Update in Primary Aldosteronism

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Primary aldosteronism (PA) is a condition well worth detecting because it is a common cause of hypertension and is associated with excessive morbidity for the degree of hypertension and reduced quality of life, all of which can be abrogated with specific surgical or medical treatment. Recent years have seen an explosion in knowledge concerning the genetic bases of this disorder, and particularly of somatic mutations associated with aldosterone-producing adenomas and germline mutations causing rare familial forms, both involving genes encoding ion channels. Inroads have also been made into understanding molecular pathways that may be involved in the development of PA. With evidence continuing to mount for non-blood pressure-dependent adverse effects of aldosterone excess and for superior effects of specific over non-specific treatment, the need for accurate yet readily applicable and available diagnostic approaches and methodologies has become a matter of urgency. Advances in approaches to confirmatory testing, subtype differentiation, and assay methodology are helping to improve feasibility and reliability of the diagnostic workup for PA, and new treatment approaches are emerging. (J Clin Endocrinol Metab 100: 1–10, 2015)

Primary aldosteronism (PA) remains in a “golden era” in terms of clinical and research interest that has evolved over the past two to three decades with the realization that it is highly prevalent among hypertensive populations and, through non-blood pressure-dependent means, is associated with cardiovascular, renal, and psychological morbidity that appears to be excessive for the degree of hypertension. The last few years have seen ongoing, intense research activity into its causations, diagnostic workup, and management that appears unlikely to wane anytime soon. Perhaps the most exciting advances have been in understanding the pathophysiology of PA, and especially its genetic bases, opening doors to newer and better methods of diagnosis and treatment. Evidence that the adverse effects of aldosterone excess go way beyond merely causing hypertension has been further strengthened. Challenges to physicians keen to pursue the diagnosis of this condition, correctly determine the subtype, and offer optimal specific treatment have received considerable attention and, in some cases, have been met by the development of better diagnostic methodologies, more accurate assays, and new therapies. This report will review some of these advances.

The New Genetics of PA

It would be impossible to complete an update on PA without including the major recent developments that have been made in understanding the genetic bases of this disorder. In 2011, Choi et al (1) reported somatic mutations (G151R and L168R) in KCNJ5, encoding an inwardly rectifying potassium channel (Kir 3.4), in eight of 22 aldosterone-producing adenomas (APAs) removed from Swedish patients. In the same report, a third germline mutation (T158A) in KCNJ5 was identified in an American family with familial, florid PA with severe childhood-onset hypertension, hypokalemia, and markedly elevated aldosterone/renin ratio (ARR). Additional germline mutations were found in the patient’s parents, whose blood pressure was normal. The patient’s siblings had normal blood pressure, but three had hypokalemia and a subclinical increase in aldosterone levels. All family members had high potassium sensitivity and low plasma renin activity. The authors showed that the T158A mutation causes a decreased affinity of Kir 3.4 channels for ouabain, resulting in increased transepithelial potassium conductance and aldosterone production. Since these reports, several other studies have confirmed the association of mutations in KCNJ5 with PA, and have also identified mutations in other genes encoding potassium channels, including KCNJ2, KCNQ1, and KCNQ3. These findings have provided new insights into the pathophysiology of PA and have opened doors to newer and better methods of diagnosis and treatment.
doxosterone and hybrid steroid (18-hydroxy- and 18-oxo-cortisol) levels, which, unlike in glucocorticoid-remediable aldosteronism (familial hyperaldosteronism type I), was not glucocorticoid-suppressible. Resected adrenals (the hypertension could not be controlled with medical treatment, including spironolactone) were grossly enlarged and showed marked, diffuse hyperplasia of the zona fasciculata (2). Subsequently, several groups confirmed the presence of somatic KCNJ5 mutations with a prevalence of approximately 40% of APAs among Caucasian patients (3–6) and even higher in Japanese patients (7). Patients with APAs bearing these mutations have shown a female predominance and younger mean age than those with non-mutated tumors (1, 3–8), with a tendency to more severe PA (1, 5–7), unresponsiveness of plasma aldosterone to upright posture (4), and larger tumors (3, 4) composed predominantly of cells resembling zona fasciculata (9).

In vitro, reported KCNJ5 mutations are almost always located within the selectivity filter and associated with loss of selectivity of the mutated Kir 3.4 channels to potassium, with enhanced sodium conductance predisposing to cell membrane depolarization, influx of calcium, and ultimately stimulation of aldosterone synthesis (1, 3, 6, 9–11). How they lead to adrenal cell proliferation and tumor development remains less well understood and a fascinating area of ongoing research (12).

Among reported families with germline KCNJ5 mutations, severity of phenotype has varied considerably from mild PA readily treated with spironolactone with adrenals of apparently normal morphology on computed tomography (CT) to very florid PA requiring bilateral adrenalectomy yielding massively enlarged adrenals, and appears to be at least partly related to the KCNJ5 mutation genotype (1, 2, 10, 13–15). Surprisingly, G151E-mutated channels, which tend to be associated with a milder clinical phenotype, showed markedly higher (more abnormal) sodium conductance and conferred greater lethality to transfected HEK 293T cells than G151R-mutated channels associated with a much more florid phenotype. This increased lethality may explain the lack of adrenal hyperplasia occurring clinically in patients bearing germline G151E (vs G151R) mutations (14).

In a study of 251 Caucasian subjects with PA, we identified three heterozygous missense mutations (R52H, E246K, and G247R) and found that 12 subjects (5% of the cohort) were carriers for the rare nonsynonymous single nucleotide polymorphism rs7102584 causing E282Q substitution (16). Unlike previously described mutations associated with PA, these three mutations and rare polymorphism were all remote from the potassium channel selectivity filter. In vitro studies showed R52H, E246K, and E282Q (but not G247) substitutions to be functional, affecting the inward rectification, the ability of the Kir 3.4 channels to conduct Na+ currents, and angiotensin II-induced aldosterone release from the H295R cell line (16). Hence, germline variation in the KCNJ5 gene may have a role to play in the common sporadic form as well as the much rarer syndromic forms of PA.

Somatic mutations within APAs have since also been described in ATP1A1 (encoding the α-subunit of Na+/K+ ATPase), ATP2B3 (encodes a Ca2+ ATPase), and CACNA1D (encodes a voltage-gated calcium channel), but with much lower frequency (8, 17–20). As with KCNJ5 mutations, in vitro studies suggest these mutations also lead to zona glomerulosa cell membrane depolarization, resulting in increased aldosterone production (17–20). Unlike patients with KCNJ5-mutated APAs, however, those with APAs carrying the newer mutations have tended to be more commonly males (17, 19), with smaller APAs (8, 19) containing predominantly zona glomerulosa-like cells (19).

Pathophysiology

Several groups of investigators have recently begun to shed some light on the molecular pathways that may be involved in the development of PA. Lenzini et al (21) reported blunted expression of the TWIK-related acid-sensitive K+ channel 2 (TASK-2) gene in APAs compared with normal adrenal cortex. H295R adrenocortical cells transfected with a TASK-2 dominant-negative mutant construct showed increased aldosterone production and expression of the genes encoding aldosterone synthase (CYP11B2) and the steriodogenic acute regulatory protein. Two microRNAs (miRNAs), hsa-miR-23 and hsa-miR-34, the expression of which was shown to negatively correlate with that of TASK-2 in APAs, were found to decrease TASK-2 expression by binding to the 3’ untranslated region of the TASK-2 gene. These data suggest that lower expression of the TASK-2 channel in APA cells, perhaps induced by miRNAs, could contribute to excessive aldosterone secretion in human PA.

Robertson et al (22) investigated further the role of miRNA in APA development. Knockdown of Dicer1, a key enzyme in miRNA maturation, significantly enhanced CYP11B1 and CYP11B2 expression in H295R cells, suggesting a regulatory role for Dicer-dependent miRNAs. In vitro manipulation of one of these (miR-24, which was down-regulated in APA vs normal human adrenal cortex) confirmed its ability to modulate CYP11B1 and CYP11B2 expression, as well as cortisol and aldosterone production. If specificity to CYP11B2 could be conferred, adrenal...
miRNA may therefore be a therapeutic target for the treatment of PA.

Berthon et al (23) reported constitutive activation of WNT/β-catenin signaling, possibly due to decreased expression of the WNT inhibitor SFRP2, to be present in 70% of APAs and postulated that this may be an important step in APA development. In support of this, mice lacking Srp2 demonstrated increased aldosterone production and ectopic differentiation of zona glomerulosa cells. Furthermore, in vitro studies in H295R cells revealed a role for β-catenin to play in the control of both basal and angiotensin II-induced aldosterone secretion by activating AT1R, CYP21, and CYP11B2 transcription. The authors concluded that aberrant WNT/β-catenin activation is associated with APA development and that the WNT pathway may be worth considering as another therapeutic target in PA.

The role of the phosphoinositide 3-kinase/protein kinase B (or AKT)/mammalian target of rapamycin (mTOR) signaling pathway (involved in tumor development and metastasis in adenocortical carcinoma, pheochromocytoma, and many other human tumors) in the pathogenesis of PA was assessed by Su et al (24). Compared with normal zona glomerulosa, the levels of phospho-AKT, phospho-mTOR, phospho-S6, and vascular endothelial growth factor were significantly up-regulated in APA and hyperplastic adrenal cortex from patients with PA. No significant differences in the expression of these proteins were found between APA and hyperplasia. The mTOR inhibitor rapamycin inhibited both cell proliferation and aldosterone production by H295R cells. The authors concluded that the phosphoinositide 3-kinase/AKT/mTOR signaling pathway may mediate aldosterone hypersecretion and contribute to the development of PA.

Two groups have recently studied the potential role for agonistic autoantibodies against the angiotensin-II type-1 receptor (AT1RAA) in PA. Rossitto et al (25) found titers of AT1RAA to be higher in both PA patients (n = 46; 26 with APA, 20 with bilateral PA) and essential hypertensive patients (n = 62) than in normotensive subjects (n = 45). The titer was higher in patients with APA than in bilateral PA and appeared to allow discrimination of these two PA subtypes by receiver operator characteristics curve analysis. Plasma aldosterone concentrations fell more in AT1RAA-positive than AT1RAA-negative PA patients in response to captopril, in keeping with an agonistic role for these autoantibodies. In a smaller but more detailed analysis by Kem et al (26), sera from each of 13 patients with PA (three APA, two bilateral, and eight with indeterminate subtype) significantly increased angiotensin-II type-1 receptor (AT1R) activation in AT1R-transfected HAC15 adrenocortical cells compared with 20 control subjects, and this activity was inhibited by the selective AT1R blocker losartan. Both sera and IgG purified from AT1RAA-positive sera demonstrated vasoconstrictive effects in isolated rat cremaster arterioles (again blocked by losartan), and the IgG stimulated basal and angiotensin-induced aldosterone production by HAC15 cells (blocked by candesartan). These suggest the possibility that, at least in some individuals, the acquisition of antibodies directed against and capable of activating AT1R may contribute to the development of PA.

Non-Blood Pressure-Dependent Adverse Effects of Aldosterone Excess: Evidence Continues to Mount

Numerous published studies have demonstrated aldosterone excess to have adverse cardiovascular and renal effects that are at least partly independent of its effects on blood pressure and that appear to be reversible with specific treatment directed against aldosterone excess (27–32). Savard et al (33) compared the prevalence of cardiovascular events in 459 patients with PA and 1290 essential hypertensives matched for sex, age, and office systolic blood pressure. Patients with PA had a 2-fold greater prevalence of left ventricular hypertrophy (even after adjustment for hypertension duration) and significantly higher prevalence rates for coronary artery disease, nonfatal myocardial infarction, heart failure, and atrial fibrillation. Mulatero et al (34) reported a higher rate of cardiovascular events (and especially strokes and arrhythmias) among patients with PA (n = 270) than among essential hypertensives (n = 810) matched 1:3 in a case control fashion for blood pressure levels and several other risk factors. This was the case both for patients with APA and those with bilateral adrenal hyperplasia (BAH). Patients with PA were also more likely to develop type II diabetes during a 12-year period of follow-up after diagnosis. In this context, Fischer et al (35) reported mean glucose-induced first phase insulin secretion to be reduced in a group of 22 patients with PA, compared to that in 11 patients with essential hypertension, and to improve in nine with APA after unilateral adrenalectomy, suggesting a negative effect of aldosterone excess on pancreatic β-cell function.

Su et al (36) reported magnetic resonance imaging evidence of increased myocardial fibrosis and stiffening among 25 patients with PA vs 12 age-matched normotensive controls. Using magnetic resonance imaging to measure aortic distensibility, Mark et al (37) reported that PA patients (n = 14) displayed increased arterial stiffness compared with matched essential hypertensives (n = 33). Lin et al (38) found plasma procollagen levels to be higher
and cyclical variation of integrated backscatter (a marker of left ventricular systolic function) on echocardiography to be lower among 20 patients with APA compared to 20 with essential hypertension. Parameters improved in the APA group after adrenalectomy. These investigators also described higher carotid intima-medial thicknesses on duplex ultrasound scanning and higher arterial pulse wave velocities in the patients with APA, again improving after surgery (39). In a multivariate regression analysis of 54 patients with PA followed for a mean of 6.4 years after unilateral adrenalectomy or commencement of spironolactone, Catena et al (40) found that the degree of reduction in left ventricular mass was independently predicted not only by change in systolic blood pressure but also by pretreatment plasma aldosterone levels, further supporting a role for aldosterone in promoting left ventricular hypertrophy that is independent of the hypertension-related hemodynamic load.

**Detection and Diagnosis of PA: What’s New?**

Although still regarded as the most reliable available screening test for PA, the plasma aldosterone/renin ratio (ARR) can be affected, and results thereby can be confounded by many factors (41), indicating a need for improved approaches. Pizzolo et al (42) reported on the use of N-terminal probrain natriuretic peptide (NT-proBNP) measurement as an adjunct to the ARR. Among 132 hypertensive subjects who underwent ARR testing, 81 with high ARR underwent iv saline infusion suppression testing (SST). NT-proBNP levels correlated positively with ARR and negatively with renin level, were higher in patients with high ARR and in those with positive SST, and were an independent predictor of positive SST. The proportion of patients with a positive SST ranged from only 23% in females with a low NT-proBNP to 93% in males with a high NT-proBNP, leading the authors to conclude that NT-proBNP may prove useful to identify which patients with a high ARR should receive formal confirmatory testing (42).

In patients who screen positive for PA, confirmatory testing is necessary to definitely confirm the diagnosis before proceeding to adrenal venous sampling (AVS) to differentiate unilateral, surgically correctable forms of PA from bilateral forms usually treated medically. Four confirmatory tests are commonly used, namely, oral sodium loading, saline infusion, fludrocortisone administration with oral sodium loading, and captopril challenge testing. Of these, fludrocortisone suppression testing (FST) has been considered the most reliable (43), but it is cumbersome, difficult to perform, and relatively expensive, requiring hospital admission for several days. Gouli et al (44) recently reported on a modified form of the test in which 1 mg of dexamethasone was administered on the last day of the FST to eliminate any stimulatory effect of endogenous stress-induced adrenocorticotropic on aldosterone secretion, a recognized confounder of test results (43). Westerdalh et al (45) recently reported captopril challenge testing to have poor discriminatory power for PA in hypertensive patients with high basal ARR. We previously found SST, performed in the recumbent position, to lack sensitivity for PA, missing most patients (including those with APA) who tested positive by FST (46). Willenberg et al (47) similarly found SST to lead to greater suppression of plasma aldosterone than FST. Because aldosterone levels can be higher upright (eg, seated) than recumbent in patients with PA and upright levels are used during FST, we hypothesized that seated SST (SSST) is more sensitive than recumbent SST (RSST), especially for posture-responsive PA. In a pilot study of 31 patients who underwent FST (upright plasma aldosterone levels measured at 10 AM basally and after 4 d of fludrocortisone 0.1 mg every 6 h and oral salt loading) and SST (aldosterone levels measured basally at 8 AM and after infusion of 2 L normal saline over 4 h), both recumbent and seated in randomized order, FST confirmed PA in 23 patients, excluded PA in three, and was originally “inconclusive” in five (48). However, one patient with inconclusive FST had PA confirmed by lateralizing AVS and was reclassified “unilateral PA.” Of the 24 with confirmed PA (eight unilateral, 11 bilateral, and five undetermined subtype), 23 (96%) tested positive by SSST compared to only eight (33%) by RSST (P < .001) (Table 1). RSST missed one unilateral, all 11 bilateral, and four with as yet undeter-

**Table 1. Comparison of Results of SSST vs RSST in 24 Patients With Confirmed PA**

<table>
<thead>
<tr>
<th></th>
<th>SSST Positive</th>
<th>RSST Positive</th>
</tr>
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<tbody>
<tr>
<td>Total patients with PA</td>
<td>23 (96)**</td>
<td>8 (33)</td>
</tr>
<tr>
<td>Unilateral PA</td>
<td>23 (96)*</td>
<td>8 (33)</td>
</tr>
<tr>
<td>Bilateral PA</td>
<td>11 (91)**</td>
<td>0 (0)</td>
</tr>
<tr>
<td>PA subtype yet to be determined</td>
<td>5 (100)*</td>
<td>1 (20)</td>
</tr>
<tr>
<td>Posture-responsive</td>
<td>13 (93)**</td>
<td>1 (7)</td>
</tr>
<tr>
<td>Posture-unresponsive</td>
<td>10 (100)</td>
<td>7 (70)</td>
</tr>
</tbody>
</table>

Data are expressed as number or number (percentage). Posture-responsive denotes plasma aldosterone responsive to upright posture (levels measured at 1000 AM after 3 hours of upright posture at least 50% higher than those measured basally at 7 AM after overnight recumbency). Posture-unresponsive denotes plasma aldosterone unresponsive to upright posture (upright levels less than 50% higher or lower than recumbent). * P < .01; and ** P < .001 vs RSST.
mined subtype. RSST was positive in seven of 10 (70%) posture-unresponsive vs one of 14 (7.1%) posture-responsive patients ($P < .005$). Clearly, larger studies are required, but these preliminary results suggest that SSST may be superior to RSST in terms of sensitivity for detecting PA, especially posture-responsive forms, and may represent a reliable, yet much simpler, cheaper, and quicker alternative to FST.

**Subtype Differentiation**

Currently, the only reliable way to differentiate unilateral (mainly APA) from bilateral PA and to lateralize APAs preoperatively is by adrenal venous sampling (AVS). However, achieving high success rates for cannulation of the adrenal veins (and especially the right, which is more difficult to locate) represents a challenge to institutions offering AVS. Recent studies have demonstrated that limiting the number of proceduralists at each institution to one or two and providing them with a higher throughput of procedures probably have the most profound effects on improving cannulation success rates (judged by adrenal/peripheral venous cortisol gradients) (49, 50). Rapid, point-of-care cortisol assays can alert the proceduralist, at the time of AVS, whether adrenal venous blood has been successfully obtained, and have also resulted in higher success rates in centers using this technique (51–53). We previously reported on the use of CT to localize the right adrenal vein before AVS (46, 54). The use of CT during angiography to determine correctness of catheter placement was recently reported by two groups to lead to catheter repositioning in approximately 10% of patients and to be associated with achievement of high (>90%) rates of successful cannulation (55, 56). Improved success rates have also been reported with the use of ACTH stimulation during AVS (57, 58). Presumably, at least in some of these instances, cannulation had already been achieved but was not confirmed by cortisol gradients because of a transient lack of cortisol production at the time of sampling. A concern about ACTH stimulation has been that, in a patient with APA, ACTH could lead to stimulation of the contralateral gland, causing results to suggest bilateral, rather than unilateral, disease. In the study by Monticone et al (58), the use of more stringent (for example, adrenal/peripheral venous cortisol gradients of $> 3.0$ basally and $> 4.0$ post-ACTH) criteria for cannulation proved critical for optimizing pre- vs post-ACTH diagnostic reproducibility.

Traditional AVS provides information about the relative degree of aldosterone production by an entire adrenal gland vs the other gland. In a recent advance, Japanese workers have reported on the use of superselective AVS that has the ability to sample different branches of the adrenal vein and thereby compare aldosterone production by different regions within a gland (59, 60). In this way, it has been possible to localize small aldosterone-producing lesions with sufficient resolution to permit their removal by partial, rather than total unilateral, adrenalectomy (59, 60). Although this may only seem clinically relevant for a minority of patients with PA (for example, those suspected as having bilateral APAs, or who have only one functioning adrenal that may contain an APA, who otherwise would require steroid replacement therapy postoperatively), it nevertheless represents a promising new approach with potentially wider future applications.

Once considered a rarity, autonomous tumorous cosecretion of cortisol has been recently reported by two groups to be relatively common (>$10%$) among patients with APA, even in the absence of obvious clinical features of Cushing syndrome (“subclinical Cushing syndrome”) (61, 62). This has implications not only for perioperative management with the need, at least in some patients, to consider glucocorticoid replacement, but also for interpretation of AVS results. Cosecretion of cortisol would be expected to raise cortisol levels in the adrenal vein draining the APA (thus lowering the ipsilateral aldosterone/cortisol ratio), while causing chronic suppression of ACTH and thus lowering of cortisol secretion by the contralateral gland (thus raising the contralateral aldosterone/cortisol ratio and lowering the adrenal/peripheral venous cortisol ratio). This could result in loss of lateralization (and a lost opportunity to offer potentially curative surgery) or give the impression that cannulation had failed on the contralateral side (and thereby lead to inappropriate consideration for repeat AVS). One potential solution to this problem is the measurement of an alternative, non-ACTH-dependent adrenal hormone to cortisol. Dekkers et al (63), for example, found measurement of plasma metanephrine during AVS to be superior to measuring cortisol in assessing success of cannulation, with adrenal/peripheral ratios 6-fold higher among procedures deemed to be successful according to cortisol criteria. Baba et al (64) reported catecholamine levels to be useful for judging not only cannulation success but also lateralization.

The search for reliable alternative approaches that are less costly, invasive, and technically demanding than AVS has been ongoing in recent years but has met with limited success. A clinical prediction score developed by Küpers et al (65), who reported the presence of a unilateral mass lesion on CT “typical” for APA plus a serum potassium of $< 3.5$ mmol/L or an estimated glomerular filtration rate of at least 100 mL/min/1.73 m$^2$ to have 100% specificity (but only 53% sensitivity) for unilateral PA, was found to
be much less reliable when applied to patients within the German Conn’s Registry (66), except in those under 40 years of age. Sze et al (67) also found the clinical prediction score to have a lower specificity (88%) among a UK cohort of patients with PA and were unable to show superiority over an imaging-based strategy. Other more promising new approaches for differentiating APA from BAH, which require validation in different centers and with larger numbers, have included the use of semiquantification of 111I-6β-iodomethyl-norcholesterol (NP-59) single photon emission CT/CT (68) and 11C-metomidate positron emission tomography/CT (69).

Studies attempting to assess (including by receiver operator characteristics analysis) and validate approaches to distinguishing unilateral from bilateral PA have been challenged by the lack of a robust “gold standard” for diagnosing APA. The blood pressure response to surgery, for example, may be incomplete if the patient is studied too soon after adrenalectomy or has other causes of residual hypertension. The response to surgery of plasma aldosterone plus or minus renin levels and the ARR is better, but it is still imperfect because cutoff criteria are arbitrary to some extent and the ARR is associated with false positives and negatives. Our own approach is to repeat the FST postoperatively (70), but, as mentioned above, this is time consuming, cumbersome, and expensive to perform. Demonstration of an adenoma in surgically removed adrenals does not prove that it was actually producing aldosterone, and reliance on this would have the potential to misclassify unilateral PA due to micronodular hyperplasia as bilateral PA. Recent studies have reported on the use of immunohistochemical staining of resected adrenals in an attempt to confirm APA removal (71–75). Using isoform-specific monoclonal antibodies against the 3β-hydroxysteroid dehydrogenase/isomerase family, Doi et al (71) reported hyperplasia of zona glomerulosa seen in bilateral PA to be accompanied by a robust expression of the HSD3B1 isoform. In contrast, tumor cells in APA were immunonegative to HSD3B1 but showed expression of HSD3B2. Volpe et al (72) found APAs to show strong immunoreactivity for aldosterone synthase. In adrenals from a significant minority of patients, they found either “adenomas” (presumably defined as adrenocortical tumors of at least 1 cm in diameter) that were not CYP11B2 positive or no adenoma at all, but they found smaller nodules with strong CYB11B2 immunoreactivity, indicating aldosterone-producing nodular hyperplasia. Nanba et al (73), in a study of 32 patients with PA who underwent unilateral adrenalectomy, found 22 to show positive CYP11B2 immunostaining in their tumors (designated APAs) and eight with either CYP11B2-negative adenomas (n = 4) or without tumors on CT to show aldosterone-producing cell clusters with CYP11B2 immunostaining in the zona glomerulosa. In APAs, CYP11B2 score adjusted for tumor volume was positively correlated with plasma aldosterone and negatively with serum potassium. APAs with either similar or greater amounts of CYP11B1 (11β-hydroxylase, which catalyzes the last step of cortisol biosynthesis) to CYP11B2 staining showed significantly higher serum cortisol after 1 mg dexamethasone and larger tumor size than the CYP11B2-dominant APA group. High-quality antibodies produced in the laboratory of Celso Gomez-Sanchez (University of Mississippi) have also been used to demonstrate correlations between the degree of expression of CYP11B2 in APAs and preoperative plasma (and urinary) aldosterone levels (74). In addition, this group has shown expression levels to be similar in zona glomerulosa within normal adrenals and in those removed from patients with PA due to BAH (and cited increased expression of upstream enzymes to be a possible explanation for the greater aldosterone production in the latter group), but lower in that in zona glomerulosa adjacent to an APA (75), consistent with suppression of aldosterone production.

Importance of Subtype Differentiation

For both cardiovascular outcomes (76–78) and quality of life (79, 80), beneficial effects of surgery in unilateral PA have been reported to be superior (more marked and/or more rapid) to those of medical therapy in bilateral PA. In a prospective study of 54 patients with PA who were treated by adrenalectomy (n = 24) or with spironolactone (n = 30), Catena et al (76) reported left ventricular mass to have decreased significantly by 1 year of follow-up only in adrenalectomized patients. Although overall change from baseline to the end of a mean of 6.4 years of follow-up was comparable in the two groups, there remains the possibility that the apparent delay in benefit afforded by medical (over surgical) treatment could translate to less impressive reductions in cardiovascular morbidity and mortality. A nationwide epidemiological study conducted in Japan, which analyzed data from 1706 patients with PA, found that among those with APA, surgical, but not medical, treatment was significantly associated with amelioration of hypertension (81). By contrast, among patients with hyperplastic forms of PA (mostly BAH), there was no relationship between either surgical or medical treatment and the prognosis of hypertension. In an observational study reported by Reincke et al (82) on 300 patients with PA, adrenalectomy (presumably mostly for unilateral PA) was associated with reduced all-cause mortality when compared with medical treatment.
Toward High-Quality Assay Development

Screening, confirmation, and subtype differentiation of PA are all dependent on accurate measurement of plasma aldosterone, and because the ARR is predominantly renin-dependent, it is critical that the methods used to assay aldosterone and renin in clinical samples are of high quality. The recent push for using automated platforms and immunometric methodology has been primarily on the basis of faster throughput and lower cost and potentially at the expense of lower reliability. In 2009, we reported on the development of a high-throughput, highly accurate, and reproducible method of measuring plasma aldosterone using HPLC and tandem mass spectrometry (LC-MS/MS) (83). Since then, several other investigators have established similar assays for aldosterone (84, 85), and at least two groups have developed assays that use LC-MS/MS to measure angiotensin I and thereby determine plasma renin activity (86, 87). The hope for incorporation of tandem mass spectrometry (LC-MS/MS) technology into routine clinical laboratory practice for measuring aldosterone and renin (or alternatively angiotensin II, with implications for the development of aldosterone/angiotensin II ratio testing as a new and possibly superior screening approach) now appears to be well within reach.

New Approaches to Treatment

The mainstays of treatment for PA continue to be unilateral adrenalectomy for most patients with unilateral forms and medications that antagonize aldosterone action (for example, spironolactone 12.50–50 mg daily, eplerenone 25–100 mg daily, or amiloride 2.5–20 mg daily) for most with bilateral PA. Both treatment approaches can result in marked blood pressure lowering in patients with hypertension, including those with resistant forms.

Recent surgical developments have included reports of single incision laparoscopic adrenalectomy (88, 89), which offers superior cosmetic results to multiple incision approaches but requires further experience to establish its value from a risk vs benefit perspective.

On the medical front, a randomized, open-label, blinded end-point study involving 34 patients with BAH reported in 2009 by Karagiannis et al (90) found eplerenone and spironolactone to be of similar efficacy in reducing blood pressure. Conversely, a more recent randomized, double-blinded trial comparing the efficacy, safety, and tolerability of eplerenone to that of spironolactone (given in relatively large daily doses of 100–300 and 75–225 mg, respectively) in patients with PA found spironolactone to be superior in terms of blood pressure lowering, but to be associated, as expected, with higher rates of male gynecomastia (21 vs 5% for eplerenone) and female mastodynia (21 vs 0%) (91). Aldosterone synthase inhibitors remain a promising treatment approach, although a recent study showed that in patients with PA, the effects of eplerenone treatment (50–100 mg twice daily) on blood pressure, plasma potassium, and renin were more marked than those of 4 weeks of treatment with the aldosterone synthase inhibitor LCI699 (0.5–1 mg twice daily) (92). An ongoing concern for these new drugs is the potential for inhibiting cortisol production in addition to that of aldosterone.

Nonsteroidal dihydropyridine-based mineralocorticoid receptor antagonists (93, 94) are a new drug class that has displayed similar in vitro potency to spironolactone, without apparent effects on androgen and progesterone receptors, and therefore presumably without the same risk of sex steroid-related side effects. BAY 94–8862, the first-in-class of this new generation of drugs, is currently in development for treatment of heart failure (93), with early results showing considerable promise (95), and studies in PA are eagerly awaited.

In both animals and humans, endogenous aldosterone and dietary salt intake appear to interact to induce target-organ deterioration. Pimenta et al demonstrated that both left ventricular mass (96) and degree of proteinuria (97) in patients with PA were dependent on 24-hour sodium excretion rate (as a marker of dietary salt intake). Increased dietary sodium and aldosterone were also shown by these workers to be related to severity of obstructive sleep apnea in patients with resistant hypertension (98). Incorporation of dietary salt restriction in the management of patients with PA may therefore help to limit and even reverse target organ damage and morbidity.

Conclusions

In an exciting time for research into PA, major inroads have been made into understanding its pathophysiology, including its underlying genetic bases, with the potential for development of new treatment modalities. Advances in approaches to confirmatory testing, subtype differentiation, assay technology, and treatment are helping to enable treating physicians to optimize diagnosis and management for patients with this important condition, with the promises of reduced morbidity and mortality and improved quality of life.

Acknowledgments

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