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Diurnal cortisol patterns are associated with physical performance in the Caerphilly Prospective Study

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Background Cross-sectional studies have suggested that elevated cortisol is associated with worse physical performance, a surrogate of ageing. We examined the relationship between repeat cortisol measures over 20 years and physical performance in later life.

Methods Middle-aged men (45–59 years) were recruited between 1979 and 1983 (Phase 1) from the Caerphilly Prospective Study (CaPS) and re-examined 20 years later at 65–83 years of age (Phase 5). Participants included 750 and 898 subjects with either Phase 1 and/or Phase 5 data on exposure and outcomes. Outcome measures were walking speed and balance time and exposures included morning fasting serum cortisol (Phase 1) and four salivary samples on 2 consecutive days (Phase 5).

Results Faster walking speed was associated with higher morning cortisol at Phase 1 [coefficient per standard deviation (SD) increase 0.68, 95% confidence interval (95% CI) 0.09–1.27; $P=0.02$] though this was attenuated after adjustment for covariates (coefficient per SD increase 0.45; 95% CI –0.16 to 1.07; $P=0.15$). Higher night-time cortisol at Phase 5 was associated with slower speed (coefficient per SD increase –1.06; 95% CI –1.60 to –0.52; $P<0.001$) and poorer balance (odds ratio of top tertile vs bottom 2.49; 95% CI 1.63–3.81; $P<0.001$). Worst performance was seen for men with a poor morning response (Phase 1) and less nocturnal decline (Phase 5).

Conclusions Dysregulation of the hypothalamic pituitary adrenal (HPA) axis is associated with worse physical performance in later life. This may reflect a causal effect of the HPA axis on ageing or that ageing itself is associated with reduced HPA reactivity.

Keywords HPA axis, physical capability, healthy ageing

Introduction

Evidence from animal¹ and human² studies suggest that the hypothalamic pituitary adrenal (HPA) axis contributes to biological ageing. The hypothalamus controls both the circadian pattern and the absolute levels of adrenocorticotrophic hormone (ACTH) secretion from the anterior pituitary, which in turn stimulates the secretion of glucocorticoid hormones (cortisol in man and corticosterone in rodents) from the adrenal cortex.³ In some studies, aged animals have been shown to have high glucocorticoid levels, which also show a delayed return to normal following a stressful event.¹ This has been postulated to lead to a positive feedback loop with reduced central inhibition ('glucocorticoid cascade' hypothesis), and thus even greater glucocorticoid responses over time. Human data testing this hypothesis are limited, though a recent report from the Whitehall II cohort found that older individuals had a flatter diurnal pattern.⁴

Sarcopenia is the loss of muscle mass that occurs with ageing.⁵ This loss of muscle mass is associated with a loss of physical performance such as the ability to rise from a chair.⁶ One potential mechanism for sarcopenia may be due to elevated life-course exposure to cortisol⁶ a potent stimulus to protein catabolism.^{7,8} For example, an excess of endogenous or exogenous corticosteroids will result in steroid myopathy.⁸ It is unclear whether a similar state of sarcopenia may result from normal physiological variations of endogenous cortisol secretion. Only five studies have reported associations between cortisol levels and physical performance tests.^{4,9–13} Three of these were based on small sample sizes of less than 60 participants (therefore underpowered to detect clinically or biologically important differences) and no study had follow-up >4 years. One study found that higher cortisol levels were associated with worse physical performance but this was inconsistent;¹² women with higher cortisol levels showed worse performance on a balance test (tandem stand), whereas men with higher levels had worse chair rises and timed walk test. The same group also demonstrated that higher cortisol was associated with a loss of grip strength over a 4-year period for men and women combined.¹¹ In the Whitehall II study, participants with higher daytime cortisol (compared with the normative group) had slower walk time tests.⁴ No study has examined whether changes in cortisol secretion are more strongly associated with physical performance. The Caerphilly Prospective Study (CaPS) has two sets of cortisol measures over a 20-year period in a large population sample of men from South Wales. We hypothesized that either elevated cortisol or lack of diurnal variability as a marker of HPA dysregulation would be associated with worse physical performance and that subjects with an increase in cortisol levels would perform worse than those whose levels simply tracked over time.

Methods

Participants

The CaPS¹⁴ was set up to examine risk factors for cardiovascular disease but as subjects became older additional age-related phenotypes were added. The baseline study (Phase 1) tried to contact all men aged 45–59 years from Caerphilly and adjacent villages. A total of 2512 men (response rate of 89% of those who were eligible) were seen between 1979 and 1983. These men have been followed up four more times, the last being Phase 5 when the men were 65–83 years old. At Phase 2, an additional 447 men were recruited who had been missed in Phase 1 ('reconstructed cohort').

Clinical and questionnaire-based data

At each phase, men completed a questionnaire and had anthropometric measures taken at an evening clinic. At Phase 2, the 30-item General Health Questionnaire, a validated measure of minor psychological morbidity, was included.¹⁵ Socio-economic status was derived using the subjects' reported occupation and the Registrar General's classification (I—professional, V—unskilled manual). Men were classified into never, past or 1–14, 15–24 and ≥25 cigarettes per day (Phase 1), or never, past or 1–12 and ≥13 cigarettes per day (Phase 5) to produce roughly equal-sized groups. Alcohol consumption (units per week) was derived from a food frequency questionnaire. At each phase, we identified subjects on corticosteroid medications. At the clinic, height was measured in metres using a stadiometer and weight in kilograms using standardized scales.

Physical performance measures

These measures were first introduced in Phase 5. The 'get up and go' test¹⁶ is a standardized objective measure of functional leg strength, power, mobility and balance integrating several basic mobility manoeuvres. The participant is timed rising from a chair, walking 3 m at their usual speed, turning, walking back and sitting down. The test was performed twice with the mean time taken for analysis. The 'flamingo' test measures the ability to maintain postural stability in the upright position¹⁷ and predicts falls in older persons.¹⁸ Participants stood on their preferred leg while lifting the other leg, keeping eyes open. The position was held for as long as possible up to a maximum of 30 s. The flamingo test was performed twice, unless the maximum score of 30 s was achieved in the first attempt. The best score was used in the analysis.

Cortisol measures

For Phase 1, participants attended an early morning clinic and had a fasting blood sample taken. Phase 1 samples for serum cortisol were frozen at –20°C,

assayed within 3 months of collection and measured by radioimmunoassay.¹⁹ The overall coefficient of variation for serum cortisol ($n=135$ pairs) was 16%.¹⁴ At Phase 5, men were shown how to collect saliva using plain cotton wool swabs (salivettes—Sarstedt) at home. Subjects were instructed to gently chew on the salivettes so that a saliva sample of 0.5–1 ml was obtained within a minute. Subjects were requested to take samples on waking, 30 min later, and at 2 pm and 10 pm on 2 consecutive days. Samples were stored in a domestic fridge until posted back to the researchers with a datasheet to indicate the actual times of sampling. Samples were frozen at -40°C and subsequently assayed by radioimmunoassay²⁰ in a laboratory specializing in high-throughput cortisol assays (Dresden LabService GmbH). The inter-assay coefficient of variation for the salivary cortisols (118 plates) was 4% at both low (5.3 nmol/l) and high (39.0 nmol/l) controls.

Statistical analysis

Cortisol has a marked circadian rhythm and therefore the time of day at which cortisol is sampled affects the cortisol level. At Phase 1, the time a man attended the morning clinic varied by 6 hours. We therefore adjusted the observed values for the time of sampling by fitting a polynomial function to the association between cortisol level and time of measurement ('fracpoly' option in the Stata software)²¹ and adding the residuals from the best fit model to the overall mean cortisol value as has been previously reported.¹⁴ The Phase 5 samples were taken at specific time points (though individuals' waking times vary) with a much narrower range of times and we found there was relatively little variability in sampling times. In addition, actual time of sampling did not improve the model fit or predict cortisol levels using either a linear or indefinite article polynomial function for time. Further analyses did not, therefore, adjust for time of sampling. Participants were requested to write any comments relating to problems with taking the samples. In five cases, subjects reported that they did not take the samples at the specified time points.

As well as the morning (mean of waking and 30 min sample) and night-time values, we derived three additional measures commonly used in epidemiological studies.²² These were 'Cortisol Awakening Response' (CAR) (difference between the 30 min post waking sample and the waking sample), diurnal drop (difference between morning and evening samples) and area under the curve (AUC) for all measures. We used the average of the measures across 2 days unless they only returned measures for 1 day.

The distribution of the get up and go test is positively skewed, so we converted it into walking speed (metres per minute), as detailed in a large recent meta-analysis examining physical capability and mortality.²³ This normalized the data to a large degree. We used the average of two attempts. Balance time

from the flamingo test was dichotomized at the 20th centile (around 3.75 s) as it was highly skewed and those in this group were classified as 'poor balance' compared with the rest of the sample. This is the same analytical approach as adopted in a recent study examining socio-economic disadvantage from childhood to adulthood and locomotor function in old age.²⁴

We used linear regression models to analyse walking speed and logistic regression analysis for balance time. Serum cortisol levels represent total protein bound and free cortisol concentrations in the blood, which are around 20 times higher than the free cortisol concentrations found in saliva. We therefore converted the absolute levels to phase-specific z-scores by calculating the difference between the observed value from the mean and dividing by the standard deviation. We modelled cortisol both as a continuous variable and in three equal groups (tertiles) to examine for departures from linearity. As night-time cortisol and the AUC for cortisol were positively skewed, we transformed them (\log_e) before converting them to z-scores. We chose potential confounders for the multivariable models from the previous literature and examined associations with age,²⁵ adiposity,^{25,26} socio-economic status,²⁷ smoking status,²⁸ alcohol consumption²⁹ and psychological morbidity.³⁰ As we failed to find any association between socio-economic status, alcohol and psychological morbidity with cortisol, our final multivariable model only adjusted for age, body mass index (BMI) and smoking status measured at the same phase as the cortisol assay. To examine for the long-term effects of different cortisol patterns, we undertook *post hoc* analyses and dichotomized cortisol into the top tertile vs low and middle groups at Phases 1 and 5 and created four subgroups (low-medium Phases 1 and 5, low-medium Phase 1 and high Phase 5, high Phase 1 and low-medium Phase 5, high Phases 1 and 5). We formally tested for an interaction effect between Phases 1 and 5 cortisol levels.

We undertook a series of further sensitivity analyses. As we did not have the actual time of waking for Phase 1 cortisol, it was possible that older subjects had artefactually lower morning cortisol levels by virtue of having woken earlier and, therefore, had a greater diurnal drop by the time they had arrived at clinic. We therefore examined whether the cortisol performance association differed by whether they attended clinic early or later on the basis that this bias would be less marked in subjects attending clinic later in the morning so that any difference in waking was attenuated. We also used the time of waking reported at Phase 5 as a proxy for Phase 1 and tested whether adjustment made any difference.

Some subjects were unable to perform the tests because of surgery, injury, other health problems or unwillingness and had missing data. We therefore repeated the analyses by classifying these subjects as

in the bottom 20% of the distribution. As there is no standard way to analyse diurnal drop, we carried out a sensitivity analysis using first waking cortisol and then 30 min post waking cortisol as the anchor for the diurnal drop. To maximize the precision of results, the analysis of associations between Phase 5 cortisol and physical performance included some men who did not have cortisol measured at Phase 1. We have additionally repeated analyses presented in Tables 2 and 3 including only men at Phase 5 who also had Phase 1 cortisol measures.

Ethical approval

Ethical approval was given for the CaPS by the Ethics Committee of the Division of Medicine of the former South Glamorgan Area Health Authority. Approval for Phase 5 came from the South East Wales Research Ethics Committee and for the cortisol assays from the North West Wales Research Ethics Committee.

Results

At Phase 1, 2143 men had a blood sample taken between 6 and 12 am (85% of eligible men). At Phase 5, 7837 salivary cortisol samples were collected from 1034 participants (response rate of 86.4% with mean number of samples per man of 7.6). The median time lag and inter-quartile range between Phases 1 and 5 were 21.8 (20.9–22.6) years. A total of 61 samples from 30 subjects were censored as the saliva values were thought to be artefactually high (>100 nmol/l). This problem has been observed in other large-scale epidemiological studies.²² A further 11 participants on oral corticosteroid medication were excluded. Of the remaining 962 men, 898 had walking speeds and 911 had balance times. Of the participants at Phase 1 with serum cortisol samples, 750 had walking speeds and 756 had balance times. The number of men who had cortisol measures at both Phases 1 and 5 was 735.

Table 1 shows the basic descriptive data for the exposures, confounders and outcomes. Men at Phase 5 were ~20 years older, had higher BMI and were far more likely to be never or ex-smokers. The time-adjusted serum cortisol at Phase 1 was 443.7 nmol/l, whereas the peak morning saliva level at Phase 5 was 20.7 nmol/l (assuming a 20-fold conversion factor this is equivalent to 414 nmol/l). Whereas morning cortisol decreased with age for men in Phase 1 (β per year age = -1.37; 95% CI -2.59 to -0.14; $P=0.03$), it increased with age for men in Phase 5 (β per year age = 0.25; 95% CI 0.08–0.42; $P=0.004$). Current smoking was associated with higher morning cortisol at both Phases 1 and 5 (P -value for linear trend < 0.001 and $P=0.02$, respectively).

Table 2 shows the relationship between cortisol measures in Phases 1 and 5 with walking speed.

Higher morning cortisol in Phase 1 was associated with a faster speed ($P=0.02$) though this was attenuated after adjustment for the other covariates ($P=0.15$). There was little evidence of an association between morning salivary cortisol at Phase 5 and speed. Night-time cortisol levels showed a strong inverse association so that higher levels were associated with slower speed and hence worse performance. This effect looked nonlinear as it was more marked in the top third. Similarly, subjects with the largest diurnal drop (reflecting higher morning and lower night-time levels) had greater speed and hence better performance but here the association was most marked in the middle tertile. There was no strong association between the CAR and walking speed though there was weak evidence ($P=0.07$) in the fully adjusted model that the top third of CAR had faster velocity. There was no association between the AUC and speed (data not shown).

There was no association between the Phase 1 morning cortisol and balance time (Table 3). Higher morning cortisol at Phase 5 was associated with worse balance ($P=0.006$), but after adjustment for confounding this association was markedly attenuated ($P=0.17$). As with speed, higher night-time cortisol levels were associated with worse balance but again this effect was driven by the association in the top tertile. This association persisted, albeit slightly weaker, in the fully adjusted model. Neither the diurnal drop nor CAR was associated with worse balance. Whereas greater AUC was weakly associated with worse balance ($P=0.05$), this was attenuated after adjustment ($P=0.18$) (data not shown).

There were only weak associations between the cortisol measures at Phases 1 and 5 (range between 0.04 and 0.12 Spearman's rank correlations). When we examined cortisol patterns over time, we found that the slowest walk speed was seen in subjects with low-medium levels of cortisol at Phase 1 and high night-time levels at Phase 5 (-3.13; 95% CI -4.64 to -1.63; $P<0.001$) as compared with those with high levels at both phases (-1.36; 95% CI -3.42 to 0.70; $P=0.19$) though these differences were attenuated in the final model (-2.12 vs -1.76) (Supplementary Table S1). The same patterns were seen with worse balance (Supplementary Table S2); a formal test for interaction, however, found the variations between groups to be consistent with chance ($P=0.51$ for walking speed and $P=0.39$ for balance in age-adjusted models).

Sensitivity analyses

There was no difference in the age of participants whether they attended the clinic early or late at Phase 1, a surrogate for whether they woke up early or late (data not shown). There was little effect on the associations between Phase 1 cortisol and either walking speed or balance time, stratified by whether they attended the clinic early or late and tests for

Table 1 Summary of demographic characteristics for Phases 1 and 5 of the Caerphilly cohort study

Variable	<i>n</i> (%)	Phase 1, 1979–83	<i>n</i> (%)	Phase 5, 2002–04
Age (years)	2486	52.68 (4.55)	1143	73.40 (4.17)
BMI (kg/m ²)	2470	26.16 (3.63)	1130	27.88 (4.30)
Serum cortisol (nmol/l) ^a	2143	443.68 (132.26)	–	–
Salivary cortisol (nmol/l) ^b				
On waking	–	–	982	18.74 (10.99)
30 min post waking	–	–	993	20.68 (12.16)
Afternoon (2 pm)	–	–	995	8.24 (7.55)
Evening (10 pm)	–	–	998	4.17 (7.46)
Get up and go (m/min)	–	–	1061	34.67 ^c (8.08)
	–	–	–	35.00 ^d
Flamingo tests (s)	–	–	1070	16.71 ^e (11.67)
	–	–	–	15.66 ^f
Smoking status				
Never	392 (17.5)		322 (30.3)	
Ex-smoker	722 (32.3)		592 (55.6)	
Current smoker	1124 (50.2)		150 (14.1)	

Results are presented as mean (SD), unless otherwise stated.

^aSerum cortisol level is adjusted for the time of sampling.

^bSalivary cortisol samples are mean of samples on consecutive days and are unadjusted for the time of sampling.

^cMean and get up and go speed from two attempts.

^dMedian get up and go speed from two attempts.

^eHighest flamingo time from two attempts.

^fMedian flamingo time.

interactions found variations to be consistent with chance (data not shown). Adjustment for waking time at Phase 5 made little difference to the results (data not shown). Including subjects who could not complete the tests made little difference to the results. We found little difference in the association between diurnal drop and walk speed when using waking cortisol ($\beta=0.37$; 95% CI: -0.14 to 0.87 ; $P=0.15$; age adjusted) or 30 min post waking ($\beta=0.43$; 95% CI -0.07 to 0.93 ; $P=0.10$; age adjusted) as the anchor point for the diurnal drop. As before, there was no evidence of an association between diurnal drop and balance. When we repeated our analysis including only men with cortisol measures at both Phases 1 and 5, the associations with walk speed, if anything, slightly increased for most of the exposures except the morning cortisol measure [Phase 5 morning cortisol ($\beta=0.31$; 95% CI -0.29 to 0.90 ; $P=0.32$), night-time cortisol (\log_e) ($\beta=-1.08$; 95% CI -1.69 to -0.48 ; $P<0.001$), diurnal drop ($\beta=0.69$; 95% CI 0.11 – 1.28 ; $P=0.02$) and CAR ($\beta=0.23$; 95% CI -0.36 to 0.83 ; $P=0.44$). Similarly, it made little difference to the associations between Phase 5 cortisol measures and balance (data not shown), although the association between higher night-time cortisol (\log_e) and poorer balance was now slightly weaker (OR = 1.17; 95% CI 0.94–1.45; $P=0.16$; age adjusted).

Discussion

The results of this study showed that higher cortisol levels in middle-aged men were associated with faster walking speed in later life, whereas higher night-time cortisol and a smaller diurnal drop, measured in later life, were associated with slower speed. In contrast, we found no effect of mid-life cortisol levels on later life balance time, but higher morning cortisol and higher night-time cortisol, measured contemporaneously, were associated with worse balance. Across the 20 years, the most adverse pattern associated for both speed and balance was seen in men with low–medium cortisol in mid-life and high night-time levels in later life, though this may have occurred by chance and we were not well powered to detect modest interactions.

There is increasing evidence that the pattern of tissue exposure to glucocorticoids is a very important determinant of glucocorticoid signalling and gene activation,³¹ although in the current study we were unable to assess ultradian rhythmicity. We had predicted that either elevated cortisol or lack of diurnal variability, as markers of HPA dysregulation, would be associated with worse physical performance and that subjects with increasing cortisol levels would perform worse than those whose levels simply tracked over time. In fact, our best predictor was higher

Table 2 Relationship between cortisol measures and get up and go speed in Phases 1 and 5

	Unadjusted			Age adjusted			Age, BMI and smoking status adjusted		
	Get up and go (m/min)			Get up and go (m/min)			Get up and go (m/min)		
	β	95% CI	<i>P</i> -value	β	95% CI	<i>P</i> -value	β	95% CI	<i>P</i> -value
Phase 1, <i>n</i> = 750									
Cortisol ^a	0.68	0.09 to 1.27	0.02	0.54	-0.03 to 1.11	0.07	0.45	-0.16 to 1.07	0.15
Tertile 1	-	-	-	-	-	-	-	-	-
Tertile 2	0.97	-0.41 to 2.34	0.17	1.01	-0.32 to 2.34	0.14	0.81	-0.57 to 2.19	0.25
Tertile 3	1.51	0.11 to 2.91	0.04	1.19	-0.16 to 2.54	0.09	1.10	-0.33 to 2.53	0.13
Phase 5, <i>n</i> = 898									
Morning cortisol ^b	-0.08	-0.61 to 0.45	0.76	0.11	-0.40 to 0.62	0.67	-0.12	-0.64 to 0.40	0.65
Tertile 1	-	-	-	-	-	-	-	-	-
Tertile 2	1.01	-0.25 to 2.27	0.12	1.27	0.06 to 2.49	0.04	0.89	-0.35 to 2.13	0.16
Tertile 3	0.04	-1.23 to 1.30	0.95	0.66	-0.56 to 1.89	0.29	0.28	-0.96 to 1.51	0.66
Night-time cortisol ^c	-1.28	-1.80 to -0.76	<0.001	-0.96	-1.46 to -0.45	<0.001	-1.06	-1.60 to -0.52	<0.001
Tertile 1	-	-	-	-	-	-	-	-	-
Tertile 2	-0.87	-2.11 to 0.37	0.17	-0.21	-1.41 to 1.00	0.74	-0.37	-1.59 to 0.84	0.55
Tertile 3	-3.48	-4.71 to -2.24	<0.001	-2.76	-3.96 to -1.55	<0.001	-2.48	-3.73 to -1.22	<0.001
Diurnal drop ^d	0.33	-0.20 to 0.85	0.22	0.40	-0.11 to 0.91	0.12	0.33	-0.20 to 0.86	0.22
Tertile 1	-	-	-	-	-	-	-	-	-
Tertile 2	1.95	0.68 to 3.21	0.003	2.02	0.81 to 3.24	0.001	1.66	0.42 to 2.90	0.01
Tertile 3	1.08	-0.18 to 2.34	0.09	1.50	0.28 to 2.72	0.02	1.15	-0.09 to 2.38	0.07
CAR ^e	0.20	-0.33 to 0.74	0.46	0.27	-0.25 to 0.78	0.31	0.34	-0.19 to 0.87	0.21
Tertile 1	-	-	-	-	-	-	-	-	-
Tertile 2	0.39	-0.87 to 1.66	0.54	0.44	-0.78 to 1.65	0.48	0.70	-0.52 to 1.93	0.26
Tertile 3	0.98	-0.29 to 2.25	0.13	1.02	-0.20 to 2.24	0.10	1.15	-0.08 to 2.39	0.07

n = number of observations.

All cortisol measures have been z-scored.

^aTime-adjusted serum cortisol.

^bMorning salivary cortisol is the average of the mean waking and 30 min post waking samples on consecutive days.

^cMean night-time cortisol over 2 consecutive days.

^dDiurnal drop is the difference between morning and night-time salivary cortisol over 2 consecutive days.

^eCAR is the difference between the 30 min post waking sample and the waking sample over 2 consecutive days.

night-time cortisol, which in turn contributed to less diurnal variability, and correlates in other studies with increased psychosocial stress.³² Our findings that higher mid-life morning cortisol was associated with better performance, and that there was little evidence of an association between morning salivary cortisol and physical performance in elderly men was counter to our a priori hypothesis; however, this may reflect an over-simplistic interpretation of the physiological significance of high cortisol levels as we shall discuss further.

We believe that a less responsive HPA axis, rather than absolute cortisol levels per se, is more important in determining physical performance. In our study, cortisol levels decreased with age for middle-aged

men but increased with age for elderly men. The latter observation is consistent with a Swedish study,²⁵ where higher morning cortisol was found in older men and in the Whitehall II study where participants with higher daytime cortisol (compared with the normative group) were older.⁴ Our mid-life cortisol samples were taken after an overnight fast and by venepuncture, both physiological stressors. It is possible that individuals with more dynamic (greater diurnal drop and lower night-time measures), hence a healthier HPA axis, produced a greater stress-related response and hence 'biologically' younger men had higher morning levels. We therefore believe that a heightened stress-induced morning response in a normal population may be indicative of a healthier

Table 3 Relationship between cortisol measures and flamingo in Phases 1 and 5

	Unadjusted		Age adjusted		Age, BMI and smoking status adjusted	
	Low flamingo (bottom 20%)		Low flamingo (bottom 20%)		Low flamingo (bottom 20%)	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Phase 1, <i>n</i> = 756						
Cortisol ^a	1.00 (0.83–1.20)	0.98	1.06 (0.87–1.28)	0.58	1.06 (0.86–1.32)	0.58
Tertile 1	1		1		1	
Tertile 2	1.13 (0.74–1.73)	0.58	1.14 (0.73–1.77)	0.56	1.10 (0.68–1.78)	0.71
Tertile 3	0.94 (0.60–1.46)	0.78	1.08 (0.68–1.71)	0.74	1.08 (0.65–1.81)	0.76
Phase 5, <i>n</i> = 911						
Morning cortisol ^b	1.24 (1.06–1.44)	0.006	1.20 (1.03–1.41)	0.02	1.13 (0.95–1.35)	0.17
Tertile 1	1		1		1	
Tertile 2	0.71 (0.46–1.10)	0.13	0.64 (0.41–1.01)	0.06	0.81 (0.50–1.31)	0.39
Tertile 3	1.33 (0.89–1.98)	0.17	1.15 (0.76–1.75)	0.50	1.12 (0.71–1.76)	0.64
Night-time cortisol ^c	1.33 (1.12–1.58)	0.001	1.23 (1.03–1.47)	0.02	1.27 (1.03–1.56)	0.02
Tertile 1	1		1		1	
Tertile 2	1.23 (0.78–1.96)	0.37	1.01 (0.63–1.63)	0.96	1.13 (0.68–1.89)	0.63
Tertile 3	2.49 (1.63–3.81)	<0.001	2.09 (1.35–3.24)	0.001	2.30 (1.41–3.74)	0.001
Diurnal slope ^d	1.17 (0.98–1.40)	0.07	1.18 (0.99–1.42)	0.06	1.08 (0.88–1.31)	0.47
Tertile 1	1		1		1	
Tertile 2	0.83 (0.54–1.27)	0.39	0.82 (0.52–1.27)	0.37	0.88 (0.55–1.42)	0.61
Tertile 3	1.17 (0.78–1.76)	0.45	1.08 (0.71–1.65)	0.71	0.96 (0.60–1.51)	0.86
CAR ^e	1.05 (0.88–1.26)	0.59	1.04 (0.87–1.25)	0.67	1.02 (0.84–1.25)	0.82
Tertile 1	1		1		1	
Tertile 2	0.81 (0.52–1.26)	0.34	0.79 (0.50–1.25)	0.32	0.76 (0.47–1.24)	0.27
Tertile 3	1.26 (0.83–1.90)	0.28	1.29 (0.84–1.97)	0.24	1.24 (0.78–1.95)	0.36

All cortisol measures have been z-scored.

n = number of observations

^aTime-adjusted serum cortisol.

^bMorning salivary cortisol is the average of the mean waking and 30 min post waking samples on consecutive days.

^cMean night-time cortisol over 2 consecutive days.

^dDiurnal drop is the difference between morning and nighttime salivary cortisol over 2 consecutive days.

^eCAR is the difference between the 30 min post waking sample and the waking sample over 2 consecutive days.

HPA axis, though this interpretation may not be shared by all. This is supported by the modest positive correlation between Phase 1 morning cortisol and diurnal variation at Phase 5 so that men with higher morning levels showed greater diurnal variation 20 years later. A similar argument to this has been made in relation to the CAR, which is larger in healthy individuals³³ and may reflect an adaptive 'boost' response³² where the body anticipates having to cope with stressful stimuli—the ability to mount such a response being a marker of a healthier HPA axis. Our data therefore suggest that individuals with the least responsive HPA axis have worse performance measures in later life, although we would have expected CAR to be more strongly associated with physical performance.

There are seven relevant publications from six independent studies (Supplementary Table S3). Three of these studies had less than 100 participants. The best-powered studies came from the Longitudinal Aging Study Amsterdam (LASA)^{11,12} and the Whitehall II study.⁴ The LASA study found that subjects in the top fourth (or above the top quartile) of morning and evening cortisol levels had greater loss of grip strength¹¹ and that total cortisol was associated with worse total performance score for women whereas high evening cortisol was associated with worse total performance for men and women.¹² They had no measure to derive the CAR and did not specifically test for the diurnal drop. In the Whitehall II study,⁴ individuals with slower walk time were more likely to show a smaller diurnal drop, consistent with our

findings. This was based on a large sample of subjects but was cross-sectional in nature. Our findings are also consistent with the clinical concept of clinical frailty. In a recent study of 214 older women,³⁴ a positive association was found between evening measures of cortisol and increasing levels of frailty.

The main theories of ageing can be classified as evolutionary, molecular, cellular and system based, and there may be interaction at these levels.³⁵ The neuroendocrine aspects of the system-based theory derive from studies that show that exposure to high levels of corticosteroids results in memory loss and hippocampal shrinkage, which in animals is seen to be associated with dendritic pruning. Since chronic stress results in impaired glucocorticoid feedback and enhanced levels of glucocorticoids, the glucocorticoid cascade hypothesis suggests that the allostatic load resulting from overactivity of the HPA axis weakens the ability to adapt to changes in the environment and is an important contributory cause of ageing in the CNS. This hypothesis is consistent with our observations that higher night-time levels and to a lesser degree smaller diurnal variability, were associated with worse performance.

Other studies suggest that socio-economic status,²⁷ psychosocial stressors³² and early life factors³⁶ may be important in determining the function of the HPA axis. These factors may all contribute to life-course influences on ageing and socio-economic differences in physical performance.²⁴

Strengths and limitations

This study is based on approximately 1000 men who have been followed up for 20 years. Whereas we only had a single serum measure of cortisol at baseline, this was not a simple waking sample but reflected a response to the stressful stimuli of fasting and venepuncture. At Phase 5, we had four measures of salivary cortisol over 2 consecutive days that allowed us to derive several functional measures. We used standardized tests of physical performance that are known to predict future disability,³⁷ hospital admission³⁸ and mortality.²³ The long time period between the Phase 1 measures and the Phase 5 performance tests makes it unlikely that any associations are due to reverse causation. The consistency of the pattern that a less robust circadian regulation of cortisol was associated with worse performance across this time period provides further evidence that this association may be causal. In our final models, we adjusted for BMI as a potential confounder though this may over-adjust (and hence underestimate) the true associations as it has been postulated that adiposity may be secondary to increased cortisol exposure and may be an intermediary between cortisol and physical performance.³⁹ Our findings may not be generalizable to women though the studies cited above suggest similar effects for women. By definition, our observations are based on the healthy survivors of the

cohort who were able to take part in Phase 5. We have previously demonstrated that subjects with elevated cortisol and reduced testosterone levels had a higher risk of cardiovascular disease possibly mediated through insulin resistance.¹⁴ Thus subjects with the most dysfunctional HPA axis are under-represented in the final sample and this selective loss would attenuate the true associations. Unfortunately, participants at Phase 1 were not asked what time they woke up. Hence, it is possible that the decrease in cortisol with age at Phase 1 might be explained by older men waking up earlier. We have tried to examine this hypothesis and though we have only done this indirectly, we failed to find any evidence to suggest that this is the explanation. Finally, it is important to consider the degree of measurement error in trying to characterize the HPA axis. A random measure of cortisol is a poor measure of the HPA axis and though several measures across the day are better, they are still less than ideal. More dynamic tests, such as psychosocial⁴⁰ or physiological stressors,⁴¹ may provide more valid measures but are costly and difficult to include in large population-based studies. Our observation that the Phase 5 measures were more predictive than the earlier Phase 1 measure may simply reflect the different sampling procedures, better assays methods with a smaller coefficient of variation and that data were collected over 2 days at Phase 5, hence reducing random error rather than the relative importance of more contemporaneous measures.

In conclusion, we have demonstrated that maintenance of good circadian regulation of HPA activity is associated with better physical performance in later life. This is consistent with a generalized theory that HPA axis dysfunction may be associated with reduced life expectancy and a faster rate of ageing. However, it is also possible that early dysregulation of the HPA axis is also a manifestation of more rapid ageing. Early life factors, socio-economic status and psychological stressors may all contribute to life-course influences on ageing and socio-economic differences in physical performance which may, in turn, be partially mediated by the HPA axis. Further work needs to confirm these findings across other data sets and examine whether there are modifiable factors that may ameliorate these effects so that future primary prevention strategies can be designed and evaluated.

Supplementary Data

Supplementary Data are available at *IJE* online.

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KEY MESSAGES

- This is the first study examining the relationship between repeat cortisol measures (over 20 years) and physical performance in later life.
- Dysregulation of the HPA axis is associated with worse physical performance in later life.
- Higher night-time cortisol and a smaller diurnal drop measured contemporaneously in later life were associated with worse physical performance.
- A heightened stress-induced morning cortisol response in a normal population may be indicative of a healthier HPA axis.

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