2, 4, 5, and 6 and are summarized in Table 4. Two of the mutations are stop codon mutations, 1 mutation leads to a start codon change, 1 mutation causes a splice site alteration, and 1 mutation is a missense mutation, which did not occur in the 100 healthy controls. Clinically, 1 case had a carotid body tumor and 4 cases had jugular paragangliomas (FIGURE). No patient had multiple tumors, and family history of paragangliomas was positive in only 1 case. None of the tumors was a malignant paraganglioma, but among the 4 cases with jugular paragangliomas, all had permanent palsy of at least 1 cranial nerve.

From the Pheochromocytoma Registry, as a first step we removed all index cases with mutations of the RET gene or the VHL gene or clinical signs of neurofibromatosis type 1. The remaining 371 index cases were all tested for SDHC mutations, but finally had such a mutation, whereas 21 cases had an SDHB mutation and 21 cases had an SDHD mutation.\(^2,16\)

The key features of the 5 index cases with mutations of the SDHC gene and their relatives who tested positive for mutations are summarized in TABLE 5. Among 27 living first-degree relatives, 23 had been genetically tested and 10 of those were carriers. A major result was that none of the adequately investigated carriers of an SDHC mutation had an abdominal or thoracic pheochromocytoma. Of those who had no imaging of the abdomen or thorax and no evaluation of catecholamine release, no signs or symptoms of such tumors have been reported.

We then sought to determine if genespecific clinical, pathological, or demographic features differentiate those with SDHC mutations from those with SDHB and SDHD mutations and from those with sporadic HNP. We achieved this by systematically collecting the clinical, pathological, and demographic data, as described in detail previously.\(^2,16\) A combined total of 22 patients with HNP and with SDHC mutations from this series and from the literature, all updated for this report (Tables 2 and 5), were compared with the sporadic cases (n = 90) identified in this study, as well as with those with SDHB mutations (15 cases of HNP), and those with SDHD mutations (42 cases of HNP). Only 6 cases completed the full screening, including magnetic resonance imaging of the neck, thorax, and abdomen, although clinical examination was offered to all carriers at least once. This type of uptake of offered comprehensive clinical surveillance in mutation carriers is, in our experience, no different than in those with RET or VHL mutations. Overall, clinical differences among the 4 groups, SDHC mutation carriers, SDHD mutation carriers, SDHB mutation carriers, and sporadic HNP cases, were found in tumor location (P<.001 to P = .16), multifocality of tumors (P<.001), and frequency of malignant disease (P<.001) (Table 2). Of note, those with SDHC mutations do not have

### Table 3. Cases of HNP and Their SDHB, SDHC, and SDHD Gene Mutations

<table>
<thead>
<tr>
<th>Index cases identified</th>
<th>SDHB</th>
<th>SDHC</th>
<th>SDHD</th>
<th>Sporadic</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Relatives of index cases identified</td>
<td>2</td>
<td>2</td>
<td>12</td>
<td>0</td>
<td>16</td>
</tr>
<tr>
<td>Cases found in the literature and in Lille, Freiburg, and Pittsburgh</td>
<td>5</td>
<td>15</td>
<td>10</td>
<td>2*</td>
<td>32</td>
</tr>
<tr>
<td>Total</td>
<td>15</td>
<td>22</td>
<td>42</td>
<td>90</td>
<td>169</td>
</tr>
</tbody>
</table>

Abbreviation: HNP, head and neck paraganglioma.

*These 2 patients primarily had abdominal pheochromocytomas and were also found to have HNPs during follow-up, but no mutations in SDHB, SDHC, or SDHD have been detected.

### Table 4. Germline Mutations of the SDHC Gene in Index Cases and Literature Review Cases*

<table>
<thead>
<tr>
<th>Index Case</th>
<th>Nationality</th>
<th>Exon</th>
<th>Mutation (cDNA Nucleotide)</th>
<th>Consequence (Codon and Amino Acid)</th>
<th>No. of Carriers</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Spanish</td>
<td>1</td>
<td>1 A/G</td>
<td>M11/loss of the start codon</td>
<td>4</td>
<td>Current report</td>
</tr>
<tr>
<td>2</td>
<td>German</td>
<td>2</td>
<td>39 C/A</td>
<td>C13X</td>
<td>1</td>
<td>Current report</td>
</tr>
<tr>
<td>3</td>
<td>Polish</td>
<td>4</td>
<td>214 C/T</td>
<td>R72C</td>
<td>4</td>
<td>Current report</td>
</tr>
<tr>
<td>4</td>
<td>Italian</td>
<td>6</td>
<td>439 C/T</td>
<td>Q147X</td>
<td>4</td>
<td>Current report</td>
</tr>
<tr>
<td>5</td>
<td>German</td>
<td>5</td>
<td>405 + 1 G/T</td>
<td>Splice site alteration</td>
<td>2</td>
<td>Current report</td>
</tr>
<tr>
<td>6</td>
<td>German</td>
<td>1</td>
<td>3 G/A</td>
<td>M11/loss of the start codon</td>
<td>7</td>
<td>Niemann and Müller,(^*) 2000</td>
</tr>
<tr>
<td>7</td>
<td>German</td>
<td>5</td>
<td>IVS5 + 1 G/T</td>
<td>Splice site alteration</td>
<td>1</td>
<td>Niemann et al,(^*) 2003</td>
</tr>
<tr>
<td>8</td>
<td>French</td>
<td>6</td>
<td>473 T/C</td>
<td>L158P</td>
<td>2</td>
<td>Bauters et al,(^*) 2003</td>
</tr>
<tr>
<td>9†</td>
<td>American</td>
<td>6</td>
<td>8372 base-pair deletion spanning exon 6</td>
<td>Loss of exon 6</td>
<td>6</td>
<td>Baysal et al,(^*) 2004</td>
</tr>
</tbody>
</table>

*For all index cases, the initial tumor was a head and neck paraganglioma.
†Case 10 (see Table 5) had an identical mutation to case 5, possibly related but not proven.
thoracic or abdominal pheochromocytoma or paraganglioma, in contrast with those with SDHB and SDHD mutations, as shown previously. The carotid body was the favored location of HNP in all subgroups. Tympanic paragangliomas are rare and have never been seen in SDHB mutation carriers. Multiple tumors were more frequently associated with SDHD mutation carriers than with those with SDHC mutations (24/42 vs 2/22; P < .001). The most striking feature, however, is that the pertinent clinical features of the HNP found in patients with SDHC mutations are similar to those in sporadic HNP. No malignant tumors were found in both groups; roughly 10% in both groups had multiple tumors (2/22 vs 10/90; P = .30). Carotid body tumors seemed to occur more frequently in SDHC mutation carriers than in sporadic HNP (13/22 vs 29/90; P = .03), but no significant difference in the distribution of tumors between SDHC and sporadic HNP mutations could be detected for the other tumor locations (jugular, 6/22 vs 33/90 [P = .46]; tympanic, 2/22 vs 21/90 [P = .24]; vagal, 2/22 vs 3/90 [P = .25]). The median age at diagnosis was 36 years (range, 13-67 years) among SDHC mutation carriers, 39 years (range, 21-66 years) among SDHB mutation carriers, and 46 years (range, 13-73 years) among SDHD mutation carriers, all significantly different from the median age at diagnosis of sporadic HNP (53 years; range, 15-83 years; P < .001, P = .01, and P = .03, respectively). This indicates a genotype dependence of the age-related penetrance, which confirms published data for SDHD and SDHB. However, also regarding this parameter, SDHC mutations are more similar to sporadic HNP than to SDHB or SDHD mutations. Because of multiple comparisons, it may be sensible to avoid possible spurious significances by accepting an α level of .01 or lower instead of .05 as done herein (Bonferroni correction). This strengthens the statement that the available data do not allow a clinical or statistical distinction between SDHC-related disease and sporadic paraganglioma.

COMMENT
Paraganglioma syndrome can include heritable disorders, and only recently have its susceptibility genes been identified. The syndromes associated with germline mutations of the SDHD gene (PGL1) and the SDHB gene (PGL4) are relatively well characterized in regard to mutation spectra and associated clinical features. Both can initially present with HNPs or adrenal and extra-adrenal pheochromocytomas, including those in a thoracic location. For PGL1 and PGL4, more than 20 different mutations each have been described. In contrast, little is known about the true population-based prevalence of SDHC mutations for any given PGL presentation or the clinical features found in patients with these mutations. Our current comprehensive clinicogenetic study of SDHC has revealed that the prevalence of germline SDHC mutations is 4% (5/121) in the population-based International HNP Registry (Table 4), but no SDHC mutations were found among 371 unrelated cases symptomatically presenting with thoracic or abdominal extra-adrenal or adrenal pheochromocytomas from the population-based Pheochromocytoma Registry. These observations contrast with those for SDHB and SDHD mutation carriers, who can present either with HNP or with thoracic/abdominal (including adrenal) pheochromocytomas.

All SDHC mutations known at present are summarized in Table 4. They are distributed over the entire gene except on exon 3. They comprise loss of the start codon, missense mutations (substitution of one amino acid by another), or mutations causing truncation of the putative SDHC protein due to a stop codon, a splice site alteration, or a large deletion. Not all mutations identified by this study have been described. The number of different mutations of the SDHC gene is only one third as many as those known both for SDHB and for SDHD.

To study the prevalence of germline SDHC mutations and clinical characteristics of SDHC mutation carriers, we have created and enlarged the International HNP Registry. The registry, to which systematic accrual initially occurred from Germany on a population basis, was extended to other countries and currently comprises 121 symptomatically unrelated (index) cases. The prevalence of SDHC mutations was 4% in the 121 index cases (among the German population, 2%; P = .08). Evaluation of these 121 patients for mutations in SDHB and SDHD revealed frequencies of 7% and 17%, respectively. These frequencies were not different between the German registrants and others (in the German population, mutation frequencies were 6% and 13%). This contrasts with mutation frequencies for SDHB, SDHC, and SDHD in 3 different series of 98 HNP cases, all of which were not population-based. Their overall mutation frequencies range from 12% to 41%. Mutation frequencies for SDHB range from 0% to 9% for sporadic presentations and from 9% to 33% for familial presentations; SDHD mutation frequencies range from 0% to 9% for sporadic and 33% to 82% for familial presentations. Of note, no SDHC mutations were found in these 3 series. It is difficult to precisely determine why SDHC mutations were absent from the previous 3 series of HNP, which were tested for this gene. Notably, a whole exon deletion was the cause of the initial failure to find the SDHC mutation among clinic patients ascertained in the United States. One ob-
observation is that to date, all but 1 SDHC mutation carrier (Table 4) were found in Western Europe. Why the mutation frequencies for SDHB and SDHD mutations contrast with our population-based registry is likely explained by the referral-based and hospital-based nature of the other 3 series. Indeed, the most selective series, based in 2 tertiary referral centers in the United States, has the highest frequencies of mutations.

Our current study provides evidence that carriers of SDHC mutations have a different profile compared with carriers of SDHB and SDHD mutations regarding age of disease onset, abdominal lesions, multifocal tumors, and tendency toward malignant transformation of tumors. Of interest, we could not find demographic, clinical, or pathological features that clearly differentiate between sporadic and SDHC-associated HNP. Importantly, 4 of 5 index cases (all with jugular tumors) carrying an SDHC mutation had an outcome dominated by permanent palsy of at least 1 cranial nerve. The only chance to ameliorate the prognosis is to identify carriers in an asymptomatic stage in order to operate on the tumors before development of health risks. Thus, to help differentiate SDHC-related HNP from sporadic HNP, SDHC gene testing may be clinically useful. However, it should be noted that carotid body HNPs are common in both SDHD- and SDHC-related HNP, so that such presentations may trigger SDHD followed by SDHC genetic testing. On the other hand, glomus vagale HNP presentations should signal the clinical cancer geneticist to

<p>| Table 5. Clinical Characteristics of SDHC Gene Mutation Carriers Found in This Study and in a Literature Review |
|---------------------------------|----------------------|-----------------|----------|----------|----------|----------|----------|----------|</p>
<table>
<thead>
<tr>
<th>Index Case and Relatives</th>
<th>Nationality</th>
<th>Sex</th>
<th>Age at Onset, y</th>
<th>Tumor Location</th>
<th>Malignancy</th>
<th>No. of Tumors</th>
<th>CT/MRI Results</th>
<th>Catecholamine Excretion Test Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Index case 1 Spanish F 54</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Normal</td>
</tr>
<tr>
<td>Daughter Spanish F 29</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sister Spanish F 29</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nephew Spanish M 29</td>
<td>Vagal</td>
<td>Benign</td>
<td>3</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Normal</td>
</tr>
<tr>
<td>Index case 2 German F 38</td>
<td>Jugular</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td>Index case 3 Polish M 59</td>
<td>Jugular</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td>Brother Polish M 59</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Niece Polish M 36</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Brother Polish M 50</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Index case 4 Italian M 49</td>
<td>Jugular</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td>Mother Italian F 73</td>
<td>Carotid body</td>
<td>Benign</td>
<td></td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Normal</td>
</tr>
<tr>
<td>Brother Italian M 47</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sister Italian F 37</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Normal</td>
</tr>
<tr>
<td>Index case 5 German M 38</td>
<td>Jugular</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td>Father German M 63</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Normal</td>
</tr>
<tr>
<td>Index case 6 German F 58</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Son German M 46</td>
<td>Jugular</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Daughter German M 46</td>
<td>Jugular</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Daughter German M 49</td>
<td>Tympanic + carotid body</td>
<td>Benign</td>
<td>2</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Niece German F 49</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Son German M 65</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Niece German M 71</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Index case 7 German F 31</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Positive‡</td>
</tr>
<tr>
<td>Index case 8 French M 22</td>
<td>Jugular</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>Negative</td>
<td>Negative</td>
<td>Noradrenaline elevated‡</td>
<td></td>
</tr>
<tr>
<td>Mother Algerian F 63</td>
<td>Unknown</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Positive</td>
</tr>
<tr>
<td>Index case 9 American M 20</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Niece American F 42</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Normal</td>
</tr>
<tr>
<td>Niece American F 40</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Son American M 20</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Daughter American F 13</td>
<td>Carotid body</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Index case 10 American M 45</td>
<td>Vagal</td>
<td>Benign</td>
<td>1</td>
<td>Positive</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

Abbreviations: CT, computed tomography; MRI, magnetic resonance imaging; ND, test not done.

*Carrier status was molecularly/genetically confirmed; case not clinically investigated.
†Carrier status was obligatory by pedigree position; case not clinically investigated.
‡Catecholamine decreased but did not return to normal after partial resection of the right jugular paraganglioma.
begin with SDHC testing. Importantly, because no SDHC mutations were found in our population-based registry of 371 symptomatic pheochromocytoma presentations, it is safe to recommend that such patients do not have to be offered SDHC gene testing. We therefore conclude that SDHC mutation testing is as useful a molecular diagnostic adjunct for HNP presentations as are SDHB, SDHD, VHL, and RET gene testing in pheochromocytoma cases.

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**Author Contributions:** Dr Neumann had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Drs Schiavi, Boeder, Bausch, Pęczkowska, and Gommez contributed equally to the manuscript. Study concept and design: Boeder, Pawlu, Walz, Januszewicz, Eng, Opperch, Neumann. Acquisition of data: Schiavi, Bausch, Pęczkowska, Gommez, Strabburg, Salzmann, Hoffmann, Berli, Brink, Cybulia, Muresan, Walter, Forrer, Välimäki, Kawecki, Sztukowski, Schipper, Walz, Pigny, Bauters, Willet-Brozick, Baysal, Januszewicz, Neumann. Analysis and interpretation of data: Boeder, Bausch, Pawlu, Buchta, Salzmann, Hoffmann, Walz, Willet-Brozick, Baysal, Eng, Neumann. Drafting of the manuscript: Schiavi, Pawlu, Salzmann, Pigny, Willet-Brozick, Eng, Opperch, Neumann. Critical revision of the manuscript for important intellectual content: Boeder, Bausch, Pęczkowska, Gommez, Strabburg, Pawlu, Bausch, Hoffmann, Berli, Brink, Cybulia, Muresan, Walter, Forrer, Välimäki, Kawecki, Sztukowski, Schipper, Walz, Bauters, Baysal, Januszewicz, Opperch, Neumann. Statistical analysis: Pawlu. Obtained funding: Baysal, Eng, Neumann. Administrative, technical, or material support: Boeder, Bausch, Gommez, Strabburg, Bauters, Walz, Hoffmann, Berli, Cybulia, Walter, Forrer, Välimäki, Kawecki, Sztukowski, Schipper, Walz, Willet-Brozick, Baysal, Eng, Neumann. Study supervision: Forrer, Pigny, Baysal, Eng, Opperch, Neumann.

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**REFERENCES**


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Limitations of our study include that, as a cohort study, there is a potential that unmeasured confounders caused the observed differences. Small sample size and few outcomes in some of the subgroups also limit the ability to reach definitive conclusions and may account for some of the differences in results compared with Berk et al. As a post hoc analysis, the findings should be considered hypothesis generating. Given these factors, there is a need for randomized trials of treatment for perinatal HIV infection to understand the optimal methods of preventing disease progression. Consideration must also be given to treatment in countries where triple therapy is not available.

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For the Italian Register for HIV Infection in Children

Access to Data: Dr de Martino had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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A complete list of the members of the Italian Register for HIV Infection in Children was published in reference 4.

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CORRECTIONS

Incorrect Units: In the Original Contribution entitled “Excess Dosing of Antiplatelet and Antithrombin Agents in the Treatment of Non–ST-Segment Elevation Acute Coronary Syndromes” published in the December 28, 2005, issue of JAMA (2005; 294:3108-3116), although correct in the dosing recommendation table (Table 1), incorrect units for tirofiban were printed in the accompanying text. On page 3109, the units for tirofiban should have been µg/kg per minute, not µg/kg per hour.