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Independent effects of obesity and cortisol in predicting cardiovascular risk factors in men and women

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Abstract. Walker BR, Soderberg S, Lindahl B, Olsson T (University of Edinburgh, Western General Hospital, Edinburgh, UK and the University of Umea, University Hospital, Umea, Sweden). Independent effects of obesity and cortisol in predicting cardiovascular risk factors in men and women. *J Intern Med* 2000; **247:** 198–204.

Objectives. Recent data suggest that higher plasma cortisol may be associated with hypertension and insulin resistance in otherwise healthy men, as it is in Cushing's syndrome. However, obesity in women is associated with lower plasma cortisol concentrations. This study sought to establish whether plasma cortisol is associated with cardiovascular risk factors in women as it is in men, and whether these relationships in either sex are confounded by obesity.
Design. A population-based cross-sectional study.
Setting. The MONICA study in northern Sweden.
Subjects. From a target cohort of 2500, 1921 subjects took part and 226 were randomly selected because they attended between 07.00 and 09.00 h after an overnight fast. A 75 g oral glucose tolerance test was performed and blood sampled at baseline and 2 h after glucose.
Results. Plasma cortisol was lower in relatively obese subjects: in men, this was observed only in the 2 h sample ($r = -0.23$, $P = 0.02$) and in women only in the fasting sample ($r = -0.26$, $P < 0.01$). Simple regression analysis did not identify relationships between plasma cortisol and blood pressure, serum lipids, fasting insulin or glucose tolerance. However, after adjusting for the effect of obesity by multiple regression, higher plasma cortisol was independently associated with higher diastolic blood pressure in men ($r = 0.21$, $P = 0.04$) but not in women, and higher fasting serum triglyceride levels in women ($r = 0.28$, $P < 0.001$) but not in men.
Conclusions. Increasing obesity and plasma cortisol concentrations make independent and sex-specific contributions to variations in blood pressure and aspects of the insulin resistance syndrome. Adverse cardiovascular risk is greatest in those with the combination of obesity and failure to downregulate plasma cortisol levels.

Keywords: blood pressure, glucocorticoids, insulin resistance, obesity.

Introduction

High circulating levels of cortisol in Cushing's syndrome are associated with numerous clinical and biochemical abnormalities, including hypertension, insulin resistance, glucose intolerance and central obesity. A number of recent studies have explored the possibility that a more subtle increase in cortisol action contributes to the clustering of these cardiovascular risk factors in what is commonly referred to as the 'metabolic syndrome' or the 'insulin resistance syndrome'. In patients with established cardiovascular risk factors, including essential hypertension, glucose intolerance and insulin resistance, there is evidence for a variety of changes in glucocorticoid physiology [1–7]. These include increased cortisol secretion and circulating concentrations, impaired peripheral metabolism of cortisol, and increased tissue sensitivity to glucocorticoids as measured by the intensity of skin blanching following topical application of steroids. However, in subjects who are also obese the situation is more complicated: although 24 h cortisol excretion is enhanced in obesity, plasma

cortisol concentrations are lower, at least in the morning [8–13].

Against this background, it is important to extend the existing epidemiological studies which relate cortisol levels with cardiovascular risk factors. Most of the cross-sectional studies published so far have been performed in men. One cohort included women, but only relatively elderly subjects (aged 55–80 years) [6]. By contrast, most of the case–control studies of cortisol in obese subjects have only included women. Therefore, we do not know if cortisol levels correlate with blood pressure and insulin sensitivity across the physiological range in women as they do in men, or the extent to which these relationships are confounded by coexisting obesity. In the current study, we sought to establish correlates of plasma cortisol in a large cohort of men and women.

Materials and methods

Subject recruitment and selection

The study was performed within the framework of the northern Sweden MONICA project, a part of the WHO MONICA (Monitoring of Trends and Determinants in Cardiovascular Disease) project. Using continuously updated population registers in Norbotten and Västerbotten, the two northernmost provinces of Sweden, a total population of 367 000 in the 25–74 year age range was identified. In 1994, 2500 individuals (250 men and 250 women randomly selected from each of five 10-year age groups) were invited to participate. In total, 1921 subjects (76.8%) attended for clinical and biochemical measurements [14]. In this paper, a sample of 226 subjects were selected on the random basis that they had attended the clinic between 07.00 and 09.00 h on the day of measurements. The characteristics of the 226 participants in this study did not differ from those of the total sample of 1921 subjects (data not shown).

Protocol

The study was approved by the Research Ethics Committee of Umeå University.

Participants were instructed not to use tobacco and to avoid strenuous physical activity during the hour preceding the examination. Weight, height and circumferences at the narrowest part of the waist and the broadest part of the hip were recorded with the subject standing. Subjects then sat down. After 5 min, blood pressure was recorded with a Hawksley random zero sphygmomanometer and a venous sample was withdrawn for determination of lipids, glucose, insulin and cortisol. A 75 g oral glucose tolerance test was performed and a second venous sample was withdrawn for glucose and cortisol after 2 h. Plasma samples were stored at –70°C.

Laboratory methods

Plasma cortisol was measured by radioimmunoassay (RIA) [15]. Plasma glucose was analysed by the hexokinase method (Boehringer Mannheim, Germany) on a Hitachi 717 analyser (Tokyo, Japan). Insulin was measured by microparticle enzyme immunoassay (MEIA) (Abbott Laboratories, IL, USA). The detection limit was 1.0 mU L^{-1}, and the interassay coefficient of variation (CV) was 6.7% at the level of 7.9 mU L^{-1}. Cross-reactivity with C-peptide/glucagon was non-detectable and 0.005% with proinsulin. Serum triglyceride, cholesterol and HDL cholesterol were determined by enzymatic methods (Boehringer Mannheim GmbH, Mannheim, Germany).

Statistics

One subject who was morbidly obese (BMI $>$ 40 kg m^{-2}), two pregnant subjects and seven patients receiving oral or inhaled corticosteroid therapy were excluded from all analyses. For analyses which included blood pressure, subjects taking antihypertensive medication (n = 26) were excluded.

Data are described as mean ± SD. Data for plasma insulin and cortisol had a skewed distribution and were normalized by logarithmic transformation before analysis.

Results

Characteristics of the participants

Characteristics of the participants are shown in Tables 1 and 2. Women were a little younger and thinner than men, and had lower blood pressure,

Table 1 Clinical characteristics of participants

	Male	Female	*P*-value (Student's *t*-test)
n	105	110	
Age (years)	52.0 ± 15.4 (range 25–74)	48.4 ± 13.6 (range 25–74)	0.08
Body mass index (kg m^{-2})	26.2 ± 3.3	25.2 ± 4.2	0.04
Waist/hip ratio	0.94 ± 0.06	0.82 ± 0.08	<0.001
Systolic blood pressure (mmHg)	131.3 ± 19.7	125.4 ± 22.8	0.05
Diastolic blood pressure (mmHg)	82.4 ± 11.4	77.6 ± 11.2	0.002
Taking antihypertensives	13	13	
Postmenopausal	–	52	
Taking oestrogens	0	44	
Diabetes mellitus	2	2	
Impaired glucose tolerance	8	6	
Smokers	18	24	
Established ischaemic heart disease	6	2	
Established cerebrovascular disease	3	2	

Data are means ± SD.

serum triglycerides and fasting plasma glucose, and higher serum HDL cholesterol. A few patients in each group had established cardiovascular disease or were being treated for risk factors.

In all the participants, plasma cortisol concentrations fell during the glucose tolerance test (from 375 ± 154 to 235 ± 105 nmol L^{-1}; $P < 0.0001$). Plasma cortisol was not different between sexes, between postmenopausal and premenopausal women whether or not they were receiving oestrogen, in patients with impaired glucose tolerance or diabetes, in patients with established cardiovascular disease, or in smokers (data not shown). However, although fasting plasma cortisol was not significantly different in patients receiving treatment for hypertension (341 ± 158 nmol L^{-1} in patients vs. 379 ± 153 nmol L^{-1} in controls; $P = 0.17$), post-glucose plasma cortisol was lower in hypertensive patients (187 ± 79 vs. 242 ± 106 nmol L^{-1}; $P < 0.004$).

Correlates of plasma cortisol in men and women

Given the differences in cardiovascular risk factors between men and women, relationships with plasma cortisol were explored separately in the two groups (Table 3). In men, lower 2 h post-glucose plasma cortisol was associated with generalized obesity and hyperinsulinaemia. In women, lower fasting plasma cortisol was associated with central obesity.

Table 2 Biochemical characteristics of participants

	Male	Female	*P*-value (Student's *t*-test)
Serum total cholesterol (mM)	6.16 ± 1.36	6.23 ± 1.41	0.69
Serum triglyceride (mM)	1.78 ± 1.55	1.41 ± 0.72	0.04
Serum HDL cholesterol (mM)	1.17 ± 0.30	1.52 ± 0.40	<0.001
Fasting plasma glucose (mM)	5.52 ± 0.71	5.26 ± 0.71	0.01
120 min post-glucose plasma glucose (mM)	5.42 ± 1.83	5.59 ± 1.43	0.45
Fasting plasma insulin (mU L^{-1})	7.77 ± 4.95	7.52 ± 4.20	0.69
Fasting plasma cortisol (nM)	383 ± 131	367 ± 173	0.44
120 min post-glucose plasma cortisol (nM)	232 ± 89	239 ± 119	0.65

Data are means ± SD.

Table 3 Correlates of plasma cortisol

	Fasting plasma cortisol[a]		120 min post-glucose plasma cortisol[a]	
	Male	Female	Male	Female
Age	−0.03	−0.13	−0.11	0.06
	0.74	0.18	0.26	0.53
Body mass index	0.03	−0.12	−0.23	−0.10
	0.78	0.20	0.02	0.30
Abdominal circumference	0.03	−0.25	−0.17	−0.18
	0.75	0.01	0.08	0.07
Waist/hip ratio	−0.04	−0.26	−0.06	−0.17
	0.73	<0.01	0.52	0.08
Systolic blood pressure[b]	0.17	−0.06	0.03	−0.01
	0.11	0.56	0.78	0.90
Diastolic blood pressure[b]	0.19	0.07	0.04	0.04
	0.07	0.53	0.70	0.70
Serum total cholesterol	0.01	−0.01	−0.03	0.07
	0.94	0.91	0.79	0.46
Serum triglyceride	0.10	0.15	0.01	0.13
	0.33	0.12	0.99	0.19
Serum HDL cholesterol	0.10	−0.07	0.13	0.05
	0.29	0.45	0.18	0.59
Fasting plasma glucose	−0.12	0.03	−0.12	0.04
	0.22	0.75	0.22	0.66
120 min post-glucose	−0.15	0.01	−0.17	0.07
plasma glucose	0.12	0.96	0.09	0.48
Fasting plasma insulin[a]	−0.12	0.05	−0.24	−0.04
	0.21	0.59	0.01	0.68

Each cell shows *r*-value followed by *P*-value for simple regression analyses comparing the two variables indicated by column and row headings.
[a]Analysed after logarithmic transformation.
[b]Patients on antihypertensive medication were excluded from this analysis.

Influence of obesity on association between plasma cortisol and other cardiovascular risk factors

Given that plasma cortisol was inversely correlated with indices of obesity, and that increasing obesity was associated with all other adverse cardiovascular risk factors (higher serum lipids, blood pressure, fasting plasma glucose, glucose intolerance and fasting plasma insulin/glucose ratio), it is possible that relationships between cardiovascular risk factors and plasma cortisol in simple linear regression analyses will be confounded by relationships with obesity. Relationships between cortisol and cardiovascular risk factors were therefore examined after correction for the potentially confounding effect of obesity by multiple regression analyses (Table 4). Abdominal circumference was chosen as the index of obesity in all subjects since this showed the most consistent relationship with cortisol in this study and in previous studies [16].

After adjusting for the influence of obesity, higher fasting plasma cortisol was associated with higher diastolic blood pressure in men and higher serum triglyceride and fasting plasma insulin levels in women, but not with other cardiovascular risk factors (Table 4). As a result, the highest diastolic blood pressures were recorded amongst men who had the combination of obesity and higher plasma cortisol, and the highest triglyceride and insulin levels were recorded amongst women who had the combination of obesity and higher plasma cortisol. By contrast, plasma cortisol 2 h post-glucose was not associated with other risk factors in similar analyses (Table 4).

Discussion

These data confirm that associations exist between cardiovascular risk factors and plasma cortisol concentrations, and extend previous studies by

Table 4 Correlates of plasma cortisol adjusted for relationship with obesity

	Male				Female			
	Abdo circ	Fasting cortisol[a]	Abdo circ	2 h cortisol[a]	Abdo circ	Fasting cortisol[a]	Abdo circ	2 h cortisol[a]
Systolic blood pressure[b]	0.25	0.19	0.25	0.07	0.40	0.03	0.39	0.03
	0.01	0.07	0.02	0.52	<0.001	0.74	<0.001	0.72
Diastolic blood pressure[b]	0.25	0.21	0.25	0.08	0.31	0.15	0.29	0.09
	0.01	0.04	0.02	0.44	0.002	0.14	0.01	0.39
Serum total cholesterol	0.21	0.03	0.21	0.02	0.42	0.09	0.42	0.15
	0.04	0.78	0.04	0.84	<0.001	0.32	<0.001	0.11
Serum triglyceride	0.12	0.11	0.12	0.03	0.53	0.28	0.50	0.21
	0.23	0.27	0.24	0.80	<0.001	0.001	<0.001	0.01
Serum HDL cholesterol	−0.28	0.06	−0.27	0.07	−0.28	−0.14	−0.24	−0.01
	0.01	0.52	0.01	0.44	0.01	0.15	0.01	0.92
Fasting plasma glucose	0.18	−0.14	0.17	−0.08	0.44	0.14	0.43	0.12
	0.07	0.15	0.08	0.41	<0.001	0.13	<0.001	0.19
120 min post-glucose	0.20	−0.15	0.18	−0.13	0.42	0.11	0.41	0.14
plasma glucose	0.05	0.13	0.07	0.18	<0.001	0.24	<0.001	0.12
Fasting plasma insulin[a]	0.41	−0.10	0.39	−0.18	0.56	0.19	0.52	0.05
	<0.001	0.26	<0.001	0.04	<0.001	0.02	<0.001	0.54

Abdo circ, abdominal circumference.
Each cell shows *r*-value followed by *P*-value for multiple regression analyses for the dependent variables, indicated by row headings, using independent variables indicated in the column headings (i.e. either abdominal circumference and fasting cortisol; or abdominal circumference and 2 h cortisol).
[a]Analysed after logarithmic transformation.
[b]Patients on antihypertensive medication were excluded from this analysis.

demonstrating that these associations differ in men and women. Specifically, plasma cortisol was lower in obese subjects, but after correction for the relationship with obesity, fasting plasma cortisol was higher in subjects with higher blood pressure and higher serum triglycerides. The association with blood pressure was stronger in men and the association with other features of insulin resistance (hypertriglyceridaemia and hyperinsulinaemia) was stronger in women.

The most striking inference from these data is that there are opposing influences on plasma cortisol levels. Obesity was associated with lower fasting plasma cortisol; hypertension was associated with higher fasting plasma cortisol. The mechanism whereby obesity is associated with lower morning plasma cortisol is not established. Studies which include measurement of 24 h urinary cortisol excretion, adrenal responsiveness to adrenocorticotrophic hormone (ACTH) or cortisol responses to other stimuli confirm that cortisol secretion is enhanced in obesity [9–11, 13, 17, 18]. However, the current results are consistent with previous observations that plasma and salivary cortisol concentrations during the morning peak of secretion are lower in obese subjects [5, 8, 12]. This paradox may be explained either because central control of the hypothalamic–pituitary–adrenal axis is altered in obesity resulting in a blunted diurnal variation in cortisol levels (lower in the morning but higher in the evening) [12], or because peripheral metabolic clearance of cortisol is enhanced in obesity resulting in a tendency for lower plasma cortisol concentrations which is only partly compensated for by enhanced cortisol secretion [16]. Interestingly, we have shown in another study that disturbances in cortisol metabolism in obesity differ between men and women, such that in men enhanced metabolism of cortisol by 5α-reductase in central obesity is compensated for by enhanced reactivation of cortisol by 11β-hydroxysteroid dehydrogenase type 1; this compensation was not observed in women [16]. These observations may relate to gender differences in dysregulation of growth hormone and sex steroid levels in obesity [11, 12]. They may also provide the explanation for the contrast in our data between the weak association of lower plasma cortisol with generalized obesity in men (evident only in the sample taken 2 h after glucose) and the stronger association with central obesity in women. With a

lesser confounding effect of obesity in men, the relationship between plasma cortisol and blood pressure may then be more obvious in men than in women. However, this does not explain why the positive relationship between plasma cortisol and features of insulin resistance was more apparent in women.

There are many potential explanations for the higher fasting plasma cortisol in subjects with hypertension and hypertriglyceridaemia. The assessment of circulating cortisol concentrations in epidemiological studies is by no means straightforward. All measurements, however strictly timed, are potentially influenced by the psychological stress of sample collection and by the magnitude of peak/trough excursion, as well as by the area under the circadian profile of plasma cortisol levels. Measurements in the morning, during the diurnal peak of cortisol, are subject to the greatest variation and therefore are least likely to detect subtle differences in cortisol secretion. However, despite these limitations, our observations in this and previous studies in men [5] suggest that more detailed studies of the dynamic control of the hypothalamic–pituitary–adrenal axis in subjects with cardiovascular risk factors will be highly informative. Indeed, there is already corroborative evidence of the importance of cortisol in subjects with cardiovascular risk factors from other studies: increased cortisol secretion occurs independently of obesity in other syndromes associated with insulin resistance, including polycystic ovarian syndrome [19, 20]; and cortisol metabolism, which influences the rate of decline of plasma cortisol following the diurnal peak of secretion, is also altered in essential hypertension [2, 3].

In the current study, we also report relationships between cardiovascular risk factors and a second measurement of plasma cortisol, taken 2 h after the fasting sample and following an oral glucose tolerance test. Post-glucose plasma cortisol proved to be less informative than the fasting value. There are several reasons why later measurement of cortisol may not show the same relationships as the fasting value. Not only are the two measurements separated by 2 h during which cortisol will fall due to diurnal variation, but also there may be an influence of glucose ingestion on cortisol levels [17] and the second measurement may be less affected by psychological stress once the subject has become accustomed to the experimental environment. Nevertheless, it is intriguing that cortisol in the fasting sample is correlated more strongly with other abnormalities, since this is consistent with the suggestion that difference in response to psychological stress is an important determinant of the observed relationships between cortisol and cardiovascular risk factors [12].

Finally, although the current study does not shed any new light on the mechanism of the association between obesity and lower plasma cortisol concentrations, it highlights that it may not be this observation which is most important. The multiple regression analyses in the current study demonstrate that both increasing obesity and higher fasting plasma cortisol concentrations predict adverse cardiovascular risk factors independently. So, the combination of obesity with higher plasma cortisol is the most dangerous. In other words, it may be that we should focus on the mechanisms which prevent the 'normal' fall in morning plasma cortisol that occurs in most obese subjects, since therein may lie the explanation for interindividual differences in the extent to which obesity amplifies cardiovascular risk. It is only a proportion of even morbidly obese subjects who develop clinical hypertension or diabetes, and the current data suggest that these will be characterized by higher, rather than lower, plasma cortisol levels.

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References

1 Walker BR, Best R, Shackleton CHL, Padfield PL, Edwards CRW. Increased vasoconstrictor sensitivity to glucocorticoids in essential hypertension. *Hypertension* 1996; **27**: 190–96.

2 Walker BR, Stewart PM, Shackleton CHL, Padfield PL, Edwards CRW. Deficient inactivation of cortisol by 11β-hydroxysteroid dehydrogenase in essential hypertension. *Clin Endocrinol (Oxf)* 1993; **39**: 221–27.

3 Soro A, Ingram MC, Tonolo G, Glorioso N, Fraser R. Evidence of coexisting changes in 11β-hydroxysteroid dehydrogenase and 5β-reductase activity in patients with

untreated essential hypertension. *Hypertension* 1995; **25**: 67–70.
4 Walker BR, Phillips DIW, Noon JP *et al.* Increased glucocorticoid activity in men with cardiovascular risk factors. *Hypertension* 1998; **31**: 891–95.
5 Phillips DIW, Barker DJP, Fall CHD *et al.* Elevated plasma cortisol concentrations: an explanation for the relationship between low birthweight and adult cardiovascular risk factors. *J Clin Endocrinol Metab* 1998; **83**: 757–60.
6 Stolk RP, Lamberts SWJ, de Jong FH, Pols HAP, Grobbee DE. Gender differences in the associations between cortisol and insulin sensitivity in healthy subjects. *J Endocrinol* 1996; **149**: 313–18.
7 Filipovsky J, Ducimetiere P, Eschwege E, Richard JL, Rosselin G, Claude JR. The relationship of blood pressure with glucose, insulin, heart rate, free fatty acids and plasma cortisol levels according to degree of obesity in middle-aged men. *J Hypertens* 1996; **14**: 229–35.
8 Ljung T, Andersson B, Bengtsson B, Bjorntorp P, Marin P. Inhibition of cortisol secretion by dexamethasone in relation to body fat distribution: a dose–response study. *Obesity Research* 1996; **4**: 277–82.
9 Hautanen A, Adlercreutz H. Altered adrenocorticotropin and cortisol secretion in abdominal obesity: implications for the insulin resistance syndrome. *J Intern Med* 1993; **234**: 461–69.
10 Pasquali R, Cantobelli S, Casimirri F *et al.* The hypothalamic-pituitary-adrenal axis in obese women with different patterns of body fat distribution. *J Clin Endocrinol Metab* 1993; **77**: 341–46.
11 Marin P, Darin M, Amemiya T, Andersson B, Jern S, Bjorntorp P. Cortisol secretion in relation to body fat distribution in obese premenopausal women. *Metabolism* 1992; **41**: 882–86.
12 Rosmond R, Dallman MF, Bjorntorp P. Stress-related cortisol secretion in men: relationships with abdominal obesity and endocrine, metabolic and haemodynamic abnormalities. *J Clin Endocrinol Metab* 1998; **83**: 1853–59.
13 Strain GW, Zumoff B, Kream J, Strain JJ, Levin J, Fukushima D. Sex difference in the influence of obesity on the 24 hr mean plasma concentration of cortisol. *Metabolism* 1982; **31**: 209–12.
14 Soderberg S, Olsson T, Eliasson M, Johnson O, Ahren B. Plasma leptin levels are associated with abnormal fibrinolysis in men and postmenopausal women. *J Intern Med* 1999; **245**: 533–43.
15 Whitworth JA, Stewart PM, Burt D, Atherden SM, Edwards CRW. The kidney is the major site of cortisone production in man. *Clin Endocrinol (Oxf)* 1989; **31**: 355–61.
16 Andrew R, Phillips DIW, Walker BR. Obesity and gender influence cortisol secretion and metabolism in man. *J Clin Endocrinol Metab* 1998; **83**: 1806–809.
17 Korbonits M, Trainer PJ, Nelson ML et al. Differential stimulation of cortisol and dehydroepiandrosterone levels by food in obese and normal subjects: relation to body fat distribution. *Clin Endocrinol* 1996; **45**: 699–706.
18 Pasquali R, Anconetani B, Chattat R et al. Hypothalamic-pituitary-adrenal axis activity and its relationship to the autonomic nervous system in women with visceral and subcutaneous obesity: effects of the corticotrophin-releasing factor/arginine-vasopressin test and of stress. *Metab Clin Exper* 1996; **45**: 351–56.
19 Stewart PM, Shackleton CHL, Beastall GH, Edwards CRW. 5alpha-reductase activity in polycystic ovarian syndrome. *Lancet* 1990; **335**: 431–33.
20 Rodin A, Thakkar H, Taylor N, Clayton R. Hyperandrogenism in polycystic ovary syndrome: evidence of dysregulation of 11beta-hydroxysteroid dehydrogenase. *N Engl J Med* 1994; **330**: 460–65.

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