Low-dose hydrocortisone replacement is associated with improved arterial stiffness index and blood pressure dynamics in severely adrenocorticotrophin-deficient hypopituitary male patients

Lucy-Ann Behan1, David Carmody1, Bairbre Rogers1, Mark J Hannon1, Colin Davenport1, William Tormey2,3, Diarmuid Smith1, Christopher J Thompson1, Alice Stanton4 and Amar Agha1

1Department of Endocrinology, Beaumont Hospital and RCSI Medical School, Dublin, Ireland, 2Department of Chemical Pathology, Beaumont Hospital, Dublin, Ireland, 3Biomedical Sciences, Ulster University, Coleraine, Northern Ireland, UK, and 4Department of Molecular and Cellular Therapeutics, RCSI Research Institute, Dublin, Ireland

Abstract

Objective: Increased cardiovascular and cerebrovascular morbidity and mortality in hypopituitary subjects may be linked to inappropriate glucocorticoid exposure; however, the pathophysiology remains unclear. We aimed to examine the effect of three commonly prescribed hydrocortisone (HC) regimens on vascular risk factors.

Design: An open crossover study randomising ten hypopituitary men with severe adrenocorticotrophic hormone deficiency to three HC dose regimens: dose A (20 mg mane and 10 mg tarde), dose B (10 mg mane and 10 mg tarde) and dose C (10 mg mane and 5 mg tarde).

Methods: Following 6 weeks on each regimen, participants underwent 24-h serum cortisol sampling, 24-h ambulatory blood pressure (BP) measurements, calculation of the Ambulatory Arterial Stiffness Index (AASI), oral glucose tolerance testing and fasting serum osteoprotegerin (OPG) sampling.

Results: There were no differences in 24-h BP between dose regimens and controls; however, low-dose HC replacement (dose C) was associated with the lowest AASI, indicating a less stiff arterial tree (P < 0.05) compared with the other dose regimens. Loss of the physiologic nocturnal BP dip was more common in higher HC replacement regimens, although only significant for dose B compared with dose C (P = 0.03). Twenty per cent of patients had abnormal glucose tolerance, but this was unrelated to dose regimen. OPG correlated strongly with 24-h BP in those on dose A only (r = 0.65, P = 0.04).

Conclusion: Currently prescribed HC replacement doses do not result in significant differences in absolute BP levels or improvements in insulin sensitivity. However, lower HC doses may result in lower arterial stiffness and a more physiological nocturnal BP dip. Long-term studies are required to confirm these findings and evaluate their impact on vascular morbidity in this patient group.

Introduction

Adults with hypopituitarism have increased mortality primarily due to cardiovascular and cerebrovascular diseases (1, 2, 3, 4, 5, 6), yet the pathophysiologic cause remains speculative and probably multifactorial. Up to 70% of patients with hypopituitarism have varying degrees of adrenocorticotropic hormone (ACTH)–cortisol
deficiency (1), and it has been suggested that inappropriate glucocorticoid (GC) over-replacement may result in subtle chronic overexposure to cortisol, leading to increased morbidity in this group. This hypothesis is supported by evidence from recent studies in subjects with treated acromegaly and non-functioning pituitary adenomas, which have suggested that higher doses of GC replacement are an independent risk factor for increased mortality (7, 8).

Although the vascular endothelium has 11β-hydroxysteroid dehydrogenase type 2 enzymatic activity, which inactivates cortisol to cortisone, this enzyme can become saturated at times of increased cortisol exposure, and the protective effect may be lost. Despite this potential physiologic role of hypercortisolism in vascular morbidity, there are conflicting data regarding the contribution of hypertension to morbidity and mortality in hypopituitarism. In the general population, arterial hypertension remains the primary risk factor for cerebrovascular and cardiovascular diseases. Among adult hypopituitary patients, some studies have shown no difference in absolute systolic or diastolic blood pressure (BP) compared with controls (9, 10), whereas other studies have demonstrated increased the prevalence of treated hypertension (11) or even lower 24-h ambulatory BP in growth hormone (GH)-deficient hypopituitary subjects compared with matched controls (12). Although such conflicting data may question the link between BP and the increased vascular mortality seen in this group, with the exception of one study, the others assessed BP in hypopituitarism using clinic BP recordings rather than 24-h ambulatory measurements; pituitary hormone replacement status was variable and few studies made comments regarding concurrent anti-hypertensive medication use. Vascular disease is associated with increasing stiffness of the vascular tree and with changes in BP dynamics, in particular loss of the physiological nocturnal BP dip (13). Persons and coworkers demonstrated reduced endothelial function in 17 subjects with secondary adrenal insufficiency, on varying types of GC replacement and variable pituitary hormone deficiencies, following an increase in hydrocortisone (HC) equivalent from 20 to 30 mg daily for 7 days (14). To our knowledge, no other group has evaluated the effect of commonly prescribed doses of HC replacement in this group of subjects on arterial stiffness or on BP dynamics.

Abnormal glucose homeostasis is also an established risk factor for cardiovascular disease, and insulin resistance has been shown to be associated with increased mortality (15). Even when blood glucose and plasma insulin levels are similar to those seen in controls, GH-deficient (GHD) pituitary patients are insulin resistant (16). A small number of studies have prospectively examined the effect of different GC regimens and doses in hypopituitary patients on glucose metabolism compared with matched controls and failed to demonstrate any difference in fasting glucose or insulin levels (12, 17, 18); however, in those studies, GHD subjects were not replaced with GH and only fasting levels were assessed.

In view of the limited and discrepant data regarding the effect of GC replacement in hypopituitary subjects on widely accepted vascular risk factors, we aimed to examine, in a prospective randomised-controlled manner, the effect of three commonly prescribed regimens of HC replacement on markers of cardiometabolic outcome in a group of panhypopituitary adults with severe ACTH deficiency, fully replaced with all pituitary hormones, including GH, and compared with age-, sex- and BMI-matched controls.

Subjects and methods

Patients and controls

Ten adult hypopituitary men with known severe ACTH deficiency, defined by a fasting morning total serum cortisol concentration <100 nmol/L and a stimulated peak cortisol of <400 nmol/L in response to insulin-induced hypoglycaemia (nadir glucose <2.2 mmol/L with symptoms), were included.

All ten subjects had been diagnosed and treated for sellar/parasellar tumours between 3 and 18 years prior to inclusion in the study. Five patients had been treated for non-functioning pituitary adenoma, two for macroadenoma and two for craniopharyngioma. The tenth patient had been treated and cured of Cushing’s disease with panhypopituitarism and was 8 years post-definitive treatment, still requiring HC replacement, with a morning pre-HC cortisol level <100 nmol/L. Nine patients had a complete anterior pituitary failure, and one patient was deficient in all anterior pituitary hormones, except gonadotrophins; all patients were on appropriate hormone replacement, including GH, without alteration in the dose for at least 3 months prior to and during the study. All 10 patients had diabetes insipidus and were on desmopressin with normal and stable plasma sodium levels before and during the study. Anterior pituitary hormone replacement therapy regimens were not adjusted during the study period, except for HC dose as per study protocol. No patient had known diabetes mellitus (DM), two patients had controlled hypertension and were on stable doses of angiotensin-converting enzyme inhibitor

www.eje-online.org
medication that was not changed during the study. This patient population has been described in detail in prior publications from our group (19, 20).

Exclusion criteria were as described in previous publications (19, 20), and with particular reference to this analysis, those with uncontrolled hypertension or a known diagnosis of DM were excluded. Female patients were excluded because of the variable effects of oestrogen status on a number of parameters that were being assessed in the overall study (21), and also on corticosteroid-binding globulin levels, thus affecting total cortisol concentrations and cortisol kinetics (22).

Healthy male controls (n=10), matched for age, BMI and waist circumference (WCM) with no known pre-existing DM or uncontrolled hypertension, were enrolled to undergo the same biochemical investigations and clinical examination as the patient group.

All the patients and controls gave written informed consent to participate in this study, which was approved by the Beaumont Hospital Medical Research (Ethics) Committee and the Irish Medicines Board.

Clinical trial registration
Irish Medicines Board Clinical Trial Number–CT900/459/1 EudraCT Number–2007-005018-37

Study design
In this prospective clinical trial, all ten patients were randomised in an open crossover protocol to each experience three HC dose regimens: dose A (20 mg 0800 h, 10 mg 1600 h), dose B (10 mg 0800 h and 1600 h) or dose C (10 mg 0800 h and 5 mg 1600 h). Patients were randomised to start on one of the three doses, and following completion of each dose regimen, they were then randomised to cross over to the next dose. In view of the short half-life of HC, the patients took each dose regimen for a full 6 weeks to allow adequate time for a “washout” of the previous dose. At the end of each 6-week treatment, schedule patients were admitted for 28 h to our clinical research centre to undergo metabolic investigations, which included a 24-h ambulatory BP measurement (24-h ABPM), an oral glucose tolerance test (OGTT) and fasting serum osteoprotegerin (OPG) measurement. During the admission, subjects took the predesignated HC dose at 0800, 1600 and 0800 h the following morning of the admission. All patients had a 24-h serum cortisol measurements on treatment as described previously (19, 20). Ten healthy-matched controls underwent identical biochemical profiling to the patient group. Patients and controls were ambulatory during the study admission days. Meal timing, activity levels and sleep duration were standardised for each of the three admission periods.

Vascular assessment
On each admission between 0730 and 0800 h, patients were fitted with validated oscillometric devices to record 24-h ABP (SpaceLabs 90202 or 90207), programmed to obtain BP readings at 30-min intervals for 24 h throughout each 26-h admission period (23). Failed recordings automatically triggered a repeat recording within a 2-min interval. All of the recorded clinical data were transferred into the dabl cardiovascular software package (dabl Ltd, Dublin, Ireland) in order to produce a report for each event.

A fasting serum sample was taken for measurement of OPG before HC was taken by the patient. Serum samples were centrifuged at 3000 rpm for 15 min and stored at −80°C for later analysis. OPG was measured using commercial enzyme-linked immunosorbent assay kits (Biomedica, Vienna, Austria) had intra- and inter-assay variations of <6%, with a minimal detection limit of 0.014 pmol/L.

Insulin sensitivity assessment
Subjects fasted from 2300 h during the admission and took the designated HC dose at 0800 h with a small sip of water as per the study protocol. About 75 g glucose was dissolved in 300 mL of water and was consumed as an oral glucose challenge 1 h later in order to standardise the effect of HC on the results. Paired serum samples for insulin and glucose were taken at time 0, before consumption of the glucose load, and at 30, 60, 90 and 120 min post oral consumption of glucose. Insulin samples were centrifuged at 3000 rpm for 15 min and stored in 1 mL aliquots at −20°C until analysis through a chemiluminescent immunoassay on the UniCel DxI 800 Access Immunoassay System (Beckman Coulter, Inc., Brea, California, USA). Glucose samples were processed immediately using the hexokinase method on an automated AU5400 analyser (Beckman Coulter, Inc.).

Data analysis
Ambulatory arterial stiffness index
Arterial stiffness varies nonlinearly with distending pressure: as mean arterial pressure increases, stiffness increases exponentially, and in this study, it was assessed using the non-invasive method of the Ambulatory
Arterial Stiffness Index (AASI) (24, 25), calculated from the half-hourly BP readings within the 24-h ABPM recordings. Using the data from the 24-h ABP machines, we computed, for each participant, the regression slope of diastolic pressure on systolic BP. We did not force the regression line through the origin (intercept 0). Therefore, the AASI is defined as follows: 1 – the regression slope of diastolic BP/systolic BP. The stiffer the arterial tree, the closer the regression slope, and the AASIs are to 0 and 1, respectively (25). The AASI is a validated predictor of vascular mortality (24, 26, 27, 28).

Glucose and insulin homeostasis

Normal glucose tolerance, impaired fasting glucose, impaired glucose tolerance and type 2 DM were diagnosed according to standard World Health Organisation criteria (29). Insulin resistance was calculated with the homeostatic model assessment of insulin resistance (HOMA-IR) using fasting insulin (IU/L) × glucose (mmol/L)/22.5 (30). Insulin measurements in ng/mL were multiplied by a factor of 175 to convert measurements to pmol, in keeping with international recommendations, for the purposes of HOMA2 calculation (31). Dynamic estimation of insulin sensitivity was measured using the oral glucose insulin sensitivity (OGIS) method (32), which has been validated against the hyperinsulinaemic–euglycaemic clamp (32).

Statistical analysis

Results are reported as mean (s.d.) or median (interquartile range) as appropriate. Between-group differences were assessed using ANOVA, or repeated measures ANOVA or the non-parametric equivalent as appropriate, followed by application of a multiple comparison test. Correlations were analysed using the Spearman’s or Pearson’s correlation coefficient, as appropriate based on normality tests. Significance was defined for P-values <0.05. Statistical analysis was performed using GraphPad Prism Windows version 5.0 (GraphPad Software).

Results

The baseline characteristics of patients and controls are shown in Table 1. Subjects were age, sex, BMI and WCM matched with controls. There were no differences in measured pituitary hormones, excluding cortisol, between patients and controls, suggesting adequate pituitary hormone replacement (Table 1).

Table 1 Patient and control baseline anthropometric and hormone status. Results are expressed as mean ± s.d.

<table>
<thead>
<tr>
<th></th>
<th>Patients (n = 10)</th>
<th>Controls (n = 10)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>46 ± 15</td>
<td>45 ± 15</td>
<td>0.9</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>29.8 ± 5.3</td>
<td>29.1 ± 4.6</td>
<td>0.5</td>
</tr>
<tr>
<td>WCM (cm)</td>
<td>105 ± 14</td>
<td>103 ± 11</td>
<td>0.53</td>
</tr>
<tr>
<td>Basal cortisol (nmol/L)</td>
<td>76.8 ± 6.5</td>
<td>403.3 ± 122.4</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Free T4 (pmol/L)</td>
<td>11.3 ± 2.1</td>
<td>10.9 ± 1.0</td>
<td>0.6</td>
</tr>
<tr>
<td>IGF-I (µg/L)</td>
<td>163 ± 45</td>
<td>152 ± 32</td>
<td>0.5</td>
</tr>
<tr>
<td>Testosterone (pmol/L)</td>
<td>14.2 ± 4.1</td>
<td>16.4 ± 7.7</td>
<td>0.4</td>
</tr>
<tr>
<td>Creatinine (µmol/L)</td>
<td>100 ± 5</td>
<td>77 ± 3</td>
<td>0.001</td>
</tr>
<tr>
<td>eGFR (mL/min/1.73 m²)</td>
<td>73 ± 5</td>
<td>97.6 ± 5</td>
<td>0.004</td>
</tr>
</tbody>
</table>

BMI, body mass index; WCM, waist circumference.

BP and OPG analysis

There were no differences in 24-h systolic BP or diastolic BP, between dose regimens and controls (Table 2). There was no correlation between mean 24-h cortisol levels, or mean nocturnal cortisol levels for any dose regimen, nor for the whole group combined (data not shown).

A nocturnal systolic dip <10% is considered abnormal (13, 33), and this was evident in three subjects on dose A, five on dose B, two on dose C and one control subject; no patient on dose C and no controls had a nocturnal diastolic dip <10%, whereas one patient on dose A and three on dose B did have an abnormal nocturnal diastolic dip. Overall the physiologic nocturnal dip in systolic and diastolic BP was blunted in subjects on higher dose HC replacement; however, this was significant only for dose B (10/10 mg) compared with dose C (10/5 mg) (Fig. 1). There was no difference in 24-h or nocturnal cortisol levels between those who had a nocturnal dip, and those who did not; however, the numbers may have been too small to show an association (data not shown).

The AASI was calculated as described previously. The closer the AASI is to 1, the stiffer the arterial tree. Low-dose HC replacement demonstrated the lowest AASI compared with the other two dose regimens (Fig. 2). There was no correlation between the AASI and the mean 24-h cortisol levels for each dose, or for the group as a whole (data not shown).

There was no difference between dose regimens and controls in OPG. There was no correlation between mean 24-h cortisol level and OPG in any dose regimen or for the controls. For the patient group, regardless of dose,
Clinical Study
L-A Behan and others
HC regimens and vascular risk

Table 2  The 24-h ambulatory BP levels between dose regimens and controls. Data are expressed as mean ± s.d.

<table>
<thead>
<tr>
<th>BP (mmHg)</th>
<th>Dose A (20/10 mg)</th>
<th>Dose B (10/10 mg)</th>
<th>Dose C (10/5 mg)</th>
<th>Control</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>24-h systolic</td>
<td>115 ± 12</td>
<td>117 ± 12</td>
<td>115 ± 13</td>
<td>121 ± 10</td>
<td>0.67</td>
</tr>
<tr>
<td>24-h diastolic</td>
<td>70 ± 8</td>
<td>68 ± 8</td>
<td>68 ± 7</td>
<td>73 ± 8</td>
<td>0.60</td>
</tr>
<tr>
<td>Daytime systolic</td>
<td>119 ± 10</td>
<td>120 ± 12</td>
<td>122 ± 13</td>
<td>128 ± 9</td>
<td>0.38</td>
</tr>
<tr>
<td>Daytime diastolic</td>
<td>74 ± 7</td>
<td>71 ± 8</td>
<td>74 ± 8</td>
<td>79 ± 8</td>
<td>0.16</td>
</tr>
<tr>
<td>Night-time systolic</td>
<td>105 ± 16</td>
<td>109 ± 16</td>
<td>103 ± 13</td>
<td>110 ± 13</td>
<td>0.70</td>
</tr>
<tr>
<td>Night-time diastolic</td>
<td>60 ± 10</td>
<td>62 ± 12</td>
<td>57 ± 7</td>
<td>63 ± 10</td>
<td>0.54</td>
</tr>
</tbody>
</table>

BP is expressed in mmHg.

OGP correlated positively with 24-h systolic and diastolic BP, and also with daytime and night-time BP parameters (Table 3). However, when analysed by dose regimen, only the highest dose of HC, dose A demonstrated a strong significant correlation with OPG for daytime SBP, $r=0.63$ (CI), $P=0.04$ and daytime DBP, $r=0.7$ (CI), $P=0.02$, and 24-h systolic BP only $r=0.65$ (CI), $P=0.04$. There were no significant correlations between OPG and any BP parameter for the lower dose regimens or for controls (data not shown). There were no significant correlations between the AASI and OPG for either group as a whole. When analysed by dose, there were moderate to strong correlations between the AASI and OPG; however, they were not statistically significant (Fig. 3).

Glucose homeostasis

Three patients and two controls had abnormal glucose levels at some point during the study, which was unrelated to HC dose regimen for the patient group (data not shown). There were no differences found between dose regimens and controls in any other glucose or insulin parameters, including fasting levels, post glucose load levels or area under the curve, insulin sensitivity (OGIS) or insulin resistance (HOMA-IR). There was no correlation between HOMA-IR and 24-h or nocturnal serum cortisol concentrations and the BP or metabolic

Discussion

In this randomised, controlled, crossover study of panhypopituitary subjects on complete pituitary hormone replacement, we have demonstrated that lower dose HC replacement is associated with a lower AASI and possibly a more physiological nocturnal BP profile. Although no difference in insulin and glucose metabolism between these commonly prescribed dose regimens was shown, 30% of the cohort had abnormal glucose metabolism at some point in the study compared with 20% matched controls, in the setting of normal fasting plasma glucose, indicating that fasting plasma glucose (FPG) would be an inadequate screening tool for this patient group. We could not demonstrate a relationship between 24-h or nocturnal serum cortisol concentrations and the BP or metabolic

Figure 1  Nocturnal systolic and diastolic BP dip for patients and controls. BP, blood pressure; SBP, systolic BP; DBP, diastolic BP; NS, not significant; *ANOVA outcome.
parameters assessed, but this may be due to the relatively small numbers of subjects. It is also possible that alterations result from chronic exposure to excessive GC replacement rather than from acute tissue exposure to GCs.

Although most studies suggest that the increased morbidity and mortality in hypopituitarism is primarily vascular in origin, the cause for the adverse vascular outcome is not clear. Two recent studies have demonstrated that inappropriate GC replacement is an independent predictor of mortality, one of which Sherlock and coworkers demonstrated that daily doses of HC of greater than or equal to 25 mg per day were associated with increased mortality independent of age, sex, calendar period and radiotherapy, and over 30% of deaths were from cardiovascular causes (8). The majority of studies evaluating cardiometabolic risks in hypopituitarism have focussed on the effects of GH deficiency and, in some cases, on GH replacement rather than on the effects of GC replacement itself. In one study assessing the impact of GC replacement, Filipsson and coworkers demonstrated an adverse metabolic profile, including dylipidaemia, an elevated HbA1c and WCM in GHD subjects treated for ACTH deficiency compared with GHD and ACTH-sufficient patients. Interestingly, ACTH-deficient patients on HC-equivalent doses <20mg daily did not differ in metabolic outcomes compared with their ACTH sufficient counterparts, whereas the adverse metabolic profile was associated with HC-equivalent doses >20mg and was more pronounced in those on doses >25mg daily (34).

It is known that 24-h ABM has more predictive power compared with random office BP readings (23), yet we demonstrated no difference in 24-h ABPM between dose regimens and controls in our group of GH-replaced hypopituitary subjects. This is consistent with data from the only other prospective study in hypopituitary subjects to use 24-h ABP, in which 13 hypopituitary GHD subjects underwent HC dose reduction from 30 to 15 mg daily and repeated 24-h ABPM 3 months later and demonstrated no difference; however, they did note that the patient cohort had lower BP, regardless of dose compared with controls (12).

Blunting of the nocturnal physiological dip in BP is a predictor of increased cardiovascular risk, independent of absolute 24-h BP (13), and is associated with increased arterial stiffness (35). Loss of the nocturnal systolic and/or diastolic BP dip and has been identified in Cushing’s syndrome (36) and with the use of exogenous high-dose GCs prescribed for anti-inflammatory purposes (37). There is little data regarding the effects of currently prescribed GC replacement regimens on the circadian rhythm of BP. We have demonstrated a blunted nocturnal dip in subjects on the higher dose replacement regimens, although this was only statistically significant for dose B compared with dose C and control subjects. Such alteration in circadian BP may reflect the abnormal cortisol dynamics in subjects on exogenous GC replacement with inappropriate nocturnal exposure, which results from higher afternoon doses (in this case 10 vs 5 mg) and possibly from increased nocturnal sensitivity to GCs. We were unable to demonstrate a difference in mean night-time cortisol levels between our subjects who had a dip compared with those who did not; however, it is possible that this is a type II error. In 64 subjects with primary adrenal insufficiency, a switch to a dual-release GC preparation resulted in a BP reduction, although this was not based on 24-h ABP measurement, and therefore, no information regarding the nocturnal dip is available (38). The only other study to use 24-h BP to evaluate the effect of different dose regimens in the

Table 3 Correlation of OPG with 24-h ambulatory BP parameters in the group as a whole.

<table>
<thead>
<tr>
<th>Ambulatory BP</th>
<th>Correlation with OPG (CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>24-h systolic BP</td>
<td>$r = 0.49 (0.16–0.72)$</td>
<td>0.006</td>
</tr>
<tr>
<td>24-h diastolic BP</td>
<td>$r = 0.45 (0.11–0.70)$</td>
<td>0.01</td>
</tr>
<tr>
<td>Daytime systolic BP</td>
<td>$r = 0.39 (0.03–0.66)$</td>
<td>0.03</td>
</tr>
<tr>
<td>Daytime diastolic BP</td>
<td>$r = 0.41 (0.07–0.68)$</td>
<td>0.02</td>
</tr>
<tr>
<td>Night-time systolic BP</td>
<td>$r = 0.54 (0.22–0.75)$</td>
<td>0.002</td>
</tr>
<tr>
<td>Night-time diastolic BP</td>
<td>$r = 0.43 (0.09–0.69)$</td>
<td>0.01</td>
</tr>
</tbody>
</table>

$r$, Pearson’s correlation; CI, confidence interval; BP, blood pressure.
Hypopituitary patient population did not report whether there was any alteration in circadian BP rhythms (12). In view of these results and the mixed evidence regarding the contribution of hypertension to the morbidity and mortality in hypopituitary subjects, it is likely that loss of circadian variation in BP is an important marker of vascular risk in this patient group.

Vascular disease is associated with increasing stiffness of the arterial tree, and a number of studies in hypopituitary patients have demonstrated reduced large vessel reactivity (39) or reduced endothelium-derived dilation as a reflection of abnormal vascular function (40, 41). Arterial stiffness as measured by pulse wave velocity is a predictor of cardiovascular events, independent of pulse pressure (42, 43). We have demonstrated that low-dose HC replacement is associated with reduced AASI. The AASI has been shown to identify arterial dysfunction at a younger age compared with pulse pressure (24). In a study of 11,291 adults not on antihypertensive medication at the time of ABPM recording, the AASI has been shown to predict death from stroke and was found in normotensive subjects to be more predictive of stroke and cardiovascular mortality than pulse pressure (28). Our findings regarding the AASI are particularly relevant in hypopituitarism in which cerebrovascular mortality is increased anywhere from a standardised mortality rate (SMR) of 1.7–4.9 (1) and notably in those diagnosed at a young age (44).

Reduced AASI in our cohort on low-dose HC replacement is unlikely to represent under-replacement with GC, as no patient reported postural symptoms, a finding that is also consistent with reported data from Dunne and coworkers who reduced HC from 30 to 15 mg daily without any onset of postural symptoms. The same group also demonstrated higher forearm blood flow on the lower dose HC regimen (15 mg daily), compared with the higher dose, indicative of improved vascular reactivity (12).

Our findings with respect to the nocturnal dip and the AASI suggest controlled HC dose reduction may be beneficial for long-term health of the vascular tree. Although our control group had better renal function compared with patients, based on the estimated glomerular filtration rate (eGFR) and serum creatinine, only one patient had an eGFR < 60 mL/min/1.73 m², and there was no difference in renal function between the three HC dose regimens across the duration of the study; therefore, differences in the AASI between dose regimens is unlikely to be explained by renal function.

In our cohort of panhypopituitary patients, fully replaced on GH, we were unable to demonstrate any difference in markers of insulin sensitivity or resistance between dose regimens. This is consistent with findings of other groups that assessed glucose metabolism using fasting levels only (12, 17). OPG, a member of the TNF receptor subfamily, is present in a number of tissues, including the bone, the immune system and vasculature (45, 46) and in humans levels are positively correlated with vascular morbidity and mortality (45, 46). As OPG is an emerging novel vascular marker and was never studied before in hypopituitary patients, we were interested in examining its role in this cohort. We found a positive correlation between OPG and 24-h BP parameters, which became a significant strong positive correlation when analysed for the highest dose replacement regimen, dose A. This may reflect a higher cardiovascular risk category for the highest dose group.

There are a few limitations to this study; it is conceivable that lack of difference in 24-h ABP parameters between doses and compared with controls is a type II statistical error due to the small number of subjects; however, previous studies with similar small numbers
demonstrated differences in BP in a similar patient population size and similar baseline BP readings, so it is unlikely that this is the case. The lack of association between the cardiometabolic parameters and the serum cortisol levels may be due to the small numbers. There has been debate previously regarding the use of a surrogate marker of arterial stiffness such as the AASI. The AASI may also be affected by the mechanical properties of the small vessels, which may be the reason that it is a useful predictor of stroke in the normotensive population. In fact, this may be a strength for the AASI as a test in the hypopituitary cohort, in whom hypertension does not appear to be a prominent feature. It is a quick and easy calculation that can be taken from any ABPM and may be of use in the clinical setting to identify at-risk patients.

In sum, this is the first study to prospectively evaluate three commonly prescribed HC dose regimens in panhypopituitary subjects on full pituitary hormone replacement with respect to these cardiometabolic risk factors. Currently prescribed doses of HC replacement do not result in significant differences in absolute BP levels or improvements in insulin sensitivity, yet lower doses are associated with a less stiff arterial tree and a more physiological nocturnal BP dip and therefore may confer reduced vascular risk. Physicians should aim for safe dose reductions of HC in order to reduce the adverse impact of excess replacement. Hypopituitary patients should be assessed with OGTT and 24-h ABP in order to identify those at highest metabolic risk. Further long-term studies are warranted to confirm our findings and assess the impact these findings may have on vascular morbidity and mortality in this patient population.

Declaration of interest
The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

Funding
This research was supported by an unrestricted educational grant from Pfizer Endocrine Care.

Acknowledgments
The authors thank Dermot Kenny and the staff of the RCSi Clinical Research Centre, Dublin, Ireland, where the research was conducted, and the staff of the Ambulatory Blood Pressure Unit in Beaumont Hospital, Dublin, Ireland, for their assistance during the study.

References
European Journal of Endocrinology
799


42 Pereira T, Maldonado J, Pereira I & Conde J. Aortic stiffness is an independent predictor of stroke in hypertensive patients. *Arquivos Brasileiros de Cardiologia* 2013 **100** 437–443.


Received 6 December 2015
Revised version received 20 March 2016
Accepted 29 March 2016