



Primary Aldosteronism Takes (KCNJ)Five!

Maria-Christina Zennaro

► **To cite this version:**

Maria-Christina Zennaro. Primary Aldosteronism Takes (KCNJ)Five!. *Endocrinology, Endocrine Society*, 2012, 153 (4), pp.1575-7. <10.1210/en.2012-1146>. <inserm-00686469>

HAL Id: inserm-00686469

<http://www.hal.inserm.fr/inserm-00686469>

Submitted on 10 Apr 2013

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Primary aldosteronism Takes (KCNJ)Five!

Authors: Maria-Christina Zennaro^{1,2,3}

Affiliations :

¹INSERM, UMRS_970, Paris Cardiovascular Research Center, Paris, France

²Université Paris Descartes, Sorbonne Paris Cité, Paris, France

³Assistance Publique-Hôpitaux de Paris, Hôpital Européen Georges Pompidou, Paris, France

Correspondance to:

Maria-Christina Zennaro, MD, PhD

INSERM U970 – PARCC

56 rue Leblanc

75015 Paris

France

Tel: +33 1 53 98 80 42

Fax: +33 1 53 98 79 52

maria-christina.zennaro@inserm.fr

Primary aldosteronism (PA) is the most common form of endocrine hypertension, due to autonomous aldosterone production from the adrenal cortex. Patients with PA typically present with hypertension, high plasma aldosterone levels associated with low plasma renin activity, and varying degrees of hypokalemia and metabolic alkalosis (1). The prevalence of PA increases with the severity of hypertension and is currently estimated around 10% in referred patients and 4% in primary care [(2) and references therein; (3)], but as high as 20% in patients with resistant hypertension (4-5). The two main causes of PA are aldosterone producing adenoma (APA) and bilateral adrenal hyperplasia (BAH), also called idiopathic hyperaldosteronism. Efficient and timely screening for PA is of major importance, given the severe cardiovascular outcome of aldosterone excess that is independent of blood pressure levels (6-7). Yet, the pathogenic mechanisms leading to aldosterone hypersecretion and cell proliferation are largely unknown.

Recently, a few recurrent somatic mutations of the *KCNJ5* gene, coding for the potassium channel Kir3.4 have been implicated as a cause of APA, while an inherited mutation was identified in a family with a Mendelian form of early severe hypertension which features massive adrenal hyperplasia and is referred to as familial hyperaldosteronism type 3 [FH3, (8-9)]. These mutations all lie near or within the selectivity filter of the Kir3.4 channel; they result in a loss of channel selectivity, with increased sodium conductance leading to membrane depolarization. These changes are presumed to be responsible for constitutive aldosterone secretion and cell proliferation by promoting opening of membrane voltage-dependent calcium channels which is followed by activation of the calcium signaling pathway, the main trigger for aldosterone production in adrenal zona glomerulosa cells. In this issue of *Endocrinology*, Oki et al now formally establish a causal relationship between *KCNJ5* mutations and hyperaldosteronism. They demonstrate that the inherited *KCNJ5* T158A mutation produces a marked stimulation in aldosterone biosynthesis which is

dependent on membrane depolarization followed by calcium influx into adrenal cortical carcinoma cells (Oki et al, 2012).

The paper by Oki et al indeed tackles a central issue that had not previously been addressed, i.e. the causal link between *KCNJ5* mutations, membrane depolarization and aldosterone overproduction and cell proliferation. By transiently infecting adrenocortical HAC15 cells with a lentivirus expressing wild type *KCNJ5* or mutated *KCNJ5*-T158A, the authors show that expression of channels harboring the T158A mutation potentiated basal aldosterone production, which was further stimulated by angiotensin II and the protein kinase A activator, forskolin. Using different fluorescent dyes, they confirmed the enhanced sodium influx through the mutated Kir3.4 channel, leading to membrane depolarization and increased intracellular calcium concentrations. *KCNJ5*-T158A transduced cells presented significantly increased expression of *CYP11B2*, the gene coding for aldosterone-synthase which ensures the last three enzymatic steps of aldosterone biosynthesis. Expression of *CYP11B1*, coding for 11 β -hydroxylase, was also increased, as was basal and stimulated cortisol production. *KCNJ5*-T158A also induced significant production of 18-oxocortisol, a hybrid steroid largely produced in affected members of the original family with FH3 (10). *CYP11B2* expression and aldosterone production were both inhibited by calcium channel antagonists and calmodulin inhibitors, confirming the mechanistic link to activation of the calcium signaling pathway.

While these results confirm the pathogenic role of the *KCNJ5* T158A mutation in promoting aldosterone-overproduction, the same does not hold true for its effects on cell proliferation. Indeed, *KCNJ5*-T158A had an inhibitory effect (~30% reduction) on HAC15 cell proliferation, in stably infected cells, an effect which was not due to increased apoptosis and was unrelated to calcium signaling. Interestingly, a similar negative effect of *KCNJ5* mutations on cell proliferation was recently described for two other mutants. The *KCNJ5* G151R mutation is one of the recurrent somatic mutations found in APA and has recently

been described as inherited mutation in two families with early onset, severe progressive hyperaldosteronism and adrenal cortex hyperplasia, requiring bilateral adrenalectomy in childhood to control blood pressure (11). Despite this adrenal phenotype, *in vitro* studies demonstrated that the G151R mutation significantly reduced cell survival when transfected into human embryonic kidney 293T cells. A second mutation affecting the same amino acid, G151E, was recently described in a family diagnosed with non-glucocorticoid remediable familial hyperaldosteronism (12). This mutation was associated with a much milder phenotype of the two affected family members, compared to classical FH3, with blood pressure levels and hypokalemia easily corrected by medical therapy. Remarkably, the adrenals appeared normal by CT scanning, and hybrid steroids were produced at a rate comparable to that of other patients with sporadic PA. Again, the KCNJ5-G151E mutant channel was similarly permeable for sodium and potassium, resulting in depolarization of the plasma membrane and a continuous sodium influx (12). The KCNJ5 G151E mutation was reported in two additional families with early onset hyperaldosteronism of unknown cause (11). These subjects also had easily controlled hypertension and no evidence of adrenal hyperplasia. Remarkably, enough, in this study KCNJ5-G151E channels produced a much larger sodium conductance as compared to KCNJ5-G151R, resulting in rapid sodium-dependent cell lethality (11). Although G151E carriers do not present adrenal cortex hyperplasia, indicating that the mutation may limit adrenocortical cell proliferation yet promoting aldosterone overproduction, it is difficult to conceive how the G151R mutation, which also leads to reduced cell survival in the same study, induces bilateral adrenal hyperplasia when inherited and APA formation when occurring somatically. Based on these studies and the work by Oki et al, it remains therefore unclear whether and how *KCNJ5* mutations are responsible for increased cell proliferation. Alternatively, the observed effects raise the possibility that *KCNJ5* mutations are responsible for aldosterone hypersecretion,

while increased cell proliferation is triggered by other, yet to be identified, mechanisms. Consistent with this hypothesis is the observation that inactivation of several relevant adrenal potassium channels in mice leads to increased aldosterone production, but not to adrenal cortex hyperplasia or APA formation (Table 1).

Somatic *KCNJ5* mutations have been demonstrated to be present in a large proportion of patients with APA, with an estimated prevalence in unselected patients of ~34% (13), and even higher frequencies described depending on the sample size, the screening procedures for selecting patients for adrenalectomy and genetic background (14-15). However, although germinal *KCNJ5* mutations are responsible for inherited forms of familial hyperaldosteronism, they are not similarly causative for sporadic BAH (13). Even though unilateral adrenalectomy represents the treatment of choice for PA in specialized departments, the fact that only 30% of operated patients are cured and that a subset of patients don't even go into surgery because of the multi-tiered process for subtype identification, sets medical treatment as an attractive therapeutic option even in lateralized forms of PA. Kir3.4 potassium channels may thus represent interesting new drug targets for a subset of APA not eligible for surgery. Cell as well as animal models expressing mutant Kir3.4 channels may provide valuable tools to address the functional consequences of *KCNJ5* mutations, allowing to further dissect the mechanistic determinants of aldosterone overproduction and increased cell proliferation in PA.

References

1. **Funder JW, Carey RM, Fardella C, Gomez-Sanchez CE, Mantero F, Stowasser M, Young WF, Jr., Montori VM** 2008 Case detection, diagnosis, and treatment of patients with primary aldosteronism: an endocrine society clinical practice guideline. *J Clin Endocrinol Metab* 93:3266-3281
2. **Hannemann A, Wallaschofski H** 2011 Prevalence of Primary Aldosteronism in Patient's Cohorts and in Population-based Studies - A Review of the Current Literature. *Horm Metab Res*
3. **Plouin PF, Amar L, Chatellier G** 2004 Trends in the prevalence of primary aldosteronism, aldosterone-producing adenomas, and surgically correctable aldosterone-dependent hypertension. *Nephrol Dial Transplant* 19:774-777.
4. **Douma S, Petidis K, Doumas M, Papaefthimiou P, Triantafyllou A, Kartali N, Papadopoulos N, Vogiatzis K, Zamboulis C** 2008 Prevalence of primary hyperaldosteronism in resistant hypertension: a retrospective observational study. *Lancet* 371:1921-1926
5. **Calhoun DA, Nishizaka MK, Zaman MA, Thakkar RB, Weissmann P** 2002 Hyperaldosteronism among black and white subjects with resistant hypertension. *Hypertension* 40:892-896
6. **Rossi GP, Sechi LA, Giacchetti G, Ronconi V, Strazzullo P, Funder JW** 2008 Primary aldosteronism: cardiovascular, renal and metabolic implications. *Trends Endocrinol Metab* 19:88-90
7. **Milliez P, Girerd X, Plouin PF, Blacher J, Safar ME, Mourad JJ** 2005 Evidence for an increased rate of cardiovascular events in patients with primary aldosteronism. *J Am Coll Cardiol* 45:1243-1248
8. **Choi M, Scholl UI, Yue P, Bjorklund P, Zhao B, Nelson-Williams C, Ji W, Cho Y, Patel A, Men CJ, Lolis E, Wisgerhof MV, Geller DS, Mane S, Hellman P, Westin G, Akerstrom G, Wang W, Carling T, Lifton RP** 2011 K⁺ channel mutations in adrenal aldosterone-producing adenomas and hereditary hypertension. *Science* 331:768-772
9. **Mulatero P** 2008 A new form of hereditary primary aldosteronism: familial hyperaldosteronism type III. *J Clin Endocrinol Metab* 93:2972-2974
10. **Geller DS, Zhang J, Wisgerhof MV, Shackleton C, Kashgarian M, Lifton RP** 2008 A novel form of human mendelian hypertension featuring nonglucocorticoid-remediable aldosteronism. *J Clin Endocrinol Metab* 93:3117-3123
11. **Scholl UI, Nelson-Williams C, Yue P, Grekin R, Wyatt RJ, Dillon MJ, Couch R, Hammer LK, Harley FL, Farhi A, Wang WH, Lifton RP** 2012 Hypertension with or without adrenal hyperplasia due to different inherited mutations in the potassium channel KCNJ5. *Proc Natl Acad Sci U S A*
12. **Mulatero P, Tauber P, Zennaro MC, Monticone S, Lang K, Beuschlein F, Fischer E, Tizzani D, Pallauf A, Viola A, Amar L, Williams TA, Strom TM, Graf E, Bandulik S, Penton D, Plouin PF, Warth R, Allolio B, Jeunemaitre X, Veglio F, Reincke M** 2012 KCNJ5 Mutations in European Families With Nonglucocorticoid Remediable Familial Hyperaldosteronism. *Hypertension* 59:235-240
13. **Boukroun S, Beuschlein F, Rossi GP, Golib-Dzib JF, Fischer E, Amar L, Mulatero P, Samson-Couterie B, Hahner S, Quinkler M, Fallo F, Letizia C, Allolio B, Ceolotto G, Cicala MV, Lang K, Lefebvre H, Lenzini L, Maniero C, Monticone S, Perrocheau M, Pilon C, Plouin PF, Rayes N, Seccia TM, Veglio F, Williams TA, Zinamosca L, Mantero F, Benecke A, Jeunemaitre X, Reincke M,**

- Zennaro MC** 2012 Prevalence, Clinical, and Molecular Correlates of KCNJ5 Mutations in Primary Aldosteronism. Hypertension
14. **Azizan EA, Murthy M, Stowasser M, Gordon R, Kowalski B, Xu S, Brown MJ, O'Shaughnessy KM** 2012 Somatic Mutations Affecting the Selectivity Filter of KCNJ5 Are Frequent in 2 Large Unselected Collections of Adrenal Aldosteronomas. Hypertension
 15. **Taguchi R, Yamada M, Nakajima Y, Satoh T, Hashimoto K, Shibusawa N, Ozawa A, Okada S, Rokutanda N, Takata D, Koibuchi Y, Horiguchi J, Oyama T, Takeyoshi I, Mori M** 2012 Expression and Mutations of KCNJ5 mRNA in Japanese Patients with Aldosterone-Producing Adenomas. *J Clin Endocrinol Metab*
 16. **Sausbier M, Arntz C, Bucurenciu I, Zhao H, Zhou XB, Sausbier U, Feil S, Kamm S, Essin K, Sailer CA, Abdullah U, Krippeit-Drews P, Feil R, Hofmann F, Knaus HG, Kenyon C, Shipston MJ, Storm JF, Neuhuber W, Korth M, Schubert R, Gollasch M, Ruth P** 2005 Elevated blood pressure linked to primary hyperaldosteronism and impaired vasodilation in BK channel-deficient mice. *Circulation* 112:60-68
 17. **Grimm PR, Irsik DL, Settles DC, Holtzclaw JD, Sansom SC** 2009 Hypertension of *Kcnmb1*^{-/-} is linked to deficient K secretion and aldosteronism. *Proc Natl Acad Sci U S A* 106:11800-11805
 18. **Heitzmann D, Derand R, Jungbauer S, Bandulik S, Sterner C, Schweda F, El Wakil A, Lalli E, Guy N, Mengual R, Reichold M, Tegtmeier I, Bendahhou S, Gomez-Sanchez CE, Aller MI, Wisden W, Weber A, Lesage F, Warth R, Barhanin J** 2008 Invalidation of TASK1 potassium channels disrupts adrenal gland zonation and mineralocorticoid homeostasis. *Embo J* 27:179-187
 19. **Davies LA, Hu C, Guagliardo NA, Sen N, Chen X, Talley EM, Carey RM, Bayliss DA, Barrett PQ** 2008 TASK channel deletion in mice causes primary hyperaldosteronism. *Proc Natl Acad Sci U S A* 105:2203-2208
 20. **Arrighi I, Bloch-Faure M, Grahammer F, Bleich M, Warth R, Mengual R, Drici MD, Barhanin J, Meneton P** 2001 Altered potassium balance and aldosterone secretion in a mouse model of human congenital long QT syndrome. *Proc Natl Acad Sci U S A* 98:8792-8797. Epub 2001 Jul 8793.

Table 1. Mouse models inactivated for potassium channels featuring adrenal aldosterone hypersecretion.

Inactivated channel (gene)	PA Phenotype	Adrenal cortex expression	Hyperplasia/ APA	Gender difference described	Ref
maxiK (<i>KCNMA1/KCNMB1</i>)	<i>KCNMA1</i> KO: Hyperaldosteronism, hypokalemia, normal renin levels; <i>KCNMB1</i> KO: hyperaldosteronism linked to renal K retention and hyperkalemia	<i>KCNMA1</i> : ZG>>ZF, ZR <i>KCNMB1</i> : adrenal gland	NO	NO	(16-17)
TASK1 (<i>KCNK3</i>)	Glucocorticoid remediable hyperaldosteronism, decreased plasma renin, hypokalemia,	ZG, ZF, ZR	NO	Females only	(18)
TASK1/TASK3 (<i>KCNK3/KCNK9</i>)	Hyperaldosteronism, reduced plasma renin,	TASK3: ZG	NO	NO	(19)
KvLQT1/IsK (<i>KCNQ1/KCNE1</i>)	Hyperaldosteronism in <i>KCNE1</i> KO mice, normal renin, fecal sodium and potassium loss	ZG (<i>KCNQ1, KCNE1</i>)	NO	NO	(20)