Cardiovascular Topics

A primary aldosteronism-like phenotype identified with the aldosterone-to-angiotensin II ratio in black men: the SABPA study

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Abstract

Introduction: Black populations may be more likely to have primary aldosteronism (PA) due to adrenal hyperplasia or other forms of adrenal hyperactivity, with suppressed renin levels and high levels of aldosterone, which may contribute to the development of hypertension.

Methods: This sub-study involved 35 black men matched for age, gender and race, and aged 20–65 years, living in the North West Province of South Africa. RAAS triple-A analysis was carried out with LC-MS/MS quantification. Blood pressure, electrocardiography and other variables were determined with known methods.

Results: Hypertensive subjects with higher aldosterone levels showed an increased aldosterone–angiotensin II ratio (AA2 ratio) compared to the hypertensive subjects with low aldosterone levels (10.2 vs 3.0 pmol/l; $p = 0.003$). The serum potassium concentration was significantly lower in the high-aldosterone group and the serum sodium–potassium ratio was significantly higher compared to the low-aldosterone group (3.9 vs 4.5, $p = 0.016$, 34.8 vs 31.8, $p = 0.032$, respectively). Furthermore, aldosterone was positively associated with both left ventricular hypertrophy (Cornell product) (Spearman $R = 0.560$; $p = 0.037$) and kidney function [albumin-to-creatinine ratio (ACR)] (Spearman $R = 0.589$, $p = 0.021$) in the hypertensive high-serum aldosterone group.

Conclusions: The AA2 ratio, a novel screening test that is currently being validated for PA case detection, was used to identify a PA-like phenotype in black men. Excess aldosterone was associated with endothelial dysfunction and left ventricular hypertrophy, independent of blood pressure.

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Although the incidence of high blood pressure has decreased worldwide since 1975, a shift is reported from high- to low-income countries, such as those in sub-Saharan Africa.[1,2] In blacks, hypertension is characterised by a greater retention of salt and water by the kidney, with suppressed levels of renin and aldosterone.[3] Black populations may also be more likely to have primary aldosteronism (PA) due to adrenal hyperplasia or other forms of adrenal hyperactivity, with suppressed renin levels and high levels of aldosterone.[3,4]

Furthermore, black populations may also have a greater sensitivity of blood pressure to the increased secretion of aldosterone and are more likely to have hypertension.[5] PA is an overlooked but frequent cause of secondary hypertension. In a recent survey in Italy and Germany, it was found that only 7–8% of general practitioners ordered aldosterone and renin measurements, and the prevalence of diagnosed PA was only 1% of hypertensive patients.[6] The consequence is that only 1% of patients in Italy and 2% in Germany are diagnosed with the disease. From recent studies, a prevalence approaching 5–13% was found.[7-9]

From the literature, it seems that PA is a largely unrecognised and undertreated cause of hypertension. In South Africa, the case may not be different due to the fact that awareness (27%), treatment (18%) and control rates (7%) for hypertension are low.[2] PA is also a common occurrence in resistant hypertension and screening for it may improve hypertension treatment, which is already a challenge in South Africa.

We therefore aimed to evaluate the role of aldosterone as a contributory factor in hypertension in a black cohort by

identifying a PA-like phenotype with the use of the novel aldosterone–angiotensin II ratio (AA2 ratio). One parameter obtained in RAAS triple-A testing, the simultaneous LC-MS/MS-based quantification of angiotensin I (Ang I), angiotensin II (Ang II) and aldosterone, was obtained in patient samples. Concerns are raised about the accuracy of renin assays and therefore new mass spectrometric methods were employed for measuring angiotensin II, which are currently being assessed in the clinical setting.

Methods

The baseline Sympathetic Activity and Ambulatory Blood Pressure in Africans (SABPA) study was conducted in the North West Province, South Africa, during 2008 and 2009. The study was a target-population, comparative study and included black teachers aged between 20 and 65 years. All participants were working as teachers for the Department of Education in one of the four Dr Kenneth Kaunda education districts of the North West Province. The study has been well-described elsewhere.[10]

This sub-study forms part of the SABPA study and for this study, 35 black men, matched for age, gender and race, were divided into normotensive (n = 7) and hypertensive participants (n = 27). The hypertensive men were further divided into low- (n = 12) and high- (n = 15) aldosterone groups based on the median value (133.2 pmol/l) of aldosterone observed in the cohort.

The data of one participant was omitted from the analysis because of angiotensin receptor blocker (ARB) therapy, which is known to affect the AA2 ratio. The other participants receiving anti-hypertensive medication potentially interfering with classical PA screening assays were not excluded from this study as recent data suggest that the AA2 ratio is less prone to drug-mediated suppression, as described for the aldosterone-to-renin ratio.[11] Exclusion from the overarching SABPA study was based on the following criteria: ear temperature > 37.5°C, being vaccinated or having donated blood in the three months before the study commenced, clinically confirmed diabetes, and known HIV infection.

All participants signed an informed consent form. The study complied with all applicable regulations, in particular, the Helsinki Declaration of 1975 (as revised in 2008) for investigation of human participants.[12] The Ethics Review Board of the North-West University, Potchefstroom, South Africa, approved the study (NWU-00036-07-A6).

Upon arriving at the North-West University overnight facilities (consisting of 10 bedrooms, two bathrooms, kitchen, dining room and television room), participants were introduced to the experimental set-up to lessen anticipatory stress.[13] They received a standardised dinner and had their last beverages (tea/coffee) and two biscuits at 20:30 hours. The following morning a fasting overnight urine sample was obtained, followed by the anthropometric measurements.

Height and weight of participants were measured using calibrated instruments (Precision Health Scale, A & D Company, Tokyo, Japan; Invicta Stadiometer, IP 1465, UK). Measurements were taken in triplicate using standardised methods,[14] and body mass index (BMI) was calculated. Participants completed a general health questionnaire on family history, diagnosis of hypertension and renal disease, as well as medication use.

Hereafter participants remained in a semi-recumbent position for at least 30 minutes before blood pressure was measured with a sphygmomanometer using appropriate-sized cuffs. Measurements were executed in duplicate with five-minute intervals and the second measurement was used for analysis. This blood pressure reading obtained with the sphygmomanometer was used to classify the participants as hypertensive (≥ 140/90 mmHg).

A resting 12-lead electrocardiogram (ECG) of six cardiac cycles (Norav NHH1200®, Kiryat Bialik, Israel) was determined for each participant. Data from the 12-lead ECG was used to determine the Cornell product [(RaVL + SV3) × QRS duration]. Values > 244 mV/ms are indicative of left ventricular hypertrophy (LVH).

Silent myocardial ischaemic events were assessed by two-channel 24-hour (ECG) recordings (Cardiotens CE120®, Meditech, Budapest, Hungary) for 20 seconds at five-minute intervals. Before the start of the ambulatory investigation, the isoelectric reference point (PQ segment), J point, L point (80 ms after the J point) and an ST-segment detection interval of at least 3 mm as the initial ST level, were calculated individually for each participant.

An ischaemic event was recorded according to the following criteria: horizontal or descending ST-segment depression of at least 1 mm; duration of the ST-segment episode lasting ≥ one minute, and a ≥ one-minute interval from the preceding episode. In case of a horizontal or descending ST depression (1 mm: 1-min duration at a 1-min interval from the preceding episode), an ECG tracing lasting 60 seconds was recorded and an additional blood pressure measurement was automatically initiated by the trigger mechanism of the device. Data were analysed using CardioVisions 1.19 Personal Edition (Meditech, Budapest, Hungary).

Further cardiovascular variables were recorded continuously for five minutes with the Finometer (Finapres Medical Systems, Amsterdam, The Netherlands) device. Finometer measurements were processed with Beatscope 1.1 software (FMS, Finapres Medical Systems, Amsterdam, The Netherlands) from the reconstructed pressure waveform to obtain the stroke volume (SV), total peripheral resistance (TPR) and the Windkessel compliance (C_{wk}). Carotid-dorsalis pedis pulse-wave velocity (c-pPWV) was obtained with the Complior acquisition system (Artech-Medical, Pantin, France).

Hereafter, fasting blood samples were collected from the participants' right arm brachial vein branches with a sterile winged infusion set. The samples were handled and prepared according to standardised procedures. Serum and plasma samples were stored at –80°C until analysis.

For RAAS triple-A testing, serum was allowed to clot for 30 minutes at room temperature before centrifugation (4 700 rpm, 20°C, 15 minutes) on the Hettich 320 centrifuge (Andrew Hettich, GmbH & Co, KG, Germany). The supernatant was then stored at –80°C until analysis.

Following pH-controlled (7.4) *ex vivo* equilibration at 37°C for one hour, serum was stabilised and subjected to LC-MS/MS quantification of equilibrium (eq) angiotensin peptide levels (Attoquant Diagnostics, Vienna, Austria). Briefly, stable isotope-labelled internal standards for Ang I, Ang II and aldosterone were spiked to the samples at a concentration of 500 pg/ml. Following C18-based solid-phase extraction, samples were subjected to LC-MS/MS analysis using a reverse-phase analytical column (Acquity UPLC® C18, Waters) operating in

line with a XEVO TQ-S triple quadrupole mass spectrometer (Waters) in MRM mode.

Two different mass transitions were measured per peptide, and angiotensin concentrations were calculated from internal standard-normalised signals under consideration of the corresponding response factors determined by calibration curves prepared in the original sample matrix. A signal-to-noise ratio of 10 was considered as quantification threshold for endogenous peptide signals, resulting in indicated lower levels of quantification.

At 50 pmol/l the inter-assay coefficients of variability (CV) for Ang II and aldosterone were 6.1 and 7.9%, respectively. The intra-assay CVs for Ang II and aldosterone were 4.4 and 5.2%, respectively. The functional sensitivity for Ang II and aldosterone measurement was > 2.0 and > 14.0 pmol/l, respectively. The angiotensin-based biomarkers AA2 ratio (aldosterone/eq Ang II), PRA-S (eq Ang I + eq Ang II) and ACE-S (eq Ang II/eq Ang I) were calculated from molar concentrations of respective analytes.

Adrenocorticotrophic hormone (ACTH) was analysed with electro-chemiluminescence immunoassay (ECLIA), e411 (Roche, Basel, Switzerland). Inter- and intra-batch variability were 5.4 and 2.9%, respectively. Serum cortisol was analysed using an electro-chemiluminescence immunoassay on the Elecsys 2010

Table 1. Characteristics of the black men with low and high serum aldosterone levels

Variables	*Normotensive* (n = *7*)	*Hypertensive, aldosterone ≤ 133.2 pmol/l* (n = *12*)	*Hypertensive, aldosterone > 133.2 pmol/l* (n = *15*)	p-*value*[a]
Age (years)	45.0 (45.0–54.0)	50.0 (45.0–53.0)	47.0 (45.0–50.0)	0.373
BMI (kg/m^2)	25.4 (23.5–27.3)	28.8 (23.3–34.4)	28.1 (24.8–30.4)	0.792
Cardiovascular variables				
SBP (mmHg)	132.0 (120.0–138.0)	150.0 (140.0–156.5)	155.0 (130.0–180.0)	0.829
DBP (mmHg)	82.0 (80.0–88.0)	100.0 (95.0–106.5	110.0 (85.0–120.0)	0.548
SV (ml)	92.7 (87.0–142.1)	85.8 (75.8–105.4)	105.9 (83.8–113.5)	0.373
TPR (mmHg/ml/s)	0.96 (0.75–1.02)	1.12 (1.02–1.30)	0.97 (0.88–1.48)	0.516
C_{wk} (ml/mmHg)	1.81 (1.65–2.02)	1.59 (1.33–1.81)	1.84 (1.22–2.02)	0.399
c-pPWV (m/s)	10.2 (8.7–10.3)	9.6(9.4–11.6)	10.5 (9.5–11.0)	0.860
Biochemical variables				
eq Ang I (pmol/l)	10.9 (3.1–23.3)	14.7 (3.5–21.4)	7.6 (3.1–33.4)	0.755
eq Ang II (pmol/l)	33.2 (12.7–58.4)	44.0 (19.0–76.9)	25.7 (10.3–55.2)	0.277
ACE-S (eq AngII/eq AngI) (pmol/l)	2.8 (2.1–4.7)	3.5 (2.4–5.4)	2.30 (0.5–4.9)	0.183
PRA-S (Ang I + Ang II) (pmol/l)	48.8 (15.8–81.7)	53.1 (25.3–102.6)	34.9 (13.4–83.7)	0.373
Aldosterone (pmol/l)	88.4 (71.7–146.0)	101.8 (88.0–126.5)	253.3 (163.9–341.2)	< 0.001
AA2 ratio	2.7 (1.8–10.5)	3.0 (1.2–6.3)	10.2 (4.4–47.6)	0.003
Aldosterone/PRA-S	1.8 (1.3–7.0)	2.3 (0.9–4.4)	5.8 (3.1–22.0)	0.010
sACTH (pg/ml)	11.9 (9.3–33.6)	17.6 (11.8–29.4)	18.4 (12.4–25.9)	0.981
sCortisol (nmol/l	405.2 (255.2–438.0	371.3 (307.1–471.5)	347.3 (276.7–487.6)	0.943
Serum Na^+ (mmol/l)	150.4 (127.3–173.5)	145.7 (125.9–177.2)	126.7 (124.4–132.6)	0.183
Serum K^+ (mmol/l)	5.0 (4.4–5.2)	4.5 (4.0–5.7)	3.9 (3.6–4.4)	0.016
Serum Na^+–K^+ ratio	31.9 (29.4–32.8)	31.8 (29.2–33.0)	34.8 (31.5–35.5)	0.032
Urinary Na^+ (mmol/l)	91.0 (62.0–112.0)	90.0 (66.0–139.0)	86.0 (44.0–107.0)	0.474
Urinary K^+ (mmol/l)	14.0 (8.0–21.0)	18.0 (12.5–22.5)	14.0 (13.0–24.1)	0.867
Urinary Na^+–K^+ ratio	6.5 (5.3–7.8)	6.1 (4.6–7.6)	6.0 (2.5–6.6)	0.470
CRP (mg/l)	3.5 (2.9–4.9)	3.1 (1.7–5.2)	3.3 (2.1–9.3)	0.456
End-organ variables				
Cornell product (> 244 mV/ms)	51.6 (30.2–80.6)	49.9 (40.0–111.9)	93.5 (59.2–141.1)	0.134
Silent 24-h ST events (*n*)	0.0 (0.0–3.0)	12.0 (0.0–25.0)	1.0 (0.0–7.0)	0.507
Est creatinine clearance	111.4 (102.8–127.9)	112.4 (97.3–139.3)	134.7 (113.2–154.1)	0.126
ACR	0.95 (0.72–1.87)	0.99 (0.62–2.77)	1.19 (0.78–1.84)	0.574
Lifestyle variables				
Cotinine (ng/ml)	0.01 (0.01–30.00)	8.51 (0.01–28.01)	0.01 (0.01–61.01)	0.683
GGT (U/l)	53.9 (44.4–130.1)	77.0 (40.5–111.3)	57.4 (42.0–76.3)	0.548
TEE (kcal/day)	2339.9 (2228.7–2559.3)	2119.6 (1818.2–3198.2)	2627.2 (2436.5–3845.1)	0.126
Medication use, *n* (%)				
SNS blocker	–	0 (0)	1 (6.7)	–
ACE inhibitor	–	0 (0)	5 (33.3)	–
Thiazide	–	2 (16.7)	2 (13.3)	–
Calcium antagonist	–	0 (0)	6 (40)	–
Beta-blocker	–	0 (0)	2 (13.3)	–

Data presented as median (lower; upper quartile). [a]2 × 1-sided exact *p*-value between high- and low-aldosterone hypertensives.
BMI: body mass index (kg/m^2); SBP, DBP: systolic and diastolic blood pressure (mmHg), respectively; SV: stroke volume (ml); TPR: total peripheral resistance (mmHg/s/ml); C_{wk}: Windkessel compliance (ml/mmHg); c-pPWV: carotid-pedalis pulse-wave velocity (m/s); eq Ang I and eq Ang II: angiotensin I and angiotensin II (pmol/l); ACE-S: angiotensin-based ACE activity (eq AngII/eq AngI, pmol/l); PRA-S: angiotensin-based renin activity (eq Ang I + eq Ang II, pmol/l); AA2 ratio: aldosterone–angiotensin II ratio; sACTH: serum adrenocorticotrophic hormone (pg/ml); CRP: C-reactive protein (mg/l); ACR: albumin–creatinine ratio; GGT: gamma-glutamyltransferase (U/L); TEE: total energy expenditure (kcal/day); CNS blocker: central nervous system blocker; ACE inhibitor: angiotensin converting enzyme inhibitor.

apparatus (Roche, Basel, Switzerland). Both the intra- and inter-assay coefficients of variation for all the assays were less than 10%.

Serum and urinary sodium and potassium concentrations were determined making use of the Konelab TM 20i sequential multiple analyser computer (SMAC) (ThermoScientific, Vantaa, Finland). Gamma-glutamyltransferase (GGT), cotinine and high-sensitivity C-reactive protein (CRP) were analysed using the sequential multiple analyser (Konelab 20i; Thermo Scientific, Vantaa, Finland; Unicel DXC 800 – Beckman and Coulter®, Germany). The intra- and inter-coefficients of variation for all assays were below 10%.

The urinary creatinine from an eight-hour overnight fasting urine sample was determined with a calorimetric method. Albumin was determined with the turbidimetric method on a Unicel DXC 800 apparatus (Beckman and Coulter, Germany) (CV% 1.7–3.3%).

The total energy expenditure (TEE) (kcal) in 24 hours was determined using the Actical® activity monitor (Mini Mitter Co, Inc, Bend, OR; Montreal, Quebec, Canada).

Statistical analysis

Data were analysed with the TIBCO® Statistica™, version 13.3 (Palo Alto, CA, USA). Data are presented as median values with lower and upper quartiles. Due to the small sample size, non-parametric statistics were used. The Mann–Whitney *U*-test was used to determine significance between the hypertensive participants with low and high aldosterone levels. Probability values of $p \leq 0.05$ were regarded as significant. Spearman rank order correlations of aldosterone with the variables were also determined.

Results

The characteristics of the normotensive and hypertensive black men with low and high serum aldosterone levels are described in Table 1. The AA2 ratio, which is currently under evaluation to be used as a novel marker for primary aldosteronism, was significantly higher in the hypertensive high-aldosterone group compared to the hypertensive low-aldosterone group (10.2 vs 3.0; $p = 0.003$). A lower value of 2.7 for the AA2 ratio was encountered in the normotensive participants.

The serum potassium (K^+) was significantly lower and the serum sodium-to-potassium (Na^+–K^+) ratio significantly higher in the hypertensive high-aldosterone group compared to the low-aldosterone group (3.9 vs 4.5, $p = 0.016$, 34.8 vs 31.8, $p = 0.032$ respectively). No differences existed between Ang I, Ang II, PRA-S and ACE-S in the hypertensive low- and high-aldosterone groups although levels appeared non-significantly suppressed in the hypertensive high-aldosterone group (7.6 vs 14.7 pmol/l, $p = 0.755$; 25.7 vs 44.0 pmol/l, $p = 0.277$ and 34.9 vs 53.1 pmol/l, $p = 0.373$, respectively). The medication use is also shown in Table 1.

In Table 2, potassium in the hypertensive high-aldosterone men associated negatively and was borderline significant with aldosterone (Spearman $R = –0.496$, $p = 0.060$). The total peripheral resistance was positively associated with aldosterone only in the hypertensive low-aldosterone group (Spearman $R = 0.699$, $p = 0.011$). Arterial compliance associated negatively and was borderline significant (Spearman $R = –0.511$, $p = 0.052$), and cortisol associated positively with aldosterone in the hypertensive high-aldosterone group (Spearman $R = 0.500$, $p = 0.058$). Ang I was inversely associated with aldosterone in the hypertensive low-aldosterone group (Spearman $R = –0.606$, $p = 0.037$). Aldosterone, in the hypertensive high-aldosterone group associated positively and significantly with both Cornell product (Spearman $R = 0.560$; $p = 0.037$) and ACR (Spearman $R = 0.589$, $p = 0.021$).

Discussion

The primary aim of this sub-study was to evaluate the role of aldosterone as contributory factor of hypertension in a black cohort by making use of the novel AA2 ratio.[11] The main finding of this study was a higher AA2 ratio in the hypertensive high-aldosterone compared to the hypertensive low-aldosterone group, suggesting a PA-like condition represented by Ang II, and independent aldosterone secretion to be a major cause of hypertension in this subgroup.

The serum K^+ concentration was significantly lower in the hypertensive high-aldosterone group and the serum Na^+–K^+ ratio was significantly higher compared to the hypertensive

Table 2. Spearman rank order correlations of aldosterone with independent variables in hypertensive black men with low ($n = 12$) (≤ 133.2 pmol/l) and high ($n = 15$) (> 133.2 pmol/l) aldosterone levels

	Hypertensive, aldosterone ≤ 133.2 pmol/l		*Hypertensive, aldosterone > 133.2 pmol/l*	
Variables	*Spearman* R	p-*value*	*Spearman* R	p-*value*
BMI (kg/m²)	0.007	0.983	–0.014	0.960
Cardiovascular variables				
SBP (mmHg)	0.043	0.896	0.337	0.219
DBP (mmHg)	0.380	0.224	0.259	0.350
TPR (mmHg/ml/s)	0.699	0.011	0.400	0.140
C_{wk} (ml/mmHg)	–0.126	0.697	–0.511	0.052
c-pPWV (m/s)	–0.074	0.820	0.233	0.546
Biochemical variables				
eq Ang I (pmol/l)	–0.606	0.037	–0.084	0.767
eq Ang II (pmol/l)	–0.510	0.090	0.161	0.566
ACE-S (pmol/l)	0.378	0.226	–0.077	0.785
PRA-S (pmol/l)	–0.552	0.063	0.206	0.462
sACTH (pg/ml)	–0.207	0.519	0.218	0.435
sCortisol (nmol/l)	–0.105	0.746	0.500	0.058
Serum Na^+ (mmol/l)	0.287	0.366	–0.204	0.467
Serum K^+ (mmol/l)	–0.196	0.542	–0.496	0.060
Serum Na^+–K^+ ratio	0.559	0.059	0.389	0.152
Urinary Na^+ (mmol/l)	0.400	0.223	0.390	0.150
Urinary K^+ (mmol/l)	–0.193	0.549	–0.058	0.839
Urinary Na^+–K^+ ratio	0.137	0.655	0.478	0.098
End-organ variables				
Cornell product (> 244 mV/ms)	0.001	0.999	0.560	0.037
Silent 24-h ST events (*n*)	–0.233	0.491	0.122	0.664
Est creatinine clearance (ml/min)	0.231	0.471	–0.304	0.271
ACR	–0.100	0.770	0.589	0.021

BMI: body mass index (kg/m²); SBP, DBP: systolic and diastolic blood pressure (mmHg), respectively; SV: stroke volume (ml); TPR: total peripheral resistance (mmHg/s/ml); C_{wk}: Windkessel compliance (ml/mmHg); c-pPWV: carotid-pedalis pulse-wave velocity (m/s); Ang I and Ang II: angiotensin I and angiotensin II (pmol/l); ACE-S: surrogate for angiotensin converting enzyme (AngII/AngI, pmol/l); PRA-S: surrogate for renin activity (Ang I + Ang II, pmol/l); sACTH: serum adrenocorticotrophic hormone (pg/ml); ACR: albumin–creatinine ratio; *p*-values ≤ 0.05 regarded as significant.

low-aldosterone group. Furthermore, aldosterone was positively associated with both left ventricular hypertrophy (Cornell product) and kidney function (ACR) in the hypertensive high-aldosterone group.

The aldosterone-to-renin ratio (ARR) is the recommended screening test for PA.[15,16] Measurement and interpretation is challenging when using the ARR because several antihypertensive drugs interfere with the RAAS,[15] resulting in an increase in renin concentration and activity, which subsequently suppresses the ARR, resulting in false-negative test results. There is a need for a versatile PA screening assay that does not interfere with anti-hypertensive treatments and therefore allows a more specific identification of PA in hypertensive patients on therapy.

Preliminary data have shown that in contrast to the ARR, the AA2 ratio remains unaffected by angiotensin converting enzyme (ACE) inhibitor therapy and may therefore be a valuable alternative to currently employed screening assays.[11] In the current study, due to the small sample size and participants utilising different hypertensive medication, we could not explore the AA2 ratio in full for the diagnosis of PA.

The PA-like phenotype in hypertensive black men was further characterised by a decrease in serum K^+ levels and was associated with higher aldosterone levels, which may aggravate cardiovascular complications.[17] From the literature, it is also evident that the overall prevalence of co-morbidities was higher in hypokalaemic PA patients than in normokalaemic patients.[17] In more than 22 000 Pakistani patients, the risk of sudden cardiac attack or sudden cardiac death and all-cause mortality was associated with hyperkalaemia,[18] however no significant relationship existed between hypokalaemia and outcome.

The association between left ventricular hypertrophy (Cornell product) and aldosterone rather supports cardiac co-morbidities, as excess aldosterone levels might be a risk factor for arrhythmic disorders occurring either via left ventricular hypertrophy or cardiac fibrosis.[19] Furthermore, aldosterone has been associated with endothelial dysfunction,[17] and now also with a lack of arterial compliance, independent of blood pressure.[19] Lower K^+ levels and a lack of compliance concurrently with high aldosterone levels and associated ACR (a marker of kidney and endothelial dysfunction) may be detrimental to the cardiovascular health of hypertensive black men.

Blood pressure control was driven by the TPR and renin–angiotensin system (see Table 2) in the hypertensive low-aldosterone group. Aldosterone is a mineralocorticoid hormone, having 30–50% of its total plasma concentration in free form. Cortisol on the other hand is a glucocorticoid hormone and has 100-fold higher free levels in circulation than aldosterone, with high intrinsic mineralocorticoid activity, although its action is blunted by local conversion to cortisone at the kidney level.[20] Cortisol can bind with the same high affinity as aldosterone to the mineralocorticoid receptor[20] and it may modulate the mineralocorticoid receptor-binding effects of aldosterone.

From the literature, it is evident that the long-term increase in both cortisol and aldosterone reflect changes in risk factors for cardiovascular disease, such as increases in dietary fat, high salt intake, low levels of physical activity and high stress levels, which may lead to the phenotype of aldosterone-associated hypertension.[20] Stress can indeed alter the aldosterone phenotype, as chronic depression was associated with a desensitised renin system and volume-loading hypertension in a black cohort over three years.[21]

Our finding of a positive association between aldosterone and cortisol ($p = 0.058$) in the hypertensive high-aldosterone group further enhances previous findings where hypothalamic–pituitary–adrenal axis dysregulation and compensatory double product (systolic blood pressure × heart rate) increased and acted as a possible defence mechanism to alleviate perfusion deficits, which potentiated ischaemic heart disease risk.[22] The interaction of endogenous levels of aldosterone and cortisol may therefore disrupt blood pressure control and result in an increased proportion of hypertension.

Low-renin hypertension is more common in blacks and is characterised by increased Na^+ retention and suppressed renin and aldosterone levels,[3,21,23] and may be psychosocial in origin.[21, 23] Salt and water retention are greater in blacks, not only because there is an increased likelihood of PA, but also because of genetic variants that affect the function of the renal tubular epithelial sodium channel (ENaC). There are a number of genetic causes of both these phenotypes.[24,25]

PA is an overlooked but frequent cause of secondary hypertension and because only a few hypertensive patients are screened worldwide for PA,[6] mineralocorticoid blockade is also unlikely to be used as treatment for hypertension. It is also worth mentioning that low levels of both aldosterone and angiotensin II would identify patients with a Liddle syndrome phenotype, who would respond best to amiloride.[26,27]

Our data highlight the fact that hypertensive black patients with low K^+ levels should be screened for PA in order to treat them adequately so as to bring the high prevalence of hypertension in sub-Saharan Africa under control. Because PA is also a common occurrence in resistant hypertension, screening for it may further improve hypertension treatment and control in South Africa.

A strength of this sub-study is that use was made of state-of-the-art analysis of the RAAS parameters. A weakness is that follow-up data were not used to determine causality.

Conclusion

The AA2 ratio, which should be explored further to replace the aldosterone-to-renin ratio for the diagnosis of PA, was used to identify the PA-like phenotype in black men. Excess aldosterone was associated with endothelial dysfunction and left ventricular hypertrophy, independent of blood pressure.

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