Primary Aldosteronism

Influence of Diagnostic Criteria on the Interpretation of Adrenal Vein Sampling

Gaëlle Lethielleux, Laurence Amar, Alain Raynaud, Pierre-François Plouin, Olivier Steichen

See Editorial Commentary, pp 704–706

Abstract—Guidelines promote the use of adrenal vein sampling (AVS) to document lateralized aldosterone hypersecretion in primary aldosteronism. However, there are large discrepancies between institutions in the criteria used to interpret its results. This study evaluates the consequences of these differences on the classification and management of patients. The results of all 537 AVS procedures performed between January 2001 and July 2010 in our institution were interpreted with 4 diagnostic criteria used in experienced institutions where AVS is performed without cosyntropin (Brisbane, Padua, Paris, and Turin) and with criteria proposed by a recent consensus statement. AVS procedures were classified as unsuccessful, lateralized, or not lateralized according to each set of criteria. Almost 5× more AVS procedures were classified as unsuccessful with the strictest criteria than with the least strict criteria (18% versus 4%, respectively). Similarly, over 2× more AVS procedures were classified as lateralized with the least stringent criteria than with the most stringent criteria (60% versus 26%, respectively). Multiple samples were available from ≥1 side for 155 AVS procedures. These procedures were classified differently by ≥2 right–left sample pairs in 12% to 20% of cases. Thus, different sets of criteria used to interpret AVS in experienced institutions translate into heterogeneous classifications and hence management decisions, for patients with primary aldosteronism. Defining the most appropriate procedures and diagnostic criteria is needed for AVS to achieve optimal performance and fully justify its status as a gold standard. (Hypertension. 2015;65:849–854. DOI: 10.1161/HYPERTENSIONAHA.114.04812.)

Key Words: adrenal glands, blood supply • catheterization, peripheral • diagnosis, differential • hyperaldosteronism • reproducibility of results

Patients with primary aldosteronism (PA) have either bilateral (idiopathic hyperplasia) or unilateral (aldosterone-producing adenoma or unilateral hyperplasia) adrenal disease.1,2 Localizing the source of excessive aldosterone secretion is critical to select the appropriate therapy. Unilateral PA can be treated by laparoscopic adrenalectomy, which normalizes serum potassium concentrations in almost all cases and cures or improves hypertension in >80% of patients.3,4 By contrast, medical management with mineralocorticoid receptor antagonists is recommended as the first-line treatment in bilateral PA.5

Adrenal vein sampling (AVS) is currently considered the gold standard to differentiate unilateral from bilateral aldosterone hypersecretion and is recommended in almost all patients with PA who are eligible for surgical management.5,6 The accuracy of catheter placement in the adrenal veins can be verified from the cortisol concentration in the adrenal veins divided by that in the inferior vena cava or a peripheral vein (selectivity index [SI]). If the SI reaches the predefined cut-off, the lateralization index (LI) is then used to classify PA into unilateral or bilateral disease. The LI is defined as the highest aldosterone/cortisol concentration ratio (dominant adrenal vein) divided by the lowest aldosterone/cortisol concentration ratio (nondominant adrenal vein). Some investigators also propose the use of the contralateral suppression index, defined by the aldosterone/cortisol concentration ratio of the nondominant adrenal vein divided by the peripheral aldosterone/cortisol concentration ratio.6 However, the protocol of AVS (simultaneous or sequential bilateral AVS, use of cosyntropin or not, and the protocol of cosyntropin infusion) and the SI and LI cut-offs used to interpret the results are not currently standardized.7–9

We have interpreted a large number of AVS procedures with criteria used in 4 reference centers or proposed by a recent consensus statement.6,10–12 The first objective of this study was to examine to which extent the interpretation of AVS may differ according to the criteria used for its interpretation. The second goal was to assess the reproducibility of this interpretation when multiple blood samples were drawn from the same adrenal vein.

Methods

Patients

This retrospective study included all patients who underwent AVS in our hypertension referral center between January 2001 and July
2010. Clinical, biochemical, hormonal, and radiological data were extracted from the electronic health record database. Patients were initially referred for tests because they had resistant or hypokalemic hypertension or suspected PA. Our methods for diagnosing PA have been described previously.13 Plasma aldosterone and active renin concentrations and urinary aldosterone concentrations were measured ≥2 weeks after the discontinuation of interfering antihypertensive drugs whenever possible (6 weeks for mineralocorticoid receptor antagonists and aliskiren). If necessary, calcium antagonists and β-blockers were prescribed to control high blood pressure in patients with severe hypertension. Patients with persistent hypokalemia were given 3 to 6 g of potassium chloride daily to maintain serum potassium concentrations ≥3.5 mmol/L, and unrestricted dietary sodium intake was recommended.

A plasma aldosterone/renin ratio cut-off value of 64 pmol/mU/l on 2 occasions was used as a screening test. The minimum active renin concentration was set at 5 mU/l for this calculation to avoid overestimating aldosterone/renin ratio because of undetectable concentrations of active renin. If the aldosterone/renin ratio exceeded 64 pmol/mU/l, the diagnosis of PA was based on a recumbent plasma aldosterone concentration of >500 pmol/l, a sitting plasma aldosterone concentration of >550 pmol/l, or a urinary excretion of aldosterone of >63 nmol/24 h.

Before 2008, AVS was reserved for patients with a diagnosis of PA and without a typical solitary adenoma. However, since 2008, a suppression test (the saline infusion test) and AVS procedures have been systematically performed for all PA patients eligible for surgery, according to guidelines from the Endocrine Society.1 All patients underwent an abdominal computed tomographic scan or MRI before AVS. The retrospective use of anonymous patient data collected during routine care and the procedures followed for this study are in accordance with French institutional guidelines.

AVS Protocol

AVS procedures were performed under the same standardized conditions used for PA diagnosis. Mineralocorticoid receptor antagonists were systematically discontinued. However, 25% to 30% of patients were assessed without the complete withdrawal of other interfering antihypertensive medications because of severe hypertension.14 Bilateral simultaneous AVS without cosyntropin stimulation was performed in the morning after a night sleep in the supine position, by the same experienced vascular radiologist for a 10-year period (A.R.). Two catheters were inserted via the femoral vein, and their correct placement was verified before sampling by the injection of a small amount of contrast agent. Blood samples were then collected simultaneously from each adrenal vein and the inferior vena cava for the measurement of cortisol and aldosterone concentrations. If the radiologist had any doubt about the catheter placement during the procedure, a second or even a third sample was collected.

Diagnostic Criteria

We used Medline to search for papers providing precise descriptions of protocols and diagnostic criteria used to carry out AVS without cosyntropin at other centers between 2001 and 2010. This identified 3 centers: Brisbane, Padua, and Turin.10–12 We also used the criteria proposed by a recent consensus statement.6 Each AVS performed in our unit between January 2001 and July 2010 was analyzed according to the 5 different sets of criteria reported in Table 1.

AVS procedures were classified as (1) unsuccessful, if they did not meet the criterion of selective AVS, (2) lateralized on the left or right side, if successful and meeting the criteria for lateralization on the considered side, or (3) not lateralized, if successful but not meeting the criteria for lateralization. For AVS with multiple samples available from 2 adrenal veins, we compared the classifications resulting from the use of the arithmetic mean of all aldosterone/cortisol concentration ratios on the same side, from the most selective sample on each side, or from all possible pairs of left and right side selective samples.

Measurements

Blood pressure was recorded 3× with a semiautomatic manometer (Omrón 705SCP), in the lying-position, with a 5-minute rest period before measurements.15 Sodium and potassium concentrations were measured by a standard method. The glomerular filtration rate was estimated by the 4 variables Modification of Diet in Renal Disease (MDRD) study equation. Active renin concentrations were determined by radioimmunoassay (LIAISON; Diasorin, Antony, France), and aldosterone concentrations were determined by immunoassay (Cot-A-Count; Siemens Medical Solutions Diagnostics, Saint-Denis, France). The urinary excretion of aldosterone was calculated from free aldosterone and aldosterone from the hydrolysis of aldosterone 18-glucuronide at pH 1.

Statistical Analysis

Descriptive statistics are reported as percentages or medians (interquartile range), as appropriate, and differences were evaluated by the Fisher or the Mann–Whitney U tests, respectively.

Each AVS was classified according to each set of diagnostic criteria. The arithmetic mean of aldosterone/cortisol concentration ratios was chosen as the default method to handle multiple selective samples from the same side.

The effect of the approach used to handle multiple selective samples from the same side (arithmetic mean, consideration of the sample with the highest SI only) was evaluated in terms of AVS interpretation in each center. The reproducibility of AVS with each set of criteria was also evaluated using AVS with multiple selective samples from ≥1 side. All possible combinations of selective samples were used to simulate all LI that could have resulted by chance if only 1 selective sample had been available on each side. The LI most favorable to the left adrenal vein was then compared with the LI most favorable to the right side to evaluate the maximum discordance between samples.

The concordance of the classifications between or within centers was assessed with k coefficients.16 All analyses were performed with Stata 9.2 (StataCorp, College Station, TX).

Table 1. Interpretation Criteria and Classification of AVS in the Different Centers

<table>
<thead>
<tr>
<th>Set of Criteria</th>
<th>Selectivity Criterion</th>
<th>Unsuccessful AVS</th>
<th>Lateralization Criteria</th>
<th>Lateralization of Aldosterone Hypersecretion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brisbane</td>
<td>SL≥3</td>
<td>99 (18%)</td>
<td>LI≥2 and CSI&lt;1</td>
<td>299 (56%)</td>
</tr>
<tr>
<td>Padua</td>
<td>SL≥1.1</td>
<td>19 (4%)</td>
<td>LI≥2</td>
<td>195 (36%)</td>
</tr>
<tr>
<td>Paris</td>
<td>SL≥2</td>
<td>64 (12%)</td>
<td>LI≥5</td>
<td>308 (57%)</td>
</tr>
<tr>
<td>Turin</td>
<td>SL≥2</td>
<td>64 (12%)</td>
<td>LI≥4 or (LI≥3 and CSI&lt;1)</td>
<td>266 (50%)</td>
</tr>
<tr>
<td>Consensus</td>
<td>SL≥2</td>
<td>64 (12%)</td>
<td>LI≥2</td>
<td>174 (32%)</td>
</tr>
</tbody>
</table>

SI=selectivity index=cortisol adrenal vein /cortisol inferior vena cava, LI=lateralization index=(aldosterone/cortisol) dominant side /(aldosterone/cortisol) nondominant side, CSI=contra-lateral suppression index=(aldosterone/cortisol) contra-lateral adrenal vein / (aldosterone/cortisol) dominant adrenal vein, AVS indicates adrenal vein sampling.
The highest agreement (92%) was observed between Turin and Paris, the 2 centers with the closest SI and LI (Table 3).

### Use of the Most Selective Among Multiple Samples

Multiple blood samples were available from ≥1 side for 155 of the 537 AVS procedures (29%). After the application of SI from each center, multiple selective samples were available from ≥1 side for 155 (29%) of AVS according to Padua criteria, 142 (26%) according to Turin, Paris or consensus criteria, and 132 (25%) according to Brisbane criteria.

We used the aldosterone/cortisol concentration ratio from the most selective sample on each side instead of the mean of the ratios from all selective samples to compute the LI and interpret these 155 AVS procedures. The disagreements and $\kappa$ coefficients between both interpretations are shown in Table 4 for each set of criteria. In all cases of disagreement, the interpretation changed from lateralized to nonlateralized or vice versa but never from lateralized on one side to lateralized on the other.

### Between-Sample Reproducibility

We then computed the 2 most discordant interpretations that could have occurred if only 1 sample had been available on each side for the 155 AVS procedures with multiple selective samples. The disagreements and $\kappa$ coefficients between these 2 most extreme possible interpretations are shown in Table 4 for each set of criteria. The majority of disagreements involved changes from lateralized to nonlateralized or vice versa. However, 4 of 31 disagreements with Padua criteria and 3 of 30 with consensus criteria involved changes from lateralized on one side to lateralized on the other.

### Discussion

AVS is currently considered the gold-standard test to diagnose unilateral aldosterone excess in PA. This procedure will be increasingly used, given the raising detection rate of PA. However, it is an invasive and expensive procedure that may result in complications, and thus, it should only be used if it provides reliable and useful diagnostic information. Our findings reveal that the interpretation of AVS may differ according to the criteria used for its analysis, suggesting that some patients may be misdiagnosed and improperly managed because of a lack of standardization.

### Comparison With Previous Studies and Interpretation of Our Results

Kline et al compared 10 different AVS protocols (with or without cosyntropin stimulation) in 63 patients. The success rate of AVS ranged between 13% and 77%, according to the SI used, and only 17% of patients were uniformly classified by all diagnostic criteria (8% as having bilateral aldosterone excess and 9% as having unilateral aldosterone excess).

### Table 2. Baseline Characteristics of Patients

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Values</th>
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<tbody>
<tr>
<td>Men</td>
<td>343/524 (65%)</td>
</tr>
<tr>
<td>Age at (first) adrenal venous sampling, y</td>
<td>48 (41–54)</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>28 (24.9–31.1)</td>
</tr>
<tr>
<td>Systolic blood pressure, mmHg</td>
<td>149 (136–160)</td>
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<tr>
<td>Diastolic blood pressure, mmHg</td>
<td>93 (85–101)</td>
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<tr>
<td>Serum potassium, mmol/L</td>
<td>3.4 (3.1–3.7)</td>
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<tr>
<td>Estimated glomerular filtration rate, mL/min/1.73 m²</td>
<td>91 (73–103)</td>
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<td>Plasma aldosterone, pmol/L</td>
<td>539 (367–790)</td>
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<tr>
<td>Plasma active renin, mUI/L</td>
<td>3 (1–5)</td>
</tr>
<tr>
<td>Aldosterone:renin ratio, pmol/mUI</td>
<td>94 (65–140)</td>
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<tr>
<td>Urinary aldosterone, nmol/24 h</td>
<td>77 (54–105)</td>
</tr>
<tr>
<td>Antihypertensive drug classes, n</td>
<td>2 (1–3)</td>
</tr>
<tr>
<td>Adrenal imaging*</td>
<td></td>
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<tr>
<td>Typical unilateral adenoma</td>
<td>96/520 (19%)</td>
</tr>
<tr>
<td>Other unilateral lesions</td>
<td>129/520 (25%)</td>
</tr>
<tr>
<td>Bilateral lesions</td>
<td>96/520 (19%)</td>
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<tr>
<td>Apparently normal adrenal glands</td>
<td>199/520 (38%)</td>
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Values are given as numbers (percentages) or medians (interquartile range).

*Typical unilateral adenoma is radiolucent, smooth, and ≥8 mm; other unilateral lesions comprise unilateral hyperplasia or atypical adenoma; bilateral lesions comprise bilateral hyperplasia or bilateral adenomas.

### Table 3. Discordances Between Sets of Criteria for Adrenal Vein Sampling Interpretation

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<tbody>
<tr>
<td>Disagreement</td>
<td>40%</td>
<td>16%</td>
<td>17%</td>
<td>33%</td>
<td>26%</td>
<td>9%</td>
<td>25%</td>
<td>17%</td>
<td>31%</td>
<td>8%</td>
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<tr>
<td>$\kappa$</td>
<td>0.44</td>
<td>0.74</td>
<td>0.74</td>
<td>0.53</td>
<td>0.64</td>
<td>0.88</td>
<td>0.75</td>
<td>0.76</td>
<td>0.57</td>
<td>0.88</td>
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</table>
Mulatero et al9 reviewed 64 patients who had undergone AVS twice because the first procedure was unsuccessful according to stringent criteria. The analysis of these AVS results with 2 other sets of diagnostic criteria revealed poor agreement (35%) between the interpretation of the first and second AVS with the most lenient set of criteria (including 14% of AVS changing from unilateral in one side to unilateral in the contralateral side).

These results are consistent with our findings and show that the choice of SI and LI strongly affects the interpretation of AVS and subsequent therapeutic decisions: the same AVS may be considered selective and lateralized in one center, nonselective in a second center, and selective but not lateralized in a third center, leading to radically different patient management.

The arithmetic mean is usually used to average aldosterone/cortisol ratios from multiple selective samples from the same adrenal vein. Another option is to retain only the sample with the highest SI from each adrenal vein because the SI measures sampling quality. Our results show that this choice may affect the interpretation of AVS, although we are unable to determine which interpretation is more accurate.

The concordance between the 2 most discrepant AVS interpretation criteria (Padua and Brisbane) was similar to that reported between AVS and adrenal imaging in a meta-analysis of 38 studies.20 Discordance between AVS and adrenal imaging is commonly attributed to adrenal incidentalomas that may coexist with bilateral hyperplasia or to small adenomas or unilateral hyperplasia that is not detected by imaging.22,23 However, discordance may also be caused by the failure or misinterpretation of AVS. Aldosterone hypersecretion,25,26 and even nonsuppressible aldosterone hypersecretion,27 may persist after adrenalectomy on the basis of AVS results. Such outcomes indicate erroneously lateralized AVS. In rare cases, patients have been hormonally cured after adrenalectomy on the side of a typical Conn adenoma, despite AVS indicating bilateral PA or unilateral PA in the opposite side.28,29

Despite these disturbing reports, the clinical consequences of the inaccurate interpretation of AVS are uncertain. Mineralocorticoid receptor antagonists can correct hypertension and hypokalemia in unilateral PA.30 Thus, the main problem arises when a lateralized AVS leads to unjustified surgery because of bilateral or, even more concerning, contralateral disease. Our results show that this possibility may occur with the most lenient selectivity criteria.

### Practical Implications

Many authors have attempted to measure diagnostic accuracy of AVS.31,32 However, false negatives may go unnoticed because patients without diagnosed lateralized hypersecretion do not undergo surgery.33 False positives may also go unnoticed because it is difficult to determine whether PA has been cured after surgery: essential hypertension can persist after adrenalectomy in unilateral PA.34,35 Postoperative suppression tests are therefore necessary to ascertain postoperative cure of PA but are consistently performed in a few reference centers only. Consequently, neither the sensitivity nor the specificity of AVS can be accurately estimated.

The same methodological concerns apply to the determination of the best set of interpretation criteria. Theoretically, the optimal SI and LI thresholds should be determined as the strongest predictors of postsurgical cure in a group of patients with PA all operated on, irrespective of their AVS results.9 However, such a study would be unethical. Mulatero et al9 proposed that the best thresholds are those that achieve the most reproducible classification in patients subjected to 2 AVS procedures.9 This predictably leads to the adoption of the strictest set of criteria. Pragmatic considerations also hint toward this direction. Indeed, surgical treatment of bilateral PA is probably less desirable than medical treatment of unilateral PA. Hence, false negatives of AVS are less of a concern than false positives and high diagnostic thresholds seem preferable. Although the best value of the thresholds is unknown, it is nonetheless problematic that patients with the same AVS results may not receive the same treatment in 2 different reference centers. Ideally, diagnostic criteria should be standardized across centers.

Moreover, it is a common practice to take duplicate blood samples during AVS, and there is no consensus about what sample should be used or how they should be combined. These factors may further increase the heterogeneity of AVS interpretation. In our study, the strictest criteria (Brisbane) resulted in the highest reproducibility between multiple samples. This finding strengthens the idea that stringent diagnostic thresholds are preferable. However, it is still unclear whether the use of the contralateral suppression index (as implemented in Brisbane) improves the diagnostic accuracy of AVS in cases where the LI is high. Indeed, the lack of contralateral suppression was not associated with a lower rate of response to adrenalectomy in patients with a LI >4 in a recent study.36

The limited concordance between samples observed in our study challenges previous results showing a high concordance between single and duplicate samples.37 Variability of the aldosterone/cortisol concentration ratio between samples from the same adrenal vein could have resulted in different interpretation with the same set of criteria if only 1 of these samples had been available, resulting in the same patient receiving different treatments in the same reference center depending on the available sample.

### Limitations

Some limitations of our study need to be highlighted. First, the analysis is retrospective, but the data were recorded prospectively and few values were missing in the electronic database. Second, some patients underwent AVS without the withdrawal of interfering antihypertensive medications. Renin
unsuppression could theoretically lead to nonlateralized AVS despite unilateral disease; this would affect interpretations with strict criteria more than interpretations with lenient criteria and may therefore increase the number of discordances. However, using potent antihypertensive drugs in high-risk patients matches real-life practice, and some studies suggest that AVS remains informative in patients taking interfering drugs and even in those taking spironolactone.6,38 Third, we performed bilateral simultaneous AVS, whereas the 3 other centers perform sequential AVS. Because simultaneous sampling abolishes a source of variability, our results may underestimate the true overall variability of AVS results between centers. However, the other centers report a gap <10 to 15 minutes between sampling of the 2 adrenal veins, and this source of variability is probably minimal. Fourth, we used a radioimmunoassay assay for aldosterone measurements, and using mass spectrometry may decrease analytic variability and between-sample variability. Fifth, the design of our study does not enable us to provide recommendations about the optimal SI and LI that should be used, or about potential improvements (such as using metanephrines instead of cortisol for computing the SI), or alternatives to AVS (such as functional imaging with ¹³C-metomidate).

Perspectives

AVS is currently considered the reference test for assessing the lateralization of aldosterone hypersecretion in PA and is considered preferable to imaging. However, its results should be interpreted with caution because cut-offs determining the success of adrenal sampling and the lateralization of hypersecretion are not standardized. Different criteria currently used in experienced institutions translate into heterogeneous classifications and hence management decisions for patients with PA (with discordance in up to 40% of AVS). Moreover, the true specificity and sensitivity of AVS are unknown, and evidence of its superiority compared with adrenal imaging is methodologically weak because of the lack of independent gold standard. Standardization is a prerequisite to collaborative research and to the evaluation of the diagnostic and prognostic value of AVS. A recent expert consensus statement on the interpretation of AVS proposed a cut-off value of SI ≥2 and LI ≥2 under unstimulated conditions and SI ≥3 and LI ≥4 during cosyntropin stimulation.6 This consensus may allow the interpretation of AVS to be standardized across centers, although the use of cosyntropin, the usefulness of multiple blood samples from the same vein (and the best way to handle them), and even the true diagnostic value of AVS remain uncertain. As a further note of caution, the LI ≥2 under unstimulated conditions led 3 of our AVS with multiple selective samples to be possibly interpreted both as lateralized to the left or lateralized to the right, depending on the samples used for the LI calculation. This undesirable behavior does not occur in our AVS with multiple samples when the LI threshold is ≥3.

Disclosures

None.

References


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