Congenital Stapes Ankylosis Associated With Another Ossicular Chain Anomaly

Surgical Results in 30 Ears

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Objective: To describe the audiometric results after stapes surgery in a consecutive series of patients with stapes footplate ankylosis combined with another ossicular middle ear anomaly.


Setting: A tertiary referral center.

Patients: A total of 25 patients (30 ears) underwent exploratory tympanotomies and ossicular reconstruction.

Main Outcome Measure: Audiometric results.

Results: Overall, a mean gain in air conduction of 18 dB (from 49 dB to 31 dB) and a mean postoperative air-bone gap (ABG) of 20 dB (mean preoperative ABG, 40 dB) were observed. The ABG closure was 20 dB or less in 70% of cases, which is in agreement with the few results reported in the literature. Moreover, the audiometric results remained stable. In the group of ears with a syndrome, the mean gain of air conduction was only 19 dB, which was comparable to that observed among nonsyndromic ears.

Conclusions: Surgery for congenital stapes footplate ankylosis with a concomitant ossicular chain anomaly can provide worthwhile hearing improvement. The ABG closure was 20 dB or less in 21 of 30 ears (70%). Most ears had some sensorineural impairment (10-20 dB), which influenced the final hearing level after surgery. Over recent decades, the technique of the malleostapedotomy procedure has been improved. Preoperative assessment is mandatory for syndromal diagnoses, which might be important for patient counseling and prognosis.

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ferral center, reporting the surgical findings and outcomes of that series. We also report the outcomes of a preoperative syndromic workup. These results are compared with outcomes of similarly categorized series published in the literature.

**METHODS**

Between 1986 and 2001, exploratory tympanotomy was performed on 107 ears; congenital ossicular chain anomalies were observed in 89 patients. The medical records of 25 (30 ears) of these 89 patients, who underwent surgery for a stapes ankylosis associated with another ossicular chain anomaly, were analyzed. All patients were treated at the Department of Otorhinolaryngology, Radboud University Medical Centre, Nijmegen, the Netherlands. In this study, patients who were diagnosed as having osteogenesis imperfecta or otosclerosis were excluded. The minimum age required for surgery was 8 years. All patients had a history of hearing loss since early childhood. A history of long-term middle ear abnormalities was excluded in all cases by medical records, otologic examination findings, and information provided by the referral center. Various surgical techniques have been used to reconstruct the ossicular chain as part of a stapedotomy or stapedectomy procedure.

Computed tomography of the temporal bones was performed mainly to exclude obvious anomalies of the inner ear and the internal acoustic canal and to obtain accurate information on the risk of a stapes gusher. Additional useful information was obtained on pneumatization of the mastoid and the size of the middle ear.

Intraoperative findings were classified according to the Cremers classification of congenital middle ear anomalies (Table 1). The anomalies of the ossicular chain associated with congenital stapes ankylosis (class II) were divided into 3 subgroups: type a for the ears with congenital stapes ankylosis and an ossicular discontinuity; type b for the ears with congenital stapes ankylosis with an epitympanic fixation (Figure 1A and B); and type c for the ears with a stapes ankylosis combined with a tympanic ossicular fixation (including fossa incudis fixation) (Figure 1C).

All patients underwent general anesthesia. A transcanal approach was typically used in combination with a transtympanic approach if (epi) tympanic fixation of the ossicular chain was suspected. After the middle ear was exposed, the mobility of the malleus and the incus was determined. The incudostapedial joint was then separated. Mobilization of the ossicles could be performed by elimination of excessive bone in the epitympanum or tympanum and/or by removal of the incus and/or the head of the malleus. On confirmation of a stapes fixation, a small fenestra stapedotomy or a stapedectomy/platinectomy was performed after removal of the stapedial tendon and the posterior crus. The fenestration was performed by creating a fenestration stapedotomy in the footplate using a diamond drill. The anterior crus with capitulum of the stapes was fractured and removed. Most patients received the Fish Teflon-platinum piston prosthesis for reconstruction of the ossicular chain during incudostapedotomy.

### Table 1. Intraoperative Findings According to the Cremers Classification

<table>
<thead>
<tr>
<th>Class</th>
<th>Anomaly</th>
<th>Subclassification</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Isolated stapes footplate fixation</td>
<td></td>
</tr>
</tbody>
</table>
| II    | Stapes fixation with another congenital ossicular chain anomaly | a. Ossicular discontinuity  
      |                               | b. Epitympanic fixation  
      |                               | c. Tympnic fixation                   |
| III   | Anomaly of ossicular chain but mobile stapes footplate | a. Ossicular discontinuity  
      |                               | b. Epitympanic fixation  
      |                               | c. Tympnic fixation                   |
| IV    | Congenital aplasia/dysplasia of the oval window or round window | a. Aplasia  
      |                               | b. Dysplasia                        |
      |                               | b1. Abnormal facial nerve           
      |                               | b2. Persistent stapedial artery     |

*Adapted with permission from Teunissen and Cremers.*

**Figure 1.** The anomalies of the ossicular chain associated with congenital stapes ankylosis (class II). A, Epitympanic fixation anterior of the malleus head (transcanal approach). B, Epitympanic fixation of the incus head (transmastoid approach). C, Isolated fixation of the short process of the incus in association with an open epitympanum, without fixation (transmastoid approach).
The parameters of patients in this series, which were analyzed and tabulated in a computer database, included age, sex, unilateral or bilateral congenital middle ear anomaly, syndromal diagnosis, comorbidity, otoscopy before surgery, otologic history, preoperative audiometric testing, surgeon, perioperative findings, and surgical technique used. Postoperative hearing results and surgical failures were noted.

Preoperative pure-tone averages for 0.5, 1.0, 2.0, and 4.0 kHz for air conduction (AC), bone conduction (BC), and air-bone gap (ABG) were compared with the corresponding postoperative values. The postoperative reference mean hearing thresholds were obtained at the 1-year follow-up visit. Long-term follow-up data were also collected. Postoperative BC values were accepted as the preoperative BC level to calculate the preoperative ABG unless the postoperative BC hearing level was worse.

Air conduction thresholds in operated ears and nonoperated ears were also evaluated with the Amsterdam Hearing Evaluation Plot, which allows assessment of the individual benefit obtained after surgical intervention (Figure 2A). The 2 dotted diagonal lines enclose the area within bone conduction (BC) that changed less than 10 dB. In 2 cases (patients 13 and 23), a deterioration of the perceptive hearing threshold from 5 to 25 dB was noted. HL indicates hearing level. B, The horizontal axis represents the postoperative change in air conduction (AC), and the vertical axis represents the preoperative ABG (ABG). The solid diagonal line indicates the total closure of the gap between preoperative AC and BC. Most patients (situated between the solid and the dotted diagonal lines) showed an improvement of the ABG to within 20 dB threshold.

Consequently, every point below the diagonal solid line indicates a gain in AC that is larger than would be expected from the preoperative ABG (overclosure). An unsatisfactory surgical result in this graphic presentation is defined as a negative change in the AC threshold or a change in AC that was not sufficient to close the gap between the postoperative AC and the preoperative BC to 20 dB or less. Every point above the dotted line indicates such a result. Other studies use the 1995 American Academy of Otolaryngology–Head and Neck Surgery Committee on Hearing and Equilibrium guidelines, but in this retrospective chart review study, we did not encounter measurements at 3.0 kHz; therefore, we used the 4.0-kHz frequency. Nonetheless, there is evidence that the average of 0.5-, 1.0-, 2.0-, and 4.0-kHz mea-
IIa anomaly was found in 7 ears (10 and 20 years for 22 ears, and older than 20 years for patient aged 8 years, 2 patients aged 9 years), between age at surgery was younger than 10 years for 3 ears (1 and 12 females (17 male ears and 13 female ears). The normality. The study population consisted of 13 males

Preoperative computed tomography (n=19) of the pet-

tion, whereas 1 ear showed general tympanic fixation. In subgroup 2c, we found 11 ears with incudal fossa fixa-
type IIc anomaly was confirmed in 12 ears (Figure 3C).

The categorization of anomalies according to the Cremers classification6 is as follows: a type II ear anomaly or age of the patient (Figure 3A). A type IIb anomaly was encountered in 11 ears (Figure 3B), and a type IIc anomaly was confirmed in 12 ears (Figure 3C).

In subgroup 2c, we found 11 ears with incudal fossa fixa-
tion, whereas 1 ear showed general tympanic fixation. Preoperative audiometric evaluation of postoperative hearing level compared with the preoperative level. Statistical significance was set at P < .05.

RESULTS

Stapes fixation with a concomitant anomaly in the os-
sicular chain of the middle ear was encountered in all 25
patients (30 ears). The categorization of anomalies ac-
cording to the Cremer classification6 is as follows: a type

Table 2. Patient Characteristics and Perioperative Parameters

| Type (n=30) | Unilateral/ Bilateral | Syndrome/ Anomalies | Major | Surgery, | Argon Laser, | Inner Ear | Surgeon, | Incudostapedotomy | Teflon |
|---|---|---|---|---|---|---|---|---|---|---|
| Ear | Yes/No | Yes/No | Endaural/ Combination | Yes/No/NA | on CT (No. of Ears) | Senior Author | Other | Stapes | Gusher | Lesion |

Abbreviations: CT, computed tomography; NA, not applicable.

a Combination of retroauricular and endaural approaches.

b Dr Cremer.

c Four patients (6 ears) with Teunissen-Cremers syndrome.

Stapess fixation with a concomitant anomaly in the os-
sicular chain of the middle ear was encountered in all 25
patients (30 ears). The categorization of anomalies ac-
cording to the Cremers classification6 is as follows: a type

Table 3. Success Rate Closure of the Air-Bone Gap (ABG) in the Incudostapedotomy Group Compared With the Malleostapedotomy Group

<table>
<thead>
<tr>
<th>ABG, dB</th>
<th>Incudostapedotomy</th>
<th>Malleostapedotomy</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group (n=18)</td>
<td>Group (n=12)</td>
<td></td>
</tr>
<tr>
<td>≤10</td>
<td>3 (17)</td>
<td>4 (33)</td>
<td>.28</td>
</tr>
<tr>
<td>≤20</td>
<td>11 (61)</td>
<td>10 (83)</td>
<td>.62</td>
</tr>
<tr>
<td>&gt;20</td>
<td>7 (39)</td>
<td>2 (17)</td>
<td>.14</td>
</tr>
</tbody>
</table>

Overall (n=30), no statistical significant surgical outcome in ABG closure was noted between the 2 surgical procedures (P=.09).

The average preoperative AC for all patients was 49 dB (range, 28-96 dB) (Table 4). The mean preoperative ABG was 40 dB (range, 14-65 dB). The mean postoperative AC was 31 dB (range, 9-78 dB), with a mean postoperative ABG of 20 dB (range, 0-60 dB). A significant hearing gain was observed in this series of surgically treated congenital stapes fixation with a concomitant ossicular anomaly (P < .001). A closure of the preoperative ABG to within 20 dB was achieved in most ears (21 of 30 [70%]) (Figure 2B and 3A). The preoperative and postoperative results at 1 year and at the most recent audiographic evaluation are presented for each patient in Figure 3. No significant correlation was noted between postoperative audiometric results and type of anomaly or age of the patient (Table 6). Postoperative follow-up ranged from 11 to 218 months, with an average of approximately 7.7 years; the median follow-up was 85 months. The long-term results (Table 4) showed a mild deterioration of the audiometric results: BC from 12 to 18 dB and AC from 31 to 39 dB. The mean ABG remained stable: 20 dB after 1 year and 21 dB at the last audiographic evaluation.

In Figure 2A, the individual audiometric outcome after exploratory tympanotomies was illustrated in an alternative manner. The figure shows that the preoperative and postoperative difference in BC remained within 10-dB hearing level in almost all cases. In 2 patients (cases 13 and 23), a deterioration of the perceptive hearing thresh-
old from 5 dB to 25 dB was noted (Figure 2A). One patient (case 13) was diagnosed as having Toriello syndrome,15 and the other (case 23) had Teunissen-Cremers syndrome and was already described in the literature (Dutch case 1).16

Figure 2B shows that disappointing postoperative audiometric results were seen in 11 of the 30 ears (cases 9, 13, 15, 16, 20, 21, 23, 24, 26, 29, and 30). Reexploration was performed in 8 ears (cases 4, 9, 20, 23, 24, 26, 29, and 30). Successful reconstruction was performed in 3 ears (cases 4, 23, and 24). In 2 of these ears (cases 4 and 23), revision surgery was necessary because of Teflon piston luxation; in 1 ear (case 24), recurrent bony closure of the footplate after stapedotomy necessitated revision surgery. In ear 23, the perceptive hearing impairment was encountered during the period between 1 year after surgery and the time of reexploration. None of the reconstructions performed in the other 5 cases were successful. In 1 of 5 ears (case 9), epitympanic ossicular refixation necessitated remodeling of the incus to create a malleostapedotomy. In the second ear (case 20), the disappointing result supported reexploration, and mobilization of the fossa incudis fixation was performed. In case 26, the stapedotomy opening seemed too small, necessitating partial platinectomy, but conductive hearing loss remained after revision. In case 29, a luxation of the Teflon piston perforated the tympanic membrane; during revision surgery, a new piston was added, but the reason for the remaining ABG remains elusive. In 1 patient (case 30), acquired cholesteatoma was observed a few years after primary surgery. Perceptive hearing impairment was obtained in 5 ears (cases 9, 20, 23, 24, and 29), and usually the impairment occurred slowly after reexploration and was progressive. Although not all revisions were successful (cases 9, 20, 26, 29, and 30), the hearing results of this series can generally be considered worthwhile.

A syndromal diagnosis was made in 12 of the 30 ears (40%). Teunissen-Cremers syndrome was diagnosed in 7 ears (cases 18, 19, 21, 23, 24, 27, and 28) and confirmed in some patients by mutational analysis.16-18 Branchio-oto-renal syndrome was found in 3 patients (cases 7, 10, and 16). Other syndromes, including Toriello syndrome15 and lacrimoauriculodentodigital syndrome, were diagnosed in 2 more ears (cases 13 and 14).19 In 11 of these syndromal ears, a clear history of familial hearing was performed. In case 26, the stapedotomy opening seemed too small, necessitating partial platinectomy, but conductive hearing loss remained after revision. In case 29, a luxation of the Teflon piston perforated the tympanic membrane; during revision surgery, a new piston was added, but the reason for the remaining ABG remains elusive. In 1 patient (case 30), acquired cholesteatoma was observed a few years after primary surgery. Perceptive hearing impairment was obtained in 5 ears (cases 9, 20, 23, 24, and 29), and usually the impairment occurred slowly after reexploration and was progressive. Although not all revisions were successful (cases 9, 20, 26, 29, and 30), the hearing results of this series can generally be considered worthwhile.

### Table 4. Audiometric Parameters in 30 Ears Before and After Exploratory Tympanotomies

<table>
<thead>
<tr>
<th>Variable</th>
<th>Mean (SD)</th>
<th>Before surgery</th>
<th>One year after surgery</th>
<th>Long-term follow-up after surgery</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>(n=30)</td>
<td>(n=30)</td>
<td>(n=30)</td>
</tr>
<tr>
<td>AC, dB HL</td>
<td>49.1 (13.3)</td>
<td>31.2 (18.0)</td>
<td>38.6 (19.5)</td>
<td></td>
</tr>
<tr>
<td>BC, dB HL</td>
<td>8.8 (8.0)</td>
<td>11.5 (9.2)</td>
<td>17.6 (15.5)</td>
<td></td>
</tr>
<tr>
<td>ABG, dB</td>
<td>40.3 (12.2)</td>
<td>19.6 (14.2)</td>
<td>21.0 (12.9)</td>
<td></td>
</tr>
<tr>
<td>HG, dB</td>
<td>NA</td>
<td>18.0 (20.7)</td>
<td>19.5 (23.0)</td>
<td></td>
</tr>
<tr>
<td>SNHL, dB</td>
<td>NA</td>
<td>−2.7 (5.3)</td>
<td>−8.8 (14.9)</td>
<td></td>
</tr>
<tr>
<td>ABG Closure, dB</td>
<td>NA</td>
<td>20.7 (18.0)</td>
<td>19.3 (17.7)</td>
<td></td>
</tr>
</tbody>
</table>

### Table 5. Air-Bone Gap and Hearing Gain Following Exploratory Tympanotomy

<table>
<thead>
<tr>
<th>Variable</th>
<th>Present Study</th>
<th>Teunissen and Cremers</th>
<th>Kisilevsky et al</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before surgery</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of ears (total No. of ears in study)</td>
<td>30 (30)</td>
<td>32 (32)</td>
<td>15 (40)</td>
</tr>
<tr>
<td>Follow-up, mo</td>
<td>12</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>Mean BC, dB</td>
<td>9</td>
<td>16</td>
<td>22</td>
</tr>
<tr>
<td>Mean AC, dB</td>
<td>49</td>
<td>53</td>
<td>52</td>
</tr>
<tr>
<td>Mean ABG, dB</td>
<td>40</td>
<td>37</td>
<td>30</td>
</tr>
<tr>
<td>After surgery</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean BC, dB</td>
<td>12</td>
<td>15</td>
<td>21</td>
</tr>
<tr>
<td>Mean AC, dB</td>
<td>31</td>
<td>30</td>
<td>34</td>
</tr>
<tr>
<td>Mean ABG, dB</td>
<td>20</td>
<td>15</td>
<td>13</td>
</tr>
<tr>
<td>HG, dB</td>
<td>18</td>
<td>23</td>
<td>18</td>
</tr>
<tr>
<td>BC loss &gt;10 dB, % (No. of ears)</td>
<td>7 (2)</td>
<td>9 (3)</td>
<td>NA</td>
</tr>
<tr>
<td>ABG =20 dB, % (No. of ears)</td>
<td>70 (21)</td>
<td>63 (20)</td>
<td>NA</td>
</tr>
</tbody>
</table>

### Abbreviations
- ABG, air-bone gap
- AC, air conduction threshold
- BC, bone conduction threshold
- HG, hearing gain
- HL, hearing level
- NA, not applicable
- SNHL, sensorineural hearing loss

The mean preoperative and postoperative values were calculated from the results of stapes surgery. The mean values for AC, BC, HG, ABG, and ABG closure were based on the average thresholds from frequencies 0.5, 1.0, 2.0, and 4.0 kHz. The postoperative BC values were accepted as the preoperative BC level to calculate the preoperative ABG unless the postoperative BC HL was worse.

The SNHL represents the loss in BC thresholds calculated by subtracting the postoperative BC threshold from the preoperative BC threshold.

The ABG closure was calculated by subtracting the postoperative ABG from the preoperative ABG.

Mean, 7.7 years.
In the present series, hearing impairment as a result of congenital stapes ankylosis associated with another congenital ossicular chain anomaly (class II according to the Cremers classification) was observed. The preoperative diagnosis of conductive hearing loss due to congenital anomaly is rather challenging. A serous otitis media or recurrent acute otitis media (which frequently occurs in children) superimposed on the congenital malformation might delay the correct diagnosis and treatment. Furthermore, attempts at surgical management by placement of ventilation tubes without middle ear inspection exacerbates this delay even more, which may adversely affect speech and language development and learning ability. The right treatment is adaption of an appropriate amplification as early as possible or later on a surgical intervention. However, surgery in children younger than 10 years is reported to be less common. Bilateral rather than unilateral hearing impairment makes the need for treatment even more urgent. During the years before successful surgery in patients with bilateral hearing impairment, hearing aids should be worn.

In cases of a syndromal diagnosis with multiple congenital dysmorphic features, hearing loss as a result of a congenital anomaly seems more likely, particularly in cases in which deformities of the branchial arch derivatives are noted, such as in Treacher Collins syndrome and branchio-oto-renal syndrome. In nonsyndromic cases, without external abnormal anatomical features, an adequate diagnosis remains challenging. The tympanic membrane and external ear canal appeared normal in all nonsyndromic cases. Furthermore, conventional computed tomography of the petrous bones did not reveal any anomalies. However, very recently, new radiologic techniques have become available. Currently, the radiographic options used to visualize the middle ear are much more sophisticated. This new development will substantially facilitate preoperative assessment and scheduling for surgery in the near future. The mean preoperative AC in this study was 49 dB; overall, we calculated a gain of 18 dB. Table 4 shows the audiometric outcomes after surgery.

In Figure 2, another format is used to report the postoperative outcome according to the Amsterdam Hearing Evaluation Plots. The purpose of this figure is to visualize the hearing results of each individual ear after stapes surgery. Figure 2A shows that in 2 ears (cases 13 and 23) some iatrogenic inner ear damage was encountered. Also, in Figure 2B, we noted 11 ears (cases 9, 13, 15, 16, 20, 21, 23, 24, 26, 29, and 30) in which an unsatisfactory operative result was achieved; in 6 of these ears (cases 9, 13, 23, 24, 26, and 29), there was a postoperative deterioration in AC.

There are only a few publications about this rare pathologic middle ear entity, and only some are useful for comparison. In this series, thorough descriptions of the deformities and accurate reports enabled us to distinguish between various corresponding deformities. Therefore, it was possible to isolate the surgical findings and results of the patients who were diagnosed as having stapes ankylosis with a concomitant other ossicular chain anomaly. The data are shown in Table 5. A range in AC gain between 18 and 23 dB was obtained. These results are similar to those published previously on surgical outcomes. Moreover, the audiometric results remained stable compared with the previously published series.

The outcome of this series and a few others confirms that surgery for congenital stapes ankylosis and an associated deformity of the ossicular chain is an opportunity to improve hearing level (Table 5). In experienced hands, reconstructive middle ear surgery will generally lead to a considerable improvement in hearing. More recently, the opportunities to apply semi-implantable hearing aids might provide new treatment options, especially when more conventional microsurgical procedures do not provide the desired outcome.

In conclusion, this new, consecutive series of patients with congenital stapes ankylosis associated with another ossicular chain deformity demonstrated worthwhile hearing improvement, and the results are comparable to those in previous reports in the literature on this topic. Because of the recent improvement in surgical techniques, a patient with unilateral or bilateral congenital stapes ankylosis with a concomitant ossicular chain anomaly might therefore benefit greatly from middle ear surgery. Surgery is usually postponed until the patient is 8 to 10 years old.

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Author Contributions: Dr Thomeer had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Study concept and design: Thomeer and Cremers. Acquisition of data: Thomeer and Kunst. Analysis and interpretation of data: Thomeer and Kunst.

Table 6. Audiometric Results According to the Type of Anomaly and Age Group After Exploratory Tympanotomy

<table>
<thead>
<tr>
<th>Type</th>
<th>HG, dB</th>
<th>Closure ABG, dB</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ilb (n=7)</td>
<td>31.8 (9.9)</td>
<td>32.5 (10.4)</td>
</tr>
<tr>
<td>Ilb (n=11)</td>
<td>17.7 (17.60)</td>
<td>20.9 (14.1)</td>
</tr>
<tr>
<td>Ilc (n=12)</td>
<td>10.1 (24.6)</td>
<td>13.5 (21.7)</td>
</tr>
<tr>
<td>Age, y</td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;10 (n=3)</td>
<td>7.9 (7.5)</td>
<td>10.4 (9.0)</td>
</tr>
<tr>
<td>10-20 (n=22)</td>
<td>19.0 (21.1)</td>
<td>21.7 (18.4)</td>
</tr>
<tr>
<td>&gt;20 (n=5)</td>
<td>12.3 (19.8)</td>
<td>15.5 (16.6)</td>
</tr>
</tbody>
</table>

Abbreviations: ABG, air-bone gap; HG, hearing gain.

The mean values for HG and ABG closure were based on the average thresholds from frequencies 0.5, 1.0, 2.0, and 4.0 kHz. No significant correlation was found for any parameter mentioned (P > .05).
Statistical analysis: Thomeer and Cremers. Drafting of the manuscript: Thomeer. Critical revision of the manuscript for important intellectual content: Thomeer, Kunst, and Cremers. Statistical analysis: Thomeer. Study supervision: Kunst and Cremers.

Financial Disclosure: None reported.

Additional Contributions: Iris Post (www.irispost.nl) contributed significantly to the figures produced for this article.

REFERENCES