Major advances have been made during the past 4 years in understanding the pathophysiology of aldosterone production in patients with primary aldosteronism (PA). The breakthrough was the identification of somatic mutations in the potassium channel GIRK4 (encoded by KCNJ5) in aldosterone-producing adenomas (APAs) and the contemporaneous discovery, by the same authors, of a germline mutation responsible for familial hyperaldosteronism type III. This was followed by the identification of further somatic mutations in APAs in 2 ATPases (Na+/K+-ATPase 1 and Ca 2+-ATPase 3, encoded by ATP1A1 and ATP2B3, respectively) and in a subunit of an L-type voltage-gated Ca2+-channel, Cav1.3 (encoded by CACNA1D).

The zona glomerulosa cells of the adrenal cortex display a high resting outward potassium current through the GIRK4 potassium channel that contributes to the hyperpolarization of the cell membrane. Most of the GIRK4 mutations identified in APA are located in or within close proximity to the activity filter of the K+ channel and result in the indiscriminate conductance of Na+ that depolarizes the cell membrane and causes the opening of voltage-gated Ca2+ channels. The resultant Ca2+ influx and activation of the Ca2+ signaling pathway results in the activation of CYP11B2 gene transcription and an increase in aldosterone biosynthesis.

The other somatic APA mutations in ATP1A1, ATP2B3, and CACNA1D are also predicted, or have been demonstrated, to cause an increase in aldosterone production via the activation of the Ca2+ signaling pathway although the mechanism at source that leads to Ca2+ influx differs.

At present, the origin of the increased cell proliferation in the adenomas is unknown and is not accounted for by the increase in intracellular Ca2+ induced by the APA mutations.

In the present issue of Hypertension, Zheng et al report a strikingly high prevalence (77%) of KCNJ5 somatic mutations in 168 APA removed from Chinese patients. Consistent with previous reports, mutations in the other genes were rarer, 4 in ATP1A1 (2.8%) and 1 each in ATP2B3 and CACNA1D. This in agreement with other reports from Japanese patients in which KCNJ5 mutations were present in 65% to 69% of APAs, indicating that KCNJ5 mutations are the most frequent genetic cause of excessive aldosterone production in Eastern Asian patients with APA (Figure [A]).

These results display some differences and similarities with those in a study of somatic APA mutations in white patients. The most intriguing common feature is the predominance of somatic KCNJ5 mutations in women, which has been consistently reported in white cohorts and now in Chinese and Japanese patients. The underlying mechanism responsible for this distinction is unknown, a difference in the phenotype between sexes has been reported in the leak K+ channel TASK1 knockout animal model, in which only female adults display hyperaldosteronism, whereas males are unaffected and seem to be protected by the androgen-regulated compensatory expression of TASK3 channels.

Zheng et al also reported an apparently more severe biochemical phenotype (higher aldosterone:renin ratio and lower potassium although not a higher lateralization index at adrenal vein sampling [AVS]) compared with patients without mutations in KCNJ5, CACNA1D, ATP1A1, and ATP2B3. A similar biochemically more severe phenotype has been observed in some but not in all studies in whites and in particular was not observed in the largest study in European patients. It should be considered that the no-mutation group could be heterogeneous in that it almost certainly comprises patients with as yet unidentified genes carrying somatic APA mutations, as well as some patients in which adrenalectomy was decided on the basis of the computed tomographic scanning alone without performing AVS or using AVS criteria that are not strict enough to ensure the removal of an APA and result in the removal of 1 adrenal with a nodule in the context of a bilateral disease. If this is the case, the mutated group will always display a more severe clinical and biochemical phenotype compared with the no-mutation group. In the study by Zheng et al, less than half of the patients underwent AVS; however, the proportion of KCNJ5 mutations was not significantly different in the group operated on the basis of the computed tomographic scanning or after AVS, a finding also observed in the Paris cohort. Therefore, a major selection bias should not have interfered with the findings of the present study. By contrast, the use of AVS in a minority of patients with PA could have been responsible for the relatively low prevalence of CACNA1D mutations in this Chinese cohort. In fact, APAs harboring CACNA1D mutations are associated with smaller nodule dimensions and, therefore, it is conceivable that a lower proportion of patients carrying these mutations are observed in cohorts where AVS is not performed in all patients with PA and are adrenalectomized relying on imaging alone, a strategy that could miss a relevant proportion of APAs with
CACNA1D mutations. It should be emphasized that, at present, the phenotype presentation of patients with or without mutations in the above genes do not display any characteristics that are sensitive or specific enough to allow the detection of a patient with a somatic mutation only by relying on a specific clinical or biochemical picture. Furthermore, even if specific peripheral biomarkers should become available, AVS would still be required to determine the correct side for the adrenalectomy in those patients.

The interest in the role of KCNJ5 mutations in PA has been heightened further by the recent discovery of 3 novel germline mutations (p.Arg52His, p.Glu246Lys, and p.Gly247Arg) and a rare nonsynonymous single nucleotide polymorphism responsible for the substitution p.Glu282Gln, in a cohort of patients with sporadic PA. Interestingly, the substitutions, with the exception of the 247 mutation that had no effect, were associated with an alteration of GIRK4 function in vitro resulting in a loss of ion selectivity causing Na+-influx and stimulation of aldosterone secretion. Therefore, germline KCNJ5 mutations are not only responsible for the severe familial hyperaldosteronism type III but are also present in a significant proportion (∼6%) of patients with sporadic PA (Figure [B]). These findings also demonstrated that even mutations located at some distance from the selectivity filter display functional effects that alter GIRK4 function and determine the promiscuous entry of sodium through GIRK4 into cells, ultimately resulting in increased aldosterone secretion.

Recent years have shed light on the mechanisms responsible for the increased aldosterone production in APA and in familial hyperaldosteronism type III but are also present in a significant proportion (∼6%) of patients with sporadic PA (Figure [B]). These findings also demonstrated that even mutations located at some distance from the selectivity filter display functional effects that alter GIRK4 function and determine the promiscuous entry of sodium through GIRK4 into cells, ultimately resulting in increased aldosterone secretion. Furthermore, APAs are often associated with multiple nodules in the cortex indicating that the formation of a nodule is independent from the appearance of a mutation that confers the secretory activity to the nodule. Interestingly, a recent study investigated the association of KCNJ5 single nucleotide polymorphisms and PA in Chinese patients; one of the polymorphisms, located in the 3′ untranslated region, was associated with PA in male patients. The functional effect of this polymorphism is unknown; however, an effect on mRNA stability or processing is not expected to cause hyperaldosteronism because variations in KCNJ5 expression (including silencing and overexpression) do not result in major changes in aldosterone secretion and, therefore, the significance of this association needs to be explored more in depth and confirmed in a different population.

An advance in understanding the role of KCNJ5 in aldosterone hypersecretion has also been defined from pharmacological evidence: mutated GIRK4 channels are less sensitive to tertiapin-Q, a selective inhibitor of wild-type GIRK4. Furthermore, mutated GIRK4 activity is blocked by the epithelial sodium channel inhibitor amiloride, and even more potently by the phenylalkylamine L-type calcium channel blocker verapamil. By contrast, the dihydropiridine calcium channel blocker nifedipine is a weak blocker of the mutated channel. This information will potentially stimulate the search or design of new and selective blockers of mutated GIRK4 channels that are predicted to be effective in patients with familial hyperaldosteronism type III and in a significant subset of patients with sporadic PA. The effects of verapamil on blood pressure and aldosterone levels in patients carrying KCNJ5 mutations could be because of a dual effect of blocking the mutated GIRK4 channel in addition to the voltage-gated calcium channel activated by the cell depolarization subsequent to sodium entry through the GIRK4 channel. This should be taken into account because verapamil is considered to be neutral on aldosterone levels and is, therefore, considered one of the drugs of choice during PA diagnostic work-up.

Figure. A. Relative frequency of mutations in KCNJ5, ATP1A1, ATP2B3, and CACNA1D in Eastern Asia and white patients. B. Somatic and germline KCNJ5 mutations demonstrated in patients with aldosterone-producing adenoma, in familial hyperaldosteronism (FH)-III and in sporadic primary aldosteronism (PA).
By contrast, verapamil has the potential to reduce aldosterone production resulting in a negative screening test or in a reduced lateralization index during AVS.

In conclusion, mutations in KCNJ5 are the most frequent known alterations in patients with PA: drugs specifically blocking the mutated GIRK4 channel are expected to be effective in a large percentage of patients with PA.

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References
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