Adrenal Venous Sampling
Evaluation of the German Conn’s Registry

Oliver Vonend, Nora Ockenfels, Xing Gao, Bruno Allolio, Katharina Lang, Knut Mai, Ivo Quack, Andreas Saleh, Christoph Degenhart, Jochen Seufert, Lysann Seiler, Felix Beuschlein, Marcus Quinkler, Petr Podrabsky, Martin Bidlingmaier, Reinhard Lorenz, Martin Reincke, Lars Christian Rump, for the participants of the German Conn’s Registry

Abstract—In patients with primary aldosteronism, adrenal venous sampling is helpful to distinguish between unilateral and bilateral adrenal diseases. However, the procedure is technically challenging, and selective bilateral catheterization often fails. The aim of this analysis was to evaluate success rate in a retrospective analysis and compare data with procedures done prospectively after introduction of measures designed to improve rates of successful cannulation. Patients were derived from a cross-sectional study involving 5 German centers (German Conn’s registry). In the retrospective phase, 569 patients with primary aldosteronism were registered between 1990 and 2007, of whom 230 received adrenal venous sampling. In 200 patients there were sufficient data to evaluate the procedure. In 2008 and 2009, primary aldosteronism was diagnosed in 156 patients, and adrenal venous sampling was done in 106 and evaluated prospectively. Retrospective evaluation revealed that 31% were bilaterally selective when a selectivity index (cortisol adrenal vein/cortisol inferior vena cava) of ≥2.0 was applied. Centers completing <20 procedures had success rates between 8% and 10%. Overall success rate increased in the prospective phase from 31% to 61%. Retrospective data demonstrated the pitfalls of performing adrenal venous sampling. Even in specialized centers, success rates were poor. Marked improvements could be observed in the prospective phase. Selected centers that implemented specific measures to increase accuracy, such as rapid-cortisol-assay and introduction of standard operating procedures, reached success rates of >70%. These data demonstrate the importance of throughput, expertise, and various potentially beneficial measures to improve adrenal vein sampling. (Hypertension. 2011;57:990-995.)

Key Words: primary aldosteronism ▪ adrenal vein sampling ▪ aldosterone-producing adenoma ▪ bilateral idiopathic hyperaldosteronism ▪ rapid cortisol assay

Primary hyperaldosteronism (PA) is one of the common causes of secondary hypertension.1–2 However, guidelines for screening, confirmatory testing, and procedures to differentiate between unilateral and bilateral disease are rare,3 and cutoff parameters are not prospectively evaluated. More than 98% of patients with PA present with unilateral aldosterone-producing adenoma (APA) or bilateral idiopathic hyperaldosteronism (IHA). In addition, there are monogenic forms of PA.4,5 However, therapy of the 2 subtypes APA and IHA differs substantially. Although hypertension attributed to unilateral APA can be cured surgically by adrenalectomy, IHA is treated by mineralocorticoid receptor antagonists. Because IHA is the more common cause of PA, establishing the correct diagnosis is important to avoid unnecessary surgery. Because imaging by computed tomography (CT) or MRI may not have the diagnostic accuracy to differentiate among APA, IHA, or incidentaloma, adrenal vein sampling (AVS) became the gold standard in localization of the source of autonomous aldosterone production.6 However, AVS is a technically challenging method. In particular, the right adrenal vein, which drains directly into the inferior vena cava, is frequently missed. Although some experienced centers with dedicated and highly motivated staff report successful AVS in >90%,7 many experts suspect that

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success rate in daily reality diverges from these numbers to a considerable extent.

The German Conn’s registry was founded in 2006 to evaluate the diagnosis, treatment, and outcome of PA in Germany.9 Nine centers are currently enrolling patients for prospective analyses. The first 5 institutions provided comprehensive data from 1990 to 2007 for retrospective evaluation. In a prospective phase, 106 AVSs, performed in 2008 and 2009, were evaluated to compare the success rates with retrospective data.

Subjects and Methods

The German Conn’s Registry (www.conn-register.de) was founded in 2006 by the Section of Adrenal Disease, Steroids, and Hypertension of the German Endocrine Society (Deutsche Gesellschaft für Endokrinologie). Nine centers are currently enrolling patients. Retrospectively, 5 centers have included 569 patients diagnosed between 1990 and 2007. As described elsewhere,9 patients were entered into an electronic database after pseudonymization using an identification number. The ethics committees of the University Hospital of Munich and of the participating centers approved the protocol. Personal data protection laws were strictly adhered to.

An interim analysis of the AVS data was performed in 2007 showing a low technical success rate using the below-mentioned criteria for AVS selectivity. The data were presented and discussed during an annual Conn’s registry meeting. Each center thereafter tried to improve success rates of AVSs using different strategies.

In 2008, the German Conn’s registry started to enroll patients with PA in a prospective manner in the Multicenter Evaluation of Primary Hyperaldosteronism: Diagnostic Testing, Subdifferentiation, Therapy, Outcome, and Genetics. Screening procedures and assays were standardized in all of the centers. Diagnosis of PA required an elevated aldosterone:renin ratio, confirmed by a plasma aldosterone not suppressible during a 4-hour saline infusion (2 L IV). In borderline cases, screening using aldosterone:renin ratio and confirmation testing using the saline infusion were recommended for a second time. The AVS data of new patients studied during 2008 and 2009 were prospectively collected from the 5 centers that participated in the retrospective part to evaluate potential improvements in success rate.

The ethics committees of the University of Munich and of the participating centers approved the protocol. Personal data protection laws were strictly adhered to.

Study Population

In the retrospective part of the German Conn’s registry, until 2007, 230 of 569 patients with PA underwent AVS within the 5 participating German centers (Figure 1). Within this retrospectively analyzed data set, cross-sectional imaging (CT or MRI) reports were accessible for 178 patients.

Between 2008 and 2009, the same 5 centers collected prospective data from 156 patients with PA, of whom 112 underwent AVS. Validated cortisol and aldosterone values were obtainable for 106 AVSs.

Selectivity Index During AVS

In all 5 of the centers, AVS was performed with each adrenal vein cannulated sequentially and without adrenocorticotropic hormone stimulation. At least 2 months before performing AVS, the medication was changed to a nondihydropyridine calcium channel antagonist, hydralazine or prazosin according to the patient’s blood pressure. No patient received glucocorticoid medication ≥2 months before procedure. AVS was carried out after 2 to 3 hours of resting in supine position between 8:00 AM and 12:00 AM. A catheter was guided through the femoral vein toward the right adrenal vein and inferior vena cava. Blood was taken by applying a gentle negative pressure for measuring aldosterone (Advantage Direct-Renin, Nichols Institute Diagnostics) and cortisol (Luminescence-Immunoaassay, Roche Diagnostics) values. In centers with a rapid cortisol assay, blood was directly transferred to the laboratory for cortisol determination. After cannulation of the right adrenal vein, blood from the left adrenal vein and simultaneously from the inferior vena cava was obtained. When the rapid cortisol assay did not show a cortisol level in the adrenal vein at least twice as high as in the inferior vena cava, cannulation of the right adrenal vein and the inferior vena cava was repeated. For rapid cortisol measurements, a modified in-house procedure was chosen. Nonclotting plasma test tubes containing lithium heparin anticoagulant and a gel separator were used instead of serum test tubes. The anticoagulant in the plasma specimen inhibits the clotting process and helps to minimize the formation of fibrin after centrifugation and, therefore, reduces turnaround time. Lithium heparin is the most commonly used anticoagulant in clinical chemistry because of its wide compatibility with chemistry tests.10 In addition, we aimed for a direct sample processing to bypass routine laboratory testing. These procedures allowed to keep overall measuring time <30 minutes. Selective AVS was assumed for the purpose of this study, if cortisol of the adrenal vein was ≥100% higher than cortisol in the inferior vena cava (cortisol in adrenal vein $[C_{AV}]$/cortisol in inferior vena cava $[C_{IVC}]$ ≥2.0).

Results

Selective Catheterization

Of the 569 retrospective patients with PA, 230 had received AVS (Figure 1). In 30 cases, AVS could not be evaluated because of missing cortisol values.

Among the 5 centers, AVS was performed in 16% to 89% patients with PA in the retrospective phase and 50% to 88% in the prospective phase. Centers with only a limited number of procedures (A and C) had the lowest rate of bilaterally selective AVSs.

Using $C_{AV}/C_{IVC}$ ≥2.0 as selective adrenal sampling, 61 (30.5%) of 200 patients were successfully catheterized on both sides in the retrospective phase. In 42.5%, AVS was selective only on the left and in 10.5% only on the right. In 33 cases (16.5%), AVS was unsuccessful on both sides. Perfor-
mance of AVS improved in the prospective phase, in which 65 (61.3%) of 106 patients were successfully catheterized on both and 32 (30.2%) only on 1 side. In 9 cases (8.5%), AVS was unsuccessful on both sides. Numerous measures, such as pre-AVS guiding CT, following defined standard operation procedure protocols, rapid cortisol assay during AVS, and focusing procedure to 1 to 2 interventional staff members, were introduced to improve success rate. Furthermore, the procedure was performed with 1 of the treating endocrinologists, nephrologists, or hypertension specialists in attendance and with prompt feedback being given to the radiologist as to whether cannulation was successful or not.

In the Table, a subanalysis of the included centers summarizes the number of patients who received AVS in the retrospective and prospective phase. In addition, the individual selective adrenal sampling rate and measures introduced to improve success rates are shown.

Increasing the selectivity index in the retrospective data set led to a rapid decrease of the number of AVSs assumed to be bilateral selective (Figure 2). The rate of bilateral nonselective sampling increased from 4% to 45%, when the selectivity index was changed from ≥1.1 to ≥5.0 (Figure 2).

Similar observations were made in the prospective phase. When a more stringent selectivity index of $C_{AV}/C_{IVC}\geq 3$ instead of $\geq 2$ was applied, the overall bilateral success rate dropped from 61% to 44%.

In 178 patients of the retrospective data set, reports of cross-sectional imaging (116 CT/62 MRI) were accessible. To compare the findings of imaging with AVS, 37 patients were identified that received CT or MRI, as well as an AVS with bilateral selective cannulation (based on a selectivity index $C_{AV}/C_{IVC}\geq 2$). Using a lateralization index in AVS of $\geq 3$, in only 27% of cases the imaging reports were in agreement with AVS data. Furthermore, some patients received CT and MRI ($n=35$). In only 20 (57%) of 35 cases, both techniques gave the same findings.

### Table. Center-Specific AVS Procedure Performance

<table>
<thead>
<tr>
<th>Center</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>D</th>
<th>E</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Retrospective study (n=200)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patients received AVS/all registered patients (%)</td>
<td>19/53 (36)</td>
<td>67/75 (89)</td>
<td>13/79 (16)</td>
<td>61/294 (21)</td>
<td>40/68 (59)</td>
</tr>
<tr>
<td>Bilateral selective</td>
<td>2 (10)</td>
<td>21 (31)</td>
<td>1 (8)</td>
<td>18 (30)</td>
<td>19 (48)</td>
</tr>
<tr>
<td>Only left selective</td>
<td>10 (53)</td>
<td>30 (45)</td>
<td>3 (23)</td>
<td>28 (46)</td>
<td>14 (35)</td>
</tr>
<tr>
<td>Only right selective</td>
<td>3 (16)</td>
<td>4 (6)</td>
<td>2 (15)</td>
<td>8 (13)</td>
<td>4 (10)</td>
</tr>
<tr>
<td>Bilateral nonselective</td>
<td>4 (21)</td>
<td>12 (18)</td>
<td>7 (54)</td>
<td>7 (11)</td>
<td>3 (7)</td>
</tr>
<tr>
<td><strong>Prospective study (n=106)</strong></td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Patients received AVS/all registered patients (%)</td>
<td>31/57 (54)</td>
<td>23/26 (88)</td>
<td>7/14 (50)</td>
<td>26/30 (87)</td>
<td>19/29 (66)</td>
</tr>
<tr>
<td>Bilateral selective</td>
<td>16 (52)</td>
<td>17 (74)</td>
<td>2 (29)</td>
<td>19 (73)</td>
<td>11 (58)</td>
</tr>
<tr>
<td>Only left selective</td>
<td>9 (29)</td>
<td>4 (17)</td>
<td>3 (43)</td>
<td>5 (19)</td>
<td>6 (32)</td>
</tr>
<tr>
<td>Only right selective</td>
<td>4 (13)</td>
<td>1 (4)</td>
<td>1 (14)</td>
<td>0 (0)</td>
<td>2 (11)</td>
</tr>
<tr>
<td>Bilateral nonselective</td>
<td>2 (6)</td>
<td>1 (4)</td>
<td>1 (14)</td>
<td>2 (8)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Change in success rate bilateral selective from retrospective to prospective, %</td>
<td>+42%</td>
<td>+43%</td>
<td>+21%</td>
<td>+43%</td>
<td>+10%</td>
</tr>
<tr>
<td>Measures to improve success rate</td>
<td>1 to 4</td>
<td>1 to 5</td>
<td>1, 2, 4</td>
<td>1 to 4</td>
<td>1 to 2</td>
</tr>
</tbody>
</table>

Evaluation of patients who received AVS in 5 different centers (A through E). The individual quote of selective catheterization is given for each center. Data based on AVS using a selectivity index of $\geq 2.0$. Measures introduced to improve success rate are as follows: (1) pre-AVS guiding CT; (2) defined standard operation procedure protocol for AVS including prelabeled test tubes; (3) rapid cortisol assay during AVS; (4) focusing procedure to 1 to 2 interventional staff members; (5) interdisciplinary procedure (endocrinologist or nephrologist attending AVS). Selectivity index = cortisol in adrenal vein/cortisol in inferior vena cava. C indicates cortisol; A, aldosterone; AV, adrenal vein; IVC, inferior vena cava. AVS indicates adrenal vein sampling; CT, computed tomography.

![Figure 2. Columns represent the percentage of bilateral selective (black), only 1-side selective (light gray), and bilateral nonselective (dark gray) AVSs of retrospective and prospective data sets using selectivity indexes $\geq 1.1$, $\geq 2.0$, $\geq 3.0$, $\geq 4.0$, and $\geq 5.0$. The given numbers indicate the percentage of bilateral nonselective AVS. Selectivity index=$C_{AV}/C_{IVC}$. C indicates cortisol; AV, adrenal vein; IVC, inferior vena cava.](http://hyper.ahajournals.org/content/112/7/992/F2)
Discussion

When PA is diagnosed, one of the most challenging aspects is the differentiation between unilateral APA and bilateral IHA. Making this distinction is important, because different therapy is assigned to the individual diagnosis. Four procedures that can help to distinguish these subtypes of PA include cross-sectional imaging (CT and MRI), iodocholesterol scintigraphy, posture test, and AVS. Iodocholesterol scintigraphy, performed in combination with dexamethasone suppression, has the advantage of correlating function with anatomic abnormalities to a certain degree. However, sensitivity of this test depends heavily on the size of the adenoma. In addition, iodocholesterol scintigraphy is not available in most centers. Because the posture test often fails to detect the paradoxical drop of serum aldosterone after posture in unilateral PA, and imaging is misleading in many cases, successful AVS is the gold standard criterion to distinguish between unilateral and bilateral diseases and is therefore suggested by recent guidelines.

Although CT is widely used to detect or exclude APA, data supporting this approach are limited. The sensitivity and specificity of CT and MRI are <50% to 70%. In a recent meta-analysis, CT, MRI, and AVS were re-evaluated in 950 patients from 38 studies. In 38% of cases, CT and MRI gave incorrect diagnosis leading to a wrong decision. The actual data provide, although in a very small subgroup of patients, some evidence for an even lower correlation between imaging and AVS. In the retrospective data set, imaging did not agree with AVS in 73% of cases. One has to point out that, other than the small number of this very heterogenic subpopulation, other technical issues might explain the low correlation. The cross-sectional imaging was performed with different protocols, and findings were reported by local nonblinded staff. In the prospective phase, evaluation of radiological findings will be performed centrally by blinded staff.

However, imaging is mandatory to evaluate adrenal gland anatomy to detect large lesions that may warrant removal based on malignant potential or as ancillary information when AVS does not provide conclusive results. Moreover, CT can also be used before AVS to localize the adrenal veins (in particular, the right vein) in relationship to other anatomic structures. This can help to minimize fluoroscopy time of the patient and to reduce frustration of the staff members performing AVS.

A retrospective and a prospective AVS analysis was conducted to assess rates of successful AVS across several clinical centers with varying expertise and methodological approaches and whether this rate could be improved by the introduction of remedial measures. It is important to note that all of the data were collected by university centers that are familiar with handling PA patients.

Selective Catheterization

When evaluating all of the performed AVSs, there were several procedures with incomplete aldosterone and/or cortisol data. In a retrospective analysis, it is impossible to distinguish between incorrectly performed AVSs and missing data. However, in a number of otherwise comprehensively investigated cases, cortisol values were not collected during AVS. This is clear evidence that standard operating procedures are needed when performing AVS.

To calculate lateralization, bilateral successful cannulation is mandatory. In case of selective sampling, cortisol value in adrenal sample is higher than in inferior vena cava (Selectivity index = CIAV > CIVC). We considered AVS successful when the cortisol value in the adrenal sample was twice as much as in the inferior vena cava (CIAV/CIVC ≥2). Different cutoff values have been described. Some authors defined a 10% higher cortisol value than in inferior vena cava, whereas others rely on higher indexes, such as 2, 3, or even 5. Accessory hepatic veins often drain together with the right adrenal vein into the inferior vena cava. Under these circumstances, adrenal blood with high cortisol values mixes with hepatic blood with a low cortisol value (lower than in the peripheral circulation because of hepatic metabolism of cortisol). Therefore, a high-selectivity index may not be achievable in such instances. It is important to note that centers demanding high cutoff values often use cosyntropin infusion (ACTH) during AVS. Cutoff values are markedly higher when ACTH infusion is used. Whether ACTH infusion increases AVS accuracy remains controversial. Because all of the AVS procedures in our analysis were performed without ACTH infusion, we cannot add additional data to the discussion on that issue. There are possible advantages of ACTH infusion during AVS. ACTH minimizes stress-induced fluctuations in aldosterone secretion during sequential AVS and increases the gradient in cortisol values from the adrenal vein. However, Rossi et al point out that an increase in the rate of bilateral selective AVS by ACTH infusion comes at a price of confounding the outcome with respect to lateralization. Therefore, Rossi et al did not find an overall benefit of ACTH infusion on AVS results. Our data confirmed that a more restrictive selectivity index leads to a striking decrease in the number of AVS procedures that can be regarded as successful. Thus, the rate of bilateral unsuccessful cannulation rose from 4% to 45% when a more stringent cutoff value of 5.0 was used compared to 1.1.

In certain situations, AVS might be helpful even when only 1 side is selectively cannulated. When I AVS is not selective, lateralization of aldosterone:cortisol ratio (ACR) cannot be calculated. However, a comparison between the ACR of the selective adrenal vein and the vena cava can be done. With an elevated ratio (ACRselective/ACRIVC >1), the unilateral selective AVS does not provide useful information, because one cannot differentiate between APA and IHA. In contrast, a suppressed ratio (ACRselective/ACRIVC ≤1) might support a suspected APA on the contralateral side. However, those authors did not provide recommendations for suitable lateralization and suppression indexes.

Of 200 AVSs that had been performed, in only 30.5% of cases were both adrenal veins selectively catheterized using the post hoc defined criteria (selectivity index ≥2). Assessing data of each center individually, it became clear that centers performing a small number of AVS had the lowest rate of bilateral selective catheterization. Thus, in these centers, rates as low as 8% to 10% were apparent. However, only 1 center reached a bilateral selective catheterization rate of...
48%. Centers with a rate between 28% and 30% were “intermediate.”

As stated by others, dedication and repetition are key factors to increase success rate. In a self-critical study from Harvey et al., where 4 different radiologists performed AVS in 60 patients, the success rate of bilateral adrenal vein cannulation was only 42%. This reflects the situation found in the German Conn’s registry. We share the authors’ conclusion that AVS interventions should be limited to a few radiologists to increase their expertise. Centers with <15 AVSs per year should consider referring patients to a center with more technical expertise.

In addition to dedicated radiologists, written standard operating procedures are recommended for all of the participants (radiologists, endocrinologists, and hypertension specialists). This may include directions on the sequence of all required blood samples and to prepare and label all test tubes before AVS to avoid confusion while performing the procedure. In addition, rapid cortisol assays during the procedure can improve the success rate of AVS. Even without rapid cortisol assays, a prompt feedback to the radiologist, whether cannulation was selective or not, should accelerate the learning curve. An endocrinologist or hypertension specialist cannot manually support the interventionalist, but we recommend that, during the procedure, another knowledgeable physician is present for a significant time. The identification of the involved staff with this rather difficult procedure and the continuous encouragement during AVS seems to have a significant impact on the success rate in our experience. Discussing procedural strategies recommended in recent references may also help to improve AVS quality.

In the prospective phase of analysis, 106 AVSs were evaluated in 2008 and 2009. After communication of poor procedure quality, selected centers started to introduce defined standard operating procedures, including rapid cortisol assay and a close collaboration with the local interventional radiologist. In the actual set of data, the rate of bilateral successful AVSs increased to an overall rate of 61%. However, the AVS quality was, with bilateral success rates ranging from 29% to 74% between the centers, still very inconsistent. Centers that used 4 or 5 of the defined standard operating procedures (centers A, B, and D) increased their bilateral success rate by 42% and 43%, whereas the center that used only 3 of the methods (excluding rapid cortisol assay, center C) had a change of only 21%. The center that used only 2 of the methods (E) had an even lower change of 10%. AVS success rate depends critically on throughput and expertise and can be improved through a variety of measures, including rapid cortisol assays.

Perspectives
Retrospective analysis revealed a mean success rate of AVS of only 30.5%. This indicates that AVS should not be performed outside of specialized centers. In addition, there are several key factors that could be modified to increase AVS accuracy. In the prospective phase of German Conn’s registry it was possible to improve the success rate of AVS to 61%. By application of 4 to 5 of the suggested factors, an increase in bilateral selective cannulation rate of >40% is possible.

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