

AperTO - Archivio Istituzionale Open Access dell'Università di Torino

**Aldosterone suppression on contralateral adrenal during adrenal vein sampling does not predict blood pressure response after adrenalectomy.**

**This is the author's manuscript**

*Original Citation:*

*Availability:*

This version is available <http://hdl.handle.net/2318/154962> since 2020-06-17T09:15:29Z

*Published version:*

DOI:10.1210/jc.2014-2345

*Terms of use:*

Open Access

Anyone can freely access the full text of works made available as "Open Access". Works made available under a Creative Commons license can be used according to the terms and conditions of said license. Use of all other works requires consent of the right holder (author or publisher) if not exempted from copyright protection by the applicable law.

(Article begins on next page)

This is the author's final version of the contribution published as:

Monticone S;Sato F;Viola A;Fischer E;Vonend O;Bernini G;Lucatello B;Quinkler M;Ronconi V;Morimoto R;Kudo M;Degenhart C;Gao X;Carrara D;Willenberg HS;Rossato D;Mengozzi G;Riester A;Paci E;Iwakura Y;Burrello J;Maccario M;Giacchetti G;Veglio F;Ito S;Reincke M;Mulatero P. Aldosterone suppression on contralateral adrenal during adrenal vein sampling does not predict blood pressure response after adrenalectomy.. THE JOURNAL OF CLINICAL ENDOCRINOLOGY AND METABOLISM. 99 (11) pp: 4158-4166.  
DOI: 10.1210/jc.2014-2345

The publisher's version is available at:  
<http://press.endocrine.org/doi/10.1210/jc.2014-2345>

When citing, please refer to the published version.

Link to this full text:  
<http://hdl.handle.net/2318/154962>

## **Aldosterone Suppression on Contralateral Adrenal During Adrenal Vein Sampling Does Not Predict Blood Pressure Response After Adrenalectomy**

Silvia Monticone\*, Fumitoshi Satoh\*, Andrea Viola§, Evelyn Fischer§, Oliver Vonend, Giampaolo Bernini, Barbara Lucatello, Marcus Quinkler, Vanessa Ronconi, Ryo Morimoto, Masataka Kudo, Christoph Degenhart, Xing Gao, Davide Carrara, Holger S. Willenberg, Denis Rossato, Giulio Mengozzi, Anna Riester, Enrico Paci, Yoshitsugu Iwakura, Jacopo Burrello, Mauro Maccario, Gilberta Giacchetti, Franco Veglio, Sadayoshi Ito, Martin Reincke#, and Paolo Mulatero#

Address all correspondence and requests for reprints to: Silvia Monticone, MD, and Paolo Mulatero, MD, Division of Internal Medicine and Hypertension, Department of Medical Sciences, University of Torino, Via Genova 3, 10126 Torino, Italy. E-mail: [paolo.mulatero@libero.it](mailto:paolo.mulatero@libero.it).

\*,§, and # denotes equal contribution for S.M. and F.S., A.V. and E.F., and M.R. and P.M., respectively.

### **Abstract**

**Context:** Adrenal vein sampling (AVS) is the only reliable means to distinguish between aldosterone-producing adenoma and bilateral adrenal hyperplasia, the two most common subtypes of primary aldosteronism (PA). AVS protocols are not standardized and vary widely between centers.

**Objective:** The objective of the study was to retrospectively investigate whether the presence of contralateral adrenal (CL) suppression of aldosterone secretion was associated with improved postoperative outcomes in patients who underwent unilateral adrenalectomy for PA.

**Setting:** The study was carried out in eight different referral centers in Italy, Germany, and Japan.

**Patients:** From 585 consecutive AVS in patients with confirmed PA, 234 procedures met the inclusion criteria and were used for the subsequent analyses.

**Results:** Overall, 82% of patients displayed contralateral suppression. This percentage was significantly higher in ACTH stimulated compared with basal procedures (90% vs 77%). The CL ratio was inversely correlated with the aldosterone level at diagnosis and, among AVS parameters, with the lateralization index ( $P = .02$  and  $P = .01$ , respectively). The absence of contralateral suppression was not associated with a lower rate of response to adrenalectomy in terms of both clinical and biochemical parameters, and patients with CL suppression underwent a significantly larger reduction in the aldosterone levels after adrenalectomy.

**Conclusions:** For patients with lateralizing indices of greater than 4 (which comprised the great majority of subjects in this study), CL suppression should not be required to refer patients to adrenalectomy because it is not associated with a larger blood pressure reduction after surgery and might exclude patients from curative surgery.

## Introduction

Primary aldosteronism (PA), the most common form of secondary hypertension, is a heterogeneous group of disorders characterized by autonomous aldosterone secretion and concomitant suppression of the renin-angiotensin system.

Bilateral adrenal hyperplasia (BAH) and aldosterone-producing adenoma (APA) together account for more than 90% of all PA cases, whereas rarer PA subtypes are unilateral adrenal hyperplasia, aldosterone-producing adrenal carcinoma, and the familial forms (1).

The diagnosis of PA is of particular importance because it has been extensively demonstrated that PA patients exhibit a higher rate of cardio- and cerebrovascular complications (2, 3), target organ damage (4), and metabolic syndrome (5) compared with essential hypertensives with a similar risk profile.

As recommended by The Endocrine Society guidelines, the diagnosis of PA includes screening, confirmatory/exclusion testing, and subtype differentiation (6).

The last step is essential to assign patients to the appropriate therapy of either adrenalectomy for unilateral disease or mineralocorticoid receptor antagonists for patients with BAH. Subtype differentiation requires adrenal computed tomography (CT) scanning to rule out malignancy, but adrenal vein sampling (AVS) has emerged as the only reliable means, in terms of sensitivity and specificity, to preoperatively differentiate between unilateral and bilateral PA (6).

AVS is a technically challenging and costly procedure that requires experienced and dedicated radiologists (7). During AVS, blood samples are collected from the adrenal veins and the inferior vena cava or from a peripheral vein. Because adrenal veins are small, the blood sample is frequently obtained near the orifice of the vein and may be diluted with nonadrenal vein blood, introducing an error in the measurement of aldosterone (A) levels. The simultaneous measurement of cortisol (C) concentrations corrects for this dilution and the ratio between cortisol levels in the adrenal veins and in the inferior vena cava [selectivity index (SI)] is a measure of the adequacy of cannulation. However, protocols for AVS are not universally standardized (and vary both in terms of procedures and stimulation), and the optimal criteria to define adequacy of cannulation and lateralization of aldosterone production remain controversial, with some centers using more permissive criteria and others using more restrictive ones (8–10).

Moreover, in addition to the SI and lateralization index (LI) (Supplemental Table 1), some authors have emphasized the importance and usefulness of contralateral (CL) adrenal suppression (defined as the A to C ratio of the nondominant adrenal vein over the A to C ratio of the inferior vena cava  $< 1$ ) (11, 12) to identify the source of aldosterone production, and currently many centers require this additional criterion together with a defined LI to suggest adrenalectomy to a PA patient (13, 14). Recently two groups published reviews on AVS criteria and both noted a lack of evidence either to support or not the use of CL suppression as an additional criteria for adrenalectomy (15, 16). By contrast, there is agreement among authors that the nondominant A to C ratio can be used to help the interpretation of suboptimal studies in which only the nondominant adrenal vein is cannulated (17, 18).

After adrenalectomy, hypertension is cured in approximately 50% of patients with APA (range 33%–70%) (19), whereas the remaining patients display a significant reduction of the blood pressure (BP) levels and number of antihypertensive drugs. Several factors have been reported to be associated with BP response after adrenalectomy (20–22), but so far no clinical and outcome data are available on the role of the CL suppression to support clinicians on the decision making to address patients to surgery and in particular in predicting a larger response to adrenalectomy.

In the current multicenter study, we assessed whether the presence or absence of contralateral adrenal suppression has an impact on the postoperative clinical and biochemical parameters in a large cohort of 234 patients who underwent unilateral adrenalectomy for PA.

## Materials and Methods

### Patient selection

This retrospective study was carried out in eight different referral centers in Italy, Germany, and Japan: the Divisions of Internal Medicine and Hypertension (Torino1) and the Division of Endocrinology (Torino2), University of Torino; the Division of Endocrinology, Polytechnic University of Marche, the Division of Internal Medicine, University of Pisa; the Division of Clinical Endocrinology, University Hospital Charité, Berlin; the Department of Nephrology and the Department of Endocrinology and Diabetology, University Hospital Düsseldorf; the Medizinische Klinik und Poliklinik IV, University Hospital, Munich; and the Division of Nephrology, Endocrinology, and Vascular Medicine, Tohoku University Graduate School of Medicine.

The data were collected within our prospective registries in the different centers, and all patients gave written informed consent for the use of data including AVS. We obtained approval from the local ethics committees to use these data for our retrospective study.

Case detection and subtype differentiation were performed according to the Japan Endocrine Society (23) and The Endocrine Society guidelines (6) as described before (9, 24) (further details are available in the Supplemental Data). Differences in genetic background, salt intake, referral bias, and screening and confirmation strategies could be responsible for some of the differences between groups of patients.

Briefly, patients were screened for PA using the aldosterone to renin (or to plasma renin activity) ratio (ARR) after withdrawal of interfering medications; patients remained in the same therapy during the entire diagnostic workup (from screening to AVS). Diagnosis of PA was confirmed with iv saline loading or the fludrocortisone-suppression tests (Italian and German Units) or captopril challenge test (Sendai-Japanese Unit). All patients with confirmed PA underwent CT scanning and AVS.

AVS was performed under basal conditions in Torino2, Pisa, Dusseldorf, Munich, and Berlin. The Sendai unit performed the AVS both under basal condition and after iv bolus injection of cosyntropin (0.25 mg): this latter was used for final decision on subtype differentiation; continuous cosyntropin infusion (50 µg/h, started 30 min before sampling and continued throughout the procedure) was used in Ancona. In Torino1, continuous cosyntropin (50 µg/h) infusion was used when the procedure was performed late in the morning or when a patient, at risk for an allergic reaction to the contrast, was pretreated with dexamethasone. AVS procedures were performed as described previously (9, 24). All centers performed AVS in the sequential way; centers that do not use cosyntropin, when collecting blood from an adrenal vein, also collect blood from a peripheral vein to rule out oscillations in aldosterone and cortisol secretion.

The SI was defined as cortisol/adrenal vein/cortisol/peripheral vein and LI as aldosterone/cortisol/adrenal vein/aldosterone/cortisol/CL adrenal vein. CL suppression was defined as aldosterone/cortisol/nondominant adrenal vein/aldosterone/cortisol/peripheral vein less than 1, ipsilateral (IL) ratio was defined as aldosterone/cortisol/dominant adrenal vein/aldosterone/cortisol/peripheral vein (Supplemental Table 1).

We considered AVS studies eligible for inclusion if they met the following criteria: 1) SI of 2 or greater for basal studies and 5 or greater for ACTH-stimulated studies; 2) lateralization index 3 or greater [in all centers, an LI > 4 is considered necessary to define lateralization and to suggest adrenalectomy, but patients with LI between 3 and 4 are operated in some cases (in all cases in the Sendai unit)], and therefore, they were included in the analysis; and 3) clinical and hormonal follow-up of at least 12 months after adrenalectomy.

All patients underwent unilateral adrenalectomy and diagnosis of APA was confirmed after surgery by histological examination, cure or significant amelioration of hypertension, normokalemia, normal ARR and low aldosterone levels, and/or the normal suppressibility of aldosterone.

### Statistical analyses

IBM SPSS Statistics 19 (SPSS Inc) was used for the statistical analyses. The data are presented as mean ± SD or median (25th to 75th percentile). Data were analyzed with the Kolmogorov-Smirnov and Shapiro-Wilk

tests to determine their distributions. Statistical significance between groups was calculated in the normally distributed data by a Student t test for independent samples or a one-way ANOVA and in not normally distributed data by the Kruskal-Wallis, the Mann-Whitney U tests, and a one-way ANOVA on ranks, using Bonferroni corrections for multiple comparisons. The  $\chi^2$  test of the Fisher exact test was used for qualitative variables. A probability value of  $P < .05$  was considered statistically significant.

## Results

Among eight different referral centers in Italy, Germany, and Japan, a total of 585 consecutive AVS were evaluated: 256 patients were adrenalectomized on the basis of the local criteria and 234 AVS procedures met the predefined inclusion criteria and were used for the subsequent analyses (Figure 1 and Supplemental Table 2).

Clinical and biochemical parameters of patients participating in the study are summarized in Table 1. The main demographic and clinical characteristics were suppressed plasma renin activity (PRA) [or plasma renin concentration (PRC)], elevated plasma aldosterone levels, and hypertension, according with the typical phenotype of PA patients. Hypokalemia (defined as serum  $K^+ < 3.5$  mmol/L on at least one occasion on the patient history) was present in 71% of the patients. Patients from the Italian units displayed a more severe phenotype with higher systolic and diastolic BP, serum aldosterone, and lower potassium levels compared with the Japanese and German units. Clinical and biochemical parameters after adrenalectomy are summarized in Table 2.

Overall, 192 of 234 of AVS studies (82%) displayed CL suppression, with no statistically significant differences among centers. ACTH stimulation was associated with a significant ( $P = .014$ ) increase in the percentage of AVS showing CL suppression, compared with basal studies (90% vs 77%). CL suppression was slightly but not significantly higher in studies with bolus ACTH vs basal studies (88% vs 77%,  $P = .07$ ), significantly higher in studies with continuous ACTH infusion vs basal (91% vs 77%,  $P = .02$ ) and similar in the two types of ACTH stimulation (88% vs 91%,  $P = .44$ ).

We also evaluated the presence of absolute contralateral aldosterone suppression by calculating the ratio of aldosterone levels in the nondominant adrenal vein with the aldosterone values in the peripheral vein. Despite the potential confounding effect of dilution errors during adrenal sampling, this ratio could give an idea of the real contralateral suppression of aldosterone secretion. We observed that only 25 of 234 patients (10.6%) had aldosterone values in the nondominant adrenal vein inferior or equal to the values in the peripheral vein. This finding was more frequent in unstimulated AVS (23 of 143) than in cosyntropin-stimulated procedures (2 of 91).

Ninety-five of 234 patients underwent a saline load after adrenalectomy, showing normal suppressibility of aldosterone secretion. We did not observe a difference of aldosterone levels after saline load performed after adrenalectomy between the patients with and without CL suppression [3.3 (2–4.8) vs 3.9 (2.6–6) ng/dL,  $P = .2$ ].

Fifty-three AVS samples were performed both under basal conditions and after ACTH stimulation. In 45 of 53 cases (85%), the presence (43 cases) or absence (two cases) of CL suppression was maintained under both conditions of sampling. In three cases CL suppression was present under basal conditions but not after ACTH, whereas in five cases the CL suppression was absent in basal conditions but observed after ACTH stimulation. More in detail, nine AVS (two in Ancona and seven in Torino<sup>1</sup>) were performed both under basal conditions and after continuous cosyntropin infusion: in seven of nine cases (78%), the presence (six cases) or not (one case) of CL suppression was maintained between the two AVS samplings. In the other two of nine cases, the CL suppression was absent in basal conditions and present after cosyntropin infusion. Forty-four AVS (all in the Sendai unit) were performed both in basal conditions and after bolus ACTH: in 38 of 44 cases (86%), the presence (37 cases) or not (one case) of CL suppression was maintained between the

two AVS samplings. In the other 6 of 44 cases, the CL suppression was discordant (in three cases it was present in basal condition and not after ACTH and in three cases it was absent in the basal condition and present after ACTH). Interestingly, patients with a unilateral single nodule did not display CL suppression more frequently or a higher lateralization ratio in comparison with multiple nodules (data not shown).

We observed a statistically significant inverse correlation between the CL ratio and the serum aldosterone levels at diagnosis ( $R^2 = 0.04$ ;  $P = .02$ ) but not between the CL ratio and systolic BP (SBP), potassium, and age at diagnosis of PA. Moreover, a regression analysis showed that serum aldosterone levels but not SBP and potassium levels predicted CL suppression during AVS.

Among the AVS parameters, the CL ratio inversely correlated with lateralization index ( $R^2 = 0.028$ ;  $P = .01$ ) and patients with an LI greater than 4 displayed a significantly higher proportion of CL suppression compared with patients with LI between 3 and 4 (184 of 211, 87% vs 8 of 23, 35%;  $P < .001$ ).

We compared the clinical and biochemical preoperative parameters between the two groups: patients with CL suppression had higher plasma aldosterone levels at diagnosis than patients who did not have suppressed CL aldosterone secretion, whereas we did not observe significant differences in systolic and diastolic BP, potassium, PRA, PRC, and the number of the class of drugs used to treat the patients (Table 3). Both in basal and ACTH-stimulated conditions ( $P < .001$  and  $P = .004$ , respectively), the median LI was significantly higher in patients who showed contralateral adrenal suppression (Figure 2), whereas the median ipsilateral ratio was not significantly different between the two groups (data not shown).

Subsequently, to investigate whether the presence of CL suppression was correlated with response to adrenalectomy, we analyzed the CL suppression status with regard to the patient's clinical and biochemical postadrenalectomy parameters, as shown in Table 4.

No differences were observed between the two groups for the main clinical and biochemical parameters [SBP, diastolic BP (DBP), aldosterone, PRA, PRC, K<sup>+</sup>, number of drugs, reduction of BP levels, and the number of classes of drugs assumed by the patients], but patients with CL suppression underwent a significantly larger reduction in aldosterone levels after adrenalectomy.

We also sought to ascertain whether the IL ratio greater than 2 was associated with postadrenalectomy clinical and biochemical parameters, including BP reduction. In some units an IL ratio greater than 2 is considered to indicate adrenalectomy if the CL ratio is inferior to 1 (8, 21). The median IL ratio was 4.1 (2.2–8.0) under basal condition and 4.3 (2.8–6.4) in ACTH-stimulated studies ( $P = .452$ ). Overall 187 of 234 patients (80%) displayed an IL ratio greater than 2. We did not observe a greater reduction in SBP in patients displaying an IL ratio greater than 2 compared with patients with an IL ratio less than 2 ( $34 \pm 2$  mm Hg vs  $37 \pm 4$  mm Hg).

Finally, despite a greater reduction in aldosterone levels for patients with LI greater than 4, we could not observe a difference in BP levels and number of drugs reduction between patients with an LI greater than 4 and patients with LI between 3 and 4. These data should be taken cautiously because the number of patients in the second group was relatively small, and patients with a LI between 3 and 4 are not systematically adrenalectomized in our units.

## Discussion

AVS is the only reliable approach to distinguish between unilateral and bilateral PA preoperatively and therefore to address patients to the most effective therapy, as indicated by The Endocrine Society and the Japanese Endocrine Society Guidelines (6, 23). Subtype differentiation is of fundamental importance because individuals with APA can be cured by unilateral laparoscopic adrenalectomy, whereas patients with BAH benefit from targeted medical therapy with mineralocorticoid receptor antagonists. Unfortunately, AVS protocols vary widely, and agreement is lacking on the best criteria indicating successful cannulation and lateralization of aldosterone production (15, 16). In some centers, to define the source of aldosterone

production, in addition to the classical selectivity and lateralization indices, also the contralateral suppression, which reflects inhibition of aldosterone secretion in the adrenal gland contralateral to an adenoma during AVS, is taken into account (15, 16). There is, however, general agreement among authors that the nondominant aldosterone to cortisol ratio cannot be relied on to predict APA because up to 32% of BAH patients can have a nondominant adrenal vein aldosterone to cortisol ratio that is less than the ratio in the peripheral vein, but it can be used to help the interpretation of suboptimal studies (18). For example, in a patient in whom only one adrenal vein is successfully cannulated, suppression on that side, taken in conjunction with particularly high serum aldosterone levels and the presence of hypokalemia (which is more common in unilateral vs bilateral PA) and a discrete mass lesion in the opposite gland on CT, may be sufficient to warrant consideration for removal of the lesion-containing gland.

To date, whether the absence of contralateral suppression might have an impact on postoperative clinical and biochemical parameters, thus affecting BP response to adrenalectomy, has never been investigated (15, 16). If such an impact proved to be substantial, it would argue for CL suppression to become a mandatory (or at least highly recommended) criterion in the selection of patients for unilateral adrenalectomy, in addition to the level of the LI.

The results of the present multicentric study show a high prevalence of contralateral suppression in our group of patients who underwent unilateral adrenalectomy, in agreement with previous findings on a smaller series of patients (12, 18). Interestingly, the number of AVS displaying CL suppression was slightly but significantly higher in cosyntropin-stimulated procedures compared with basal ones. This may be due to a high percentage of ACTH-sensitive APA in our group of patients. Furthermore, in the subgroup of patients who underwent AVS both in basal and stimulated conditions, ACTH administration did not reduce the percentage patients with contralateral adrenal suppression. These results suggest that cosyntropin administration only rarely stimulates aldosterone production from the contralateral gland to an adenoma sufficiently to represent a confounding factor in the interpretation of the AVS results, which is in agreement with another recent study (9). It has been shown that cosyntropin stimulation may help centers with a lower success rate in obtaining diagnostic AVS (9): our results confirm that ACTH can be used with only minimal risk of reducing gradients between the dominant and nondominant adrenal due to the stimulation of the contralateral adrenal to the gland with an APA.

Overall, the absence of contralateral suppression was not associated with a lower rate of response to adrenalectomy in terms of both clinical and biochemical parameters. Thus, CL suppression should not be required for patients with a high LI before being addressed to unilateral adrenalectomy. It should be noted that our results apply only to patients with an LI greater than 4, which constituted the great majority of patients recruited in this study, and that is the cutoff used in most of the units. Although our results suggest that patients with an LI between 3 and 4 probably do not require CL suppression to warrant adrenalectomy, this observation should be taken cautiously because only a small proportion of these patients were recruited here and because we could have had a selection bias for these patients because they do not systematically undergo adrenalectomy in our units. Certainly our data cannot be extended to patients with an LI less than 3 that are operated in some units (25).

In our patient cohort, the presence of CL suppression was associated with higher aldosterone levels: in fact, patients with CL suppression displayed significantly higher LI, and the CL ratio was inversely related with serum aldosterone levels at diagnosis. Therefore, patients with CL suppression may be affected by a more severe aldosterone excess. On the other hand, it is conceivable that patients with absent CL suppression might have an autonomous contralateral aldosterone production that after adrenalectomy, despite biochemical cure of hyperaldosteronism, could theoretically be responsible for some (subclinical) deregulated hormonal production or recurrence in later years (26). However, the benefit of adrenalectomy was remarkable and equally evident in both groups of patients, with or without CL suppression. It also



should be noted that a percentage of patients with bilateral disease has been shown to benefit of unilateral adrenalectomy (27). Furthermore, the presence of background essential hypertension and differences in salt intake and aldosterone sensitivity (28, 29) between patients may contribute to confound the interpretation of the results of BP response to adrenalectomy in the present study.

It should be underlined that the concept of CL suppression is an operative definition that includes a relative aldosterone production in comparison with cortisol production in the adrenal gland and does not reflect a complete suppression of aldosterone secretion in the contralateral gland: in fact, the absolute aldosterone levels from the contralateral adrenal gland to an APA are almost always higher than the aldosterone levels in a peripheral vein (90% in the present study).

Interestingly, the presence of marked hyperkalemia after adrenalectomy for APA is a very rare observation, indicating that a sustained contralateral suppression of aldosterone secretion is not frequent in patients with APA (30).

This is coherent with the observation that in the adrenal cortex surrounding an APA, the zona glomerulosa is often hyperplastic and contains nodules that in many cases display aldosterone synthase activity (called aldosterone producing cell cluster) (31). These findings are reminiscent of the observation that rats under a long-term high-sodium diet display a decrease in the width of the zona glomerulosa and the number of cells that express the aldosterone synthase, but there is always a proportion of cells with high aldosterone synthase expression (32). These findings led some authors to hypothesize that a significant proportion of patients with PA and unilateral adrenal disease are in reality affected by a bilateral hyperplasia in which subsequently a nodule became dominant such as in a multinodular thyroid goiter (33). Recent studies demonstrated that somatic mutations in different genes are present in APAs, which have not been observed in BAH patients (1, 34, 35); however, even in patients with APA carrying somatic mutations, a lack of contralateral suppression has been observed during AVS: this is consistent with a two (or multiple)-hit hypothesis suggesting that two or more alterations are necessary for the formation of an APA (36).

We speculate that CL suppression, LI, and aldosterone levels after an adrenalectomy are all related to the deregulated aldosterone production; by contrast, other indices of disease severity are affected by other factors [salt intake, familial history of hypertension, underlying essential hypertension, somatic mutations in *KCNJ5*, *ATP1A1*, *ATB2B3*, and *CACNA1D* affecting aldosterone secretion (1, 34, 37)], and last but not least, genetic determinants of aldosterone sensitivity (28, 29) that could not be evaluated in the present study.

This study has some potential limitations: 1) the retrospective nature of the study; 2) a different level of referral bias due to the type of PA patients referred to our Hypertension Units that may not be entirely representative of the general PA population; 3) differences in diagnostic protocols among centers that may contribute to a nonuniform composition of the patients included in the present study; and 4) the fact that not all patients underwent a postsurgery suppression study: however, in all cases patients displayed, after adrenalectomy, a significant reduction of BP, normalization of hypokalemia when present, and normal ARR; furthermore, in most cases patients after adrenalectomy displayed aldosterone levels that were so low that a suppression test was considered unnecessary.

AVS is a complex and technically demanding procedure, but also its interpretation requires experience and careful evaluation. Unfortunately, despite the fact that it is considered indispensable for addressing a patient with suspicious unilateral adrenal aldosterone overproduction to surgery, criteria for interpreting the results of the sampling are not universally accepted. It has been recently demonstrated that the procedure is equally effective whether or not cosyntropin stimulation is used during the procedure (9) as long as the selectivity indices demonstrating cannulation are strict enough to ensure reproducible diagnoses (8).

In the present study, we add a further criterion to help the clinician in the prediction of the efficacy of adrenalectomy in terms of BP response. For patients with an LI greater than 4, the presence of CL suppression should not be considered an indispensable parameter to address patients to adrenalectomy because it is not associated with a larger BP reduction after surgery and would exclude approximately 20% of patients from curative surgery. Whether this holds true for patients with an LI less than 4 remains to be established and is worthy of further study. We believe that the results of this study represent further progress in the standardization of AVS interpretation in patients with primary aldosteronism.

**Abbreviations:**

A	aldosterone
APA	aldosterone-producing adenoma
ARR	aldosterone to renin ratio
AVS	adrenal vein sampling
BAH	bilateral adrenal hyperplasia
BP	blood pressure
C	cortisol
CL	contralateral
CT	computed tomography
DBP	diastolic BP
IL	ipsilateral
LI	lateralization index
PA	primary aldosteronism
PRA	plasma renin activity
PRC	plasma renin concentration
SBP	systolic BP
SI	selectivity index.

## References

1. Mulatero P, Monticone S, Rainey WE, Veglio F, Williams TA. Role of KCNJ5 in familial and sporadic primary aldosteronism. *Nat Rev Endocrinol*. 2013;9:104–112.
2. Milliez P, Girerd X, Plouin PF, Blacher J, Safar ME, Mourad JJ. Evidence for an increased rate of cardiovascular events in patients with primary aldosteronism. *J Am Coll Cardiol*. 2005;45:1243–1248.
3. Mulatero P, Monticone S, Bertello C, et al. Long-term cardio- and cerebro-vascular events in patients with primary aldosteronism. *J Clin Endocrinol Metab*. 2013;98:4826–4833.
4. Mulatero P, Milan A, Williams TA, Veglio F. Mineralocorticoid receptor blockade in the protection of target organ damage. *Cardiovasc Hematol Agents Med Chem*. 2006;4:75–91.
5. Fallo F, Pilon C, Urbanet R. Primary aldosteronism and metabolic syndrome. *Horm Metab Res*. 2012;44:208–214.
6. Funder JW, Carey RM, Fardella C, et al. The Endocrine Society. Case detection, diagnosis, and treatment of patients with primary aldosteronism: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab*. 2008;93:3266–3281.
7. Mulatero P, Bertello C, Rossato D, et al. Roles of clinical criteria, computed tomography scan, and adrenal vein sampling in differential diagnosis of primary aldosteronism subtypes. *J Clin Endocrinol Metab*. 2008;93:1366–1371.
8. Mulatero P, Bertello C, Sukor N, et al. Impact of different diagnostic criteria during adrenal vein sampling on reproducibility of subtype diagnosis in patients with primary aldosteronism. *Hypertension*. 2010;55:667–673. , Google Scholar CrossRef, Medline, ISI
9. Monticone S, Satoh F, Giacchetti G, et al. Effect of adrenocorticotrophic hormone stimulation during adrenal vein sampling in primary aldosteronism. *Hypertension*. 2012;59:840–846.
10. Rossi GP, Barisa M, Allolio B, et al. The Adrenal Vein Sampling International Study (AVIS) for identifying the major subtypes of primary aldosteronism. *J Clin Endocrinol Metab*. 2012;97:1606–1614.
11. Doppman JL, Gill JR Jr, Miller DL, et al. Distinction between hyperaldosteronism due to bilateral hyperplasia and unilateral aldosteronoma: reliability of CT. *Radiology*. 1992;184:677–682.
12. Espiner EA, Ross DG, Yandle TG, Richards AM, Hunt PJ. Predicting surgically remedial primary aldosteronism: role of adrenal scanning, posture testing, and adrenal vein sampling. *J Clin Endocrinol Metab*. 2003;88:3637–3644.
13. Ahmed AH, Cowley D, Wolley M, et al. Seated saline suppression testing for the diagnosis of primary aldosteronism—a preliminary study. *J Clin Endocrinol Metab*. 2014;99(8):2745–2753.
14. Kempers MJ, Lenders JW, van Outheusden L, et al. Systematic review: diagnostic procedures to differentiate unilateral from bilateral adrenal abnormality in primary aldosteronism. *Ann Intern Med*. 2009;151:329–337.
15. Monticone S, Viola A, Rossato D, et al. Adrenal vein sampling in primary aldosteronism: towards a standardised protocol [published online May 12, 2014]. *Lancet Diabetes Endocrinol*. doi:10.1016/S2213-8587(14)70069-5.
16. Rossi GP, Auchus RJ, Brown M, et al. An expert consensus statement on use of adrenal vein sampling for the subtyping of primary aldosteronism. *Hypertension*. 2014;63:151–160.
17. Auchus RJ, Wians FH Jr, Anderson ME, et al. What we still do not know about adrenal vein sampling for primary aldosteronism. *Horm Metab Res*. 2010;42:411–415.
18. Young WF, Stanson AW, Thompson GB, Grant CS, Farley DR, van Heerden JA. Role for adrenal venous sampling in primary aldosteronism. *Surgery*. 2004;136:1227–1235.
19. Mulatero P, Stowasser M, Loh KC, et al. Increased diagnosis of primary aldosteronism, including surgically correctable forms, in centers from five continents. *J Clin Endocrinol Metab*. 2004;89:1045–1050.

20. Sawka AM, Young WF, Thompson GB, et al. Primary aldosteronism: factors associated with normalization of blood pressure after surgery. *Ann Intern Med.* 2001;135:258–261.
21. Celen O, O'Brien MJ, Melby JC, Beazley RM. Factors influencing outcome of surgery for primary aldosteronism. *Arch Surg.* 1996;131:646–650.
22. Giacchetti G, Ronconi V, Rilli S, Guerrieri M, Turchi F, Boscaro M. Small tumor size as favorable prognostic factor after adrenalectomy in Conn's adenoma. *Eur J Endocrinol.* 2009;160:639–646.
23. Nishikawa T, Omura M, Satoh F, et al. Task Force Committee on Primary Aldosteronism, The Japan Endocrine Society. Guidelines for the diagnosis and treatment of primary aldosteronism—the Japan Endocrine Society 2009. *Endocr J.* 2011;58:711–721.
24. Vonend O, Ockenfels N, Gao X, et al. German Conn's Registry. Adrenal venous sampling: evaluation of the German Conn's registry. *Hypertension.* 2011;57:990–995.
25. Stowasser M, Gordon RD, Gunasekera TG, et al. High rate of detection of primary aldosteronism, including surgically treatable forms, after 'non-selective' screening of hypertensive patients. *J Hypertens.* 2003;21:2149–2157.
26. Rutherford JC, Taylor WL, Stowasser M, Gordon RD. Success of surgery for primary aldosteronism judged by residual autonomous aldosterone production. *World J Surg.* 1998;22:1243–1245.
27. Sukor N, Gordon RD, Ku YK, Jones M, Stowasser M. Role of unilateral adrenalectomy in bilateral primary aldosteronism: a 22-year single center experience. *J Clin Endocrinol Metab.* 2009;94:2437–2445.
28. Tu W, Eckert GJ, Hannon TS, et al. Racial differences in sensitivity of blood pressure to aldosterone. *Hypertension.* 63:1212–1218.
29. Funder JW. Sensitivity to aldosterone: plasma levels are not the full story. *Hypertension.* 2014;63:1168–1170.
30. Fischer E, Hanslik G, Pallauf A, et al. Prolonged zona glomerulosa insufficiency causing hyperkalemia in primary aldosteronism after adrenalectomy. *J Clin Endocrinol Metab.* 2012;97:3965–3973.
31. Nishimoto K, Nakagawa K, Li D, et al. Adrenocortical zonation in humans under normal and pathological conditions. *J Clin Endocrinol Metab.* 2010;95:2296–2305.
32. Romero DG, Yanes LL, de Rodriguez AF, et al. Disabled-2 is expressed in adrenal zona glomerulosa and is involved in aldosterone secretion. *Endocrinology.* 2007;148:2644–2652.
33. Gomez-Sanchez CE, Rossi GP, Fallo F, Mannelli M. Progress in primary aldosteronism: present challenges and perspectives. *Horm Metab Res.* 2010;42:374–381.
34. Williams TA, Monticone S, Schack VR, et al. Somatic ATP1A1, ATP2B3, and KCNJ5 mutations in aldosterone-producing adenomas. *Hypertension.* 2014;63:188–195.
35. Scholl UI, Goh G, Stölting G, et al. Somatic and germline CACNA1D calcium channel mutations in aldosterone-producing adenomas and primary aldosteronism. *Nat Genet.* 2013;45:1050–1054.
36. Gomez-Sanchez CE, Gomez-Sanchez EP. Mutations of the potassium channel KCNJ5 causing aldosterone-producing adenomas: one or two hits? *Hypertension.* 2012;59:196–197.
37. Fernandes-Rosa FL, Williams TA, Riester A, et al. Genetic spectrum and clinical correlates of somatic mutations in aldosterone-producing adenoma. *Hypertension.* 2014;64:354–361.

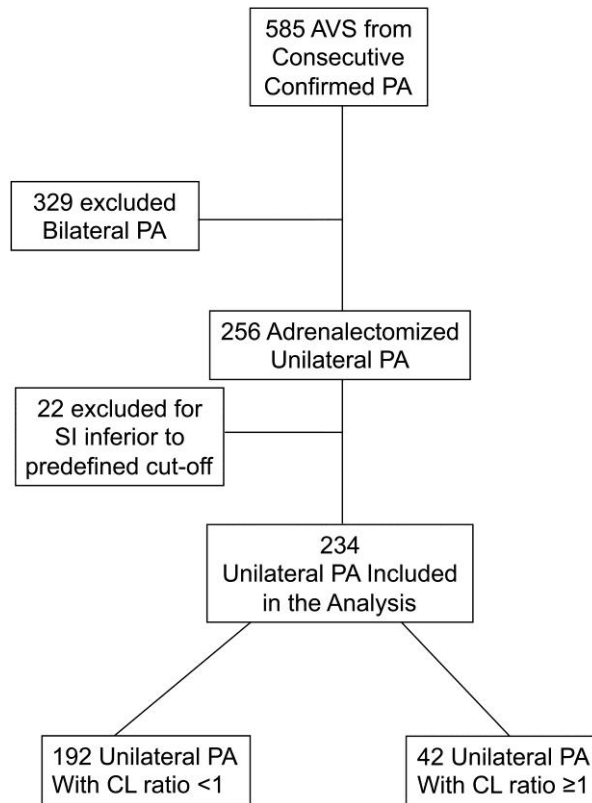


Figure 1: Flow chart of the study.

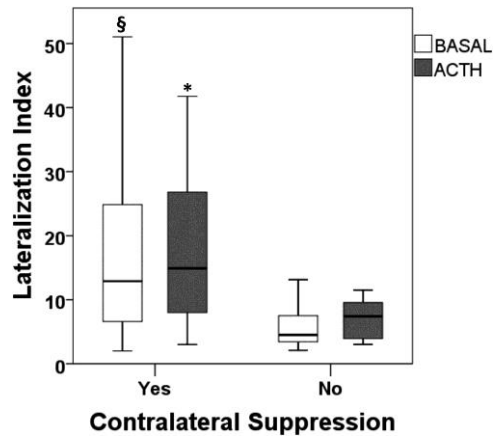


Figure 2. Box plots of the LI distribution between patients with and without contralateral adrenal suppression in basal (white) and ACTH (gray)-stimulated studies. The horizontal line indicates the median, and the box and the bar represent the 25th to 75th and fifth to 95th percentiles, respectively. \*,  $P = .004$ ; §,  $P < .001$ .

Table 1. Clinical and Biochemical Parameters of APA Patients Included in the Analysis

	Germany	Italy	Japan	Overall P Value	Germany vs Italy	Germany vs Japan	Italy vs Japan
Age at diagnosis, y	52 ± 1.4	48 ± 1.0	53 ± 1.9	.04	.10	.45	.02
Sex, male/female	33/26	67/64	23/21	.83	.32	.43	.52
Pre PRA, ng/mL · h	0.2 [0.2–0.5]	0.2 [0.14–0.4]	0.1 [0.1–0.3]	.003	1.00	.15	.007
Pre PRC, mUL	2.8 [1.6–5–1]	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
Pre aldosterone, ng/dL	22.3 [14.5–38.1]	45.3 [33.3–58]	39.9 [26.9–60.9]	<.001	<.001	.003	.47
Pre K+, mEq/L	3.1 ± 0.07	3.0 ± 0.06	3.4 ± 0.11	.002	.46	.012	<.001
Pre SBP, mm Hg	152 ± 3	169 ± 2	155 ± 3	<.001	<.001	.57	.001
Pre DBP, mm Hg	91 ± 2	103 ± 1	90 ± 2	<.001	<.001	.77	<.001
Nodule, mm	15 [11.7–19]	15 [10–20]	15 [10–19]	.73	.67	.48	.76
Pre drugs, n	3 ± 0.2	2.4 ± 0.1	2.2 ± 0.3	.013	.01	.006	.39

Abbreviation: n.a., not available; Pre, preoperative. Nodule dimensions are obtained with CT scanning. Numbers are mean ± SD or median [25th to 75th percentiles].

**Table 2.** Clinical and Biochemical Postadrenalectomy Parameters of APA Patients Included in the Analysis

	Germany	Italy	Japan	Overall P Value	Germany vs Italy	Germany vs Japan	Italy vs Japan
Post PRA, ng/mL · h	1.6 [1.1–2.3]	2.1 [1.2–3.0]	1.9 [1–3.9]	.53	.27	.46	.59
Post PRC, mU/L	20 [11–50]	n.a.	n.a.	n.a	n.a.	n.a	n.a
Post aldosterone, ng/dL	5.4 [3.5–8.4]	12.9 [8.7–19.1]	9 [5.9–12]	<.001	<.001	.005	.004
Post K <sup>+</sup> , mEq/L	4.3 ± 0.06	4.7 ± 0.04	4.4 ± 0.05	<.001	<.001	.10	.001
Post SBP, mm Hg	129 ± 2	127 ± 1	127 ± 2	.55	.28	.45	.92
Post DBP, mm Hg	82 ± 1	81 ± 1	79 ± 1	.26	.43	.10	.24
ΔSBP, mm Hg	23 ± 3	41 ± 2	28 ± 3	<.001	<.001	.35	.001
ΔDBP, mm Hg	9 ± 2	22 ± 1	11 ± 2	<.001	<.001	.51	<.001
Postoperative drugs, n	1.6 ± 1.5	1.1 ± 1.2	1.5 ± 1.3	.019	.014	.85	.045

Abbreviations: ΔDBP, diastolic blood pressure reduction after adrenalectomy; n.a., not available; ΔSBP, systolic blood pressure reduction after adrenalectomy. Data were obtained between 12 and 18 months after adrenalectomy.



Table 3. Comparison of Preoperative Parameters According to Contralateral Suppression Status

Pre Adrenalectomy	Contralateral Suppression: Yes (n = 192)	Contralateral Suppression: No (n = 42)	P value
Parameters			
PRA, ng/mL · h	0.2 [0.1–0.3]	0.2 [0.1–0.3]	.6
PRC, mU/L	3.0 [2.0–5.1]	2.4 [1.3–13.5]	.8
Aldosterone, ng/dL	41.5 [26.2–58.0]	33.3 [22.3–48.8]	.011
K <sup>+</sup> , mEq/L	3.1 ± 0.05	2.9 ± 0.1	.2
SBP, mm Hg	161 ± 2	167 ± 4	.2
DBP, mm Hg	96 ± 1	101 ± 3	.1
Drugs, n	2.5 ± 0.1	2.6 ± 0.3	.9

P < 0.05 was considered statistically significant.

Table 4. Comparison of Postoperative Parameters According to Contralateral Suppression Status

Postadrenalectomy Parameters	Contralateral Suppression: Yes (n = 192)	Contralateral Suppression: No (n = 42)	P Value
SBP, mm Hg	128 ± 11	127 ± 14	.6 (n.s.)
DBP, mm Hg	80 ± 8	82 ± 8	.2 (n.s.)
Aldosterone, ng/dL	9.80 [5.65–17.40]	9.50 [6.90–20.00]	.5 (n.s.)
K+, mEq/L	4.53 ± 0.4	4.52 ± 0.4	.9 (n.s.)
PRA, ng/mL · h	2.0 [1.20–3.08]	1.65 [1.09–3.08]	.6 (n.s.)
PRC, mU/L	18.5 [9.2–30.0]	26.8 [13.0–63.2]	.4 (n.s.)
Drugs, n	1.35 ± 1.4	1.38 ± 1.5	.9 (n.s.)
ΔSBP, mm Hg	33 ± 1.8	40 ± 4.0	.1 (n.s.)
ΔDBP, mm Hg	16 ± 1.0	19 ± 2.6	.2 (n.s.)
ΔAldosterone	28.6 ± 3.1	8.5 ± 9.9	.015
ΔDrugs, n	1.2 ± 0.1	1.2 ± 0.3	.8 (n.s.)
Number of cured, %	100 (52%)	22 (52%)	.9 (n.s.)

Abbreviations: ΔAldosterone, plasma aldosterone levels reduction after adrenalectomy; ΔDBP, diastolic blood pressure reduction after adrenalectomy; Δdrugs n, drugs number reduction after adrenalectomy; n.s., not significant; ΔSBP, systolic blood pressure reduction after adrenalectomy. P < 0.05 was considered statistically significant.