Does sympathoadrenal activity predict changes in body fat? An 18-y follow-up study1–3

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ABSTRACT

Background: Whether alterations in the sympathoadrenal system contribute to obesity or, rather, are consequences of it, is an unresolved issue.

Objective: We hypothesized that the sympathoadrenal system plays a predictive role in the development of body fat.

Design: At entry, arterial plasma epinephrine and norepinephrine concentrations were measured in 99 healthy men (x ± SD age: 19.3 ± 0.4 y) at rest and during a mental stress test and a cold pressor test. Body mass index (BMI; in kg/m²), waist circumference, and triceps skinfold thickness were measured at entry and after 18 y of follow-up.

Results: Eighty subjects (81%) were available for follow-up analyses after a mean (±SD) of 18.0 ± 0.9 y. The epinephrine responses to the mental stress test (E_{MST}) showed a negative relation to changes in BMI (P = 0.01) and waist circumference (P = 0.007). The mean increase in BMI was 6.3 among subjects in the lowest E_{MST} quartile and 3.7 in the remaining subjects. In multiple regression analyses corrected for level of exercise, BMI, waist circumference, and triiceps skinfold thickness at entry, E_{MST} was found to be a consistent negative predictor of future BMI (P = 0.005), waist circumference (P = 0.001), and triceps skinfold thickness (P = 0.05).

Conclusions: We present the first long-term follow-up study in whites showing that the epinephrine response to mental stress is a negative predictor of future BMI, waist circumference, and triceps skinfold thickness after 18 y of follow-up. These findings may provide further insights into the pathophysiology of obesity. Am J Clin Nutr 2008;87:1596–601.

INTRODUCTION

Obesity and overweight are of growing worldwide concern and reaching epidemic proportions (1). Obesity is a strong predictor of mortality (2), and it was recently shown that overweight is related to increased mortality (3). Animal and twin studies indicate that 25–40% of the variability in human body weight may be accounted for by genetic factors (4), and the sympathoadrenal system and β-adrenergic receptors are thought to play an important role (5–7). However, whether alterations in the sympathoadrenal system contribute to obesity or, rather, are consequences of it, is still an unresolved issue (8) because most of the data available are from cross-sectional studies. Only 2 longitudinal studies have been performed. Tatarnini et al (9) studied 44 Pima Indians over a period of 3.3 y, assessing the predictive power of 24-h urinary catecholamine excretion rates. They found that baseline epinephrine excretion was negatively related to changes in the waist-to-thigh circumference ratio, whereas norepinephrine excretion correlated negatively with body weight gain. Masuo et al (10), on the other hand, found resting plasma norepinephrine to be a positive predictor of changes in body mass index (BMI; in kg/m²) over a 5-y period in 433 Japanese subjects. Hence, the available data are contradictory, and the follow-up periods are short. Moreover, there are no longitudinal data on whites. The Pima Indian population has one of the highest prevalences of obesity in the world. They have lower muscle sympathetic nervous activity (11) and a lower chronotropic sensitivity to β-adrenergic stimulation than do whites (12). Thus, findings in studies on this population cannot be generalized to whites.

The present 18-y follow-up study investigated the predictive role of sympathoadrenal activity in the development of body fat in whites. We hypothesized that arterial plasma epinephrine and norepinephrine at rest and during laboratory stress were related to the increase in BMI, waist circumference, and triceps skinfold thickness. We also examined whether reactivity to 2 separate stress tests, a cold pressor test and a mental stress test, would differ in their predictive power. The 2 tests are supposed to represent different reactivity mechanisms, α- compared with β-adrenergic responses, respectively (13).

SUBJECTS AND METHODS

Participants

The local Ethics Committee approved the study, and the procedures followed were in accordance with institutional guidelines. Informed consent was obtained from each subject both at entry and at follow-up. All 19-y-old men in Norway are required to undergo a medical examination as part of the military draft procedure. The participants were seated for 5 min and then a

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2 The initial phase (entry) of the study was supported financially by the Norwegian Council of Cardiovascular Diseases. The final phase of the study (follow-up) was supported financially by the Research Council at Ullevaal University Hospital, Oslo, Norway.

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Adrenomedullary Reactivity and Body Fat

Trained physician measured blood pressure (BP) once with an automatic auscultatory device (Boso-digital II S; Bosh & Sohn GmbH, Jungingen, Germany) or a newly calibrated mercury sphygmomanometer. None of the subjects were informed about the results of the BP recordings at this stage, to avoid effects of hypertension labeling on responses to the stress tests (14, 15). Mean BP was thereafter calculated as diastolic BP + pulse pressure/3. A total of 99 subjects were selected from the military draft screening: 30 were in the 1st percentile, 30 were in the 50th percentile, and 39 were in the 95th–99th percentile of the mean BP distribution. This selection procedure was followed to ensure a good representation of BP readings, because BP is related to sympathoadrenal activity (16). All subjects were white, except one who was half Asian and half white. The subjects were healthy and had no history of diabetes, renal disease, elevated BP, or other cardiovascular disease and had normal results from a physical examination, electrocardiogram, routine blood tests, and urinalysis. None of the subjects were receiving medical treatment or abused drugs or alcohol.

Examination at entry

The protocol at baseline is described in detail elsewhere (17) and was performed in 1986–1989. Resting heart rate and BP were recorded after the subjects had rested for 15 min in a sitting position; the same equipment used during the screening was used. Height, weight, and waist circumference were measured while the subjects were standing. BMI was calculated as weight (kg) divided by height² (m). Waist circumference was measured in 34 subjects at entry. Skinfold-thickness measurements were performed with a Harpenden skinfold caliper. A fold of skin, including subcutaneous tissue, was seized between thumb and forefinger, half-way down the triceps. The mean of 2 measurements was used. A short Teflon catheter (Venflon, 19G; Viggo AB, Hälsingborg, Sweden) was introduced under local anesthesia or without epinephrine (Xylocain; AstraZeneca, Wilmington, DE) into the left brachial artery for blood sampling.

The cold pressor test lasted for 1 min. The right hand was completely immersed in ice water (0–2 °C). During the mental arithmetic test, the subjects were asked to subtract the number “13” repetitively starting from “1079” for 5 min, while a metronome made noise at a frequency of 2 Hz. They were informed about any miscalculation. After each test there was a 30-min recovery period. Arterial blood for catecholamine assay was collected after 30 min of supine rest, before and during the cold pressor test (after 0.5 and 1 min), and before and during the mental stress test (after 1, 3, and 5 min). Catecholamine responses to stress tests were calculated as the mean value during stress, subtracting the baseline value before the test. Blood was drawn into 2-mL glass tubes containing glutathione and EGTA, and plasma catecholamines were measured with a radioenzymatic technique according to Peuler and Johnsen (18) as previously reported (15, 19). All the blood samples were analyzed by the same technician.

Examination at follow-up

Follow-up examinations were conducted from 2005 to 2006. The mean (±SD) length of follow-up was 18.0 ± 0.9 y; 81 of the original 99 subjects (82%) were available for examination at follow up. A total of 18 subjects were not reexamined; 1 was excluded because of probable intravenous drug addiction, 2 lived abroad and were not able to attend the examinations, 4 did not answer any letters or calls, and 11 did not want to participate. The subjects that were not available for follow-up did not differ from the reexamined subjects in resting BP, heart rate, BMI, waist circumference, or catecholamine stress responses at entry. One subject who was reexamined had ulcerative colitis and had to be excluded from further analyses because of a previous colectomy and an excessive intake of water and salt.

Follow-up, 21 subjects (25.9%) reported having one or more of the following diseases: hypertension (n = 9), hypercholesterolemia (n = 12), diabetes mellitus (n = 3), and myocardial infarction (n = 1). Eight of these subjects used one or more of the following medications regularly: angiotensin receptor blockers (n = 3), β-blockers (n = 3), angiotensin-converting enzyme inhibitor (n = 1), statins (n = 2), antidiabetics (n = 3), and acetylsalicylic acid (n = 1). Each subject was studied in the same room at 0800 each day after fasting overnight. They abstained from any medication use or smoking for 8 h before and from alcohol for 24 h before the examination. BMI, waist circumference, and triceps skinfold thickness were measured and calculated in the same way as at entry.

Resting BP was measured 3 times on the left arm after the subjects had sat for ≥15 min, and was calculated as the mean of the last 2 measurements. Standardized questionnaires were used to collect information about concomitant diseases, medication, family history, education, occupation, and exercise. Baseline blood samples were drawn after a minimum of 30 min of supine rest.

Statistics

The data were analyzed by using the statistical package SPSS version 14.0 for WINDOWS (SPSS Inc, Chicago, IL). Paired-samples t tests were used to analyze possible changes in normally distributed continuous variables from entry to follow-up, and the Wilcoxon’s signed ranked test was applied when normality was not achieved by log-normal transformation. Categorical variables were analyzed by sign test. Associations between continuous variables were assessed by using Pearson’s correlations, whereas chi-square testing with linear-by-linear association was used for dichotomous variables.

To adjust for possible confounders, linear regression analysis was performed with BMI, waist circumference, and triceps skinfold thickness at follow-up as dependent variables and level of exercise at follow-up and selected variables measured at entry as independent variables. Exercise was graded in 3 levels: 1, <1 h of exercise per week; 2, >1 h of moderate exercise per week; and 3, >1 h of heavy exercise a week. The data are presented as means ± SD unless otherwise indicated. Null hypotheses were rejected if the 2-tailed P value was <0.05.

RESULTS

Descriptive characteristics

Characteristics of the participants at baseline and follow-up are presented in Table 1. They were 19.3 y of age (range: 18.2–20.8 y) on the first visit, and 37.3 y (range: 35.4–38.9 y) by the time of the reexamination. Systolic BP (P = 0.003) and diastolic...
Catecholamines and cardiovascular disease risk factors at entry

There were no significant relations between cardiovascular disease risk factors, the resting epinephrine concentration, or the epinephrine response to the mental stress test at entry (Table 2). The epinephrine response to the cold pressor test was positively related to resting diastolic BP ($r = 0.39$, $P = 0.001$). Smoking was positively related to resting norepinephrine ($P = 0.008$) and negatively related to the norepinephrine response to mental stress ($P = 0.004$). BMI, waist circumference, and triceps skinfold thickness at entry did not correlate with any of the catecholamine variables.

Prediction of 18-y changes in BMI, waist circumference, and triceps skinfold thickness

The absolute epinephrine response to mental stress at entry was negatively related to the changes in BMI ($r = -0.31$, $P = 0.01$), waist circumference ($r = -0.49$, $P = 0.007$), and triceps skinfold thickness ($r = -0.22$, $P = 0.10$) after 18 y. A threshold between the first and second stress response quartile is suggested in Figure 1. Whereas the mean BMIs at entry and follow-up were 22.1 and 28.4, respectively, in the first quartile, the mean BMIs in the 3 higher quartiles combined were 22.9 and 26.6, respectively. None of the other epinephrine or norepinephrine variables during rest or the cold pressor test were significantly related to changes in BMI, waist circumference, or triceps skinfold thickness.

In multiple regression analyses of BMI, waist circumference and triceps skinfold thickness at follow-up, the absolute epinephrine response to mental stress at entry was a consistent negative predictor for all 3 body fat variables after 18 y, after adjustment for level of exercise and entry BMI, waist circumference, and triceps skinfold thickness (Table 3). The norepinephrine response at entry was a weak positive predictor of future waist

### Table 1
Descriptive characteristics at entry to the study and after 18 y of follow-up[^1]

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>No. of subjects</th>
<th>Entry</th>
<th>Follow-up</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (y)</td>
<td>80</td>
<td>19.3 ± 0.4[^2]</td>
<td>37.3 ± 0.8[^3]</td>
</tr>
<tr>
<td>Systolic blood pressure (mm Hg)</td>
<td>76</td>
<td>126 ± 20</td>
<td>131 ± 16[^4]</td>
</tr>
<tr>
<td>Diastolic blood pressure (mm Hg)</td>
<td>76</td>
<td>70 ± 17</td>
<td>89 ± 10[^5]</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>75</td>
<td>66 ± 15</td>
<td>64 ± 12</td>
</tr>
<tr>
<td>BMI (kg/m^2)</td>
<td>80</td>
<td>22.4 ± 3.0</td>
<td>26.7 ± 4.3[^5]</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>34</td>
<td>83.0 ± 8.6</td>
<td>94.5 ± 11.1</td>
</tr>
<tr>
<td>Triceps skinfold thickness (mm)</td>
<td>70</td>
<td>10.0 ± 4.2</td>
<td>12.2 ± 6.1[^5]</td>
</tr>
<tr>
<td>Total serum cholesterol (mmol/L)</td>
<td>79</td>
<td>4.0 ± 0.7</td>
<td>4.9 ± 0.9[^5]</td>
</tr>
<tr>
<td>Serum HDL cholesterol (mmol/L)</td>
<td>78</td>
<td>1.1 ± 0.2</td>
<td>1.2 ± 0.3</td>
</tr>
<tr>
<td>Serum triacylglycerols (mmol/L)</td>
<td>79</td>
<td>0.8 ± 0.4</td>
<td>1.3 ± 0.9[^4]</td>
</tr>
<tr>
<td>Plasma glucose (mmol/L)</td>
<td>69</td>
<td>4.2 ± 0.5</td>
<td>5.1 ± 0.8[^4]</td>
</tr>
<tr>
<td>Daily smokers [n (%)]</td>
<td>78</td>
<td>28 (36)</td>
<td>20 (26)</td>
</tr>
<tr>
<td>Epinephrine during rest (pg/mL)</td>
<td>80</td>
<td>45.8 ± 31.4</td>
<td>—</td>
</tr>
<tr>
<td>Epinephrine during CPT (pg/mL)</td>
<td>74</td>
<td>78.5 ± 48.5</td>
<td>—</td>
</tr>
<tr>
<td>Norepinephrine during rest (pg/mL)</td>
<td>80</td>
<td>116.1 ± 76.8</td>
<td>—</td>
</tr>
<tr>
<td>Norepinephrine during CPT (pg/mL)</td>
<td>74</td>
<td>78.5 ± 48.5</td>
<td>—</td>
</tr>
<tr>
<td>Norepinephrine during CPT (pg/mL)</td>
<td>74</td>
<td>156.4 ± 75.3</td>
<td>—</td>
</tr>
</tbody>
</table>

[^1]: Blood samples were collected after the subjects fasted overnight. Catecholamines represent arterial plasma concentrations. MST, mental stress test; CPT, cold pressor test. Paired-samples t tests were used for normally distributed variables, Wilcoxon’s signed-rank test was used when normality was not achieved by log-normal transformation, and the proportions of smokers were compared by using a sign test.

[^2]: ± SD (all such values).

[^3]: Significantly different from entry: [^1] $P < 0.001$, [^4] $P < 0.05$.

### Table 2
Associations between variables and epinephrine response at entry

<table>
<thead>
<tr>
<th>Variable at entry</th>
<th>1 (11.3 ± 19.7[^1])</th>
<th>2 (44.0 ± 27.8)</th>
<th>3 (100.1 ± 54.7)</th>
<th>4 (146.0 ± 75.7)</th>
<th>$P^2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>BMI (kg/m^2)</td>
<td>22.1 ± 3.2</td>
<td>22.6 ± 2.8</td>
<td>23.7 ± 2.6</td>
<td>22.5 ± 3.5</td>
<td>0.95</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>80.3 ± 8.1</td>
<td>82.2 ± 7.2</td>
<td>87.0 ± 9.0</td>
<td>83.0 ± 13.6</td>
<td>0.42</td>
</tr>
<tr>
<td>Triceps skinfold thickness (mm)</td>
<td>10.1 ± 4.5</td>
<td>9.0 ± 3.2</td>
<td>12.9 ± 5.7</td>
<td>9.4 ± 4.0</td>
<td>0.78</td>
</tr>
<tr>
<td>Systolic blood pressure (mm Hg)</td>
<td>120.1 ± 14.5</td>
<td>120.0 ± 21.5</td>
<td>130.9 ± 21.5</td>
<td>134.0 ± 22.5</td>
<td>0.15</td>
</tr>
<tr>
<td>Diastolic blood pressure (mm Hg)</td>
<td>64.8 ± 20.7</td>
<td>71.1 ± 17.8</td>
<td>71.9 ± 14.8</td>
<td>73.6 ± 14.6</td>
<td>0.39</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>67.6 ± 20.7</td>
<td>60.1 ± 10.2</td>
<td>68.1 ± 11.3</td>
<td>61.6 ± 15.4</td>
<td>0.37</td>
</tr>
<tr>
<td>Total serum cholesterol (mmol/L)</td>
<td>4.2 ± 0.7</td>
<td>3.9 ± 0.7</td>
<td>3.9 ± 0.8</td>
<td>4.1 ± 0.8</td>
<td>0.56</td>
</tr>
<tr>
<td>Serum HDL (mmol/L)</td>
<td>1.1 ± 0.2</td>
<td>1.2 ± 0.2</td>
<td>1.0 ± 0.2</td>
<td>1.2 ± 0.2</td>
<td>0.81</td>
</tr>
<tr>
<td>Serum triacylglycerols (mmol/L)</td>
<td>0.9 ± 0.5</td>
<td>0.8 ± 0.3</td>
<td>0.9 ± 0.4</td>
<td>0.8 ± 0.4</td>
<td>0.51</td>
</tr>
<tr>
<td>Fasting plasma glucose (mmol/L)</td>
<td>4.1 ± 0.6</td>
<td>4.1 ± 0.4</td>
<td>4.2 ± 0.5</td>
<td>4.2 ± 0.5</td>
<td>0.65</td>
</tr>
<tr>
<td>Daily smokers [n (%)]</td>
<td>7 (44)</td>
<td>8 (53)</td>
<td>5 (31)</td>
<td>3 (19)</td>
<td>0.08</td>
</tr>
<tr>
<td>Epinephrine during rest (pg/mL)</td>
<td>67.5 ± 44.2</td>
<td>41.4 ± 21.4</td>
<td>49.6 ± 26.4</td>
<td>30.2 ± 16.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Norepinephrine during rest (pg/mL)</td>
<td>140.2 ± 51.9</td>
<td>126.8 ± 33.0</td>
<td>126.4 ± 41.8</td>
<td>89.2 ± 39.7</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

[^1]: ± SD (all such values).

[^2]: Univariate Pearson correlation analyses were conducted between the various variables and epinephrine response during mental stress as a continuous variable. The $P$ value for smokers was derived with a chi-square test for linear-by-linear association.
circumference and a near significant positive predictor of BMI and triceps skinfold thickness.

**DISCUSSION**

The present 18-y follow-up study is the first to demonstrate that reduced adrenal medullary reactivity to mental stress is related to future weight gain in whites. The epinephrine response to mental stress was negatively related to changes in BMI and waist circumference. In supplementary multiple regression analyses, the epinephrine response was a highly consistent negative predictor of future BMI, waist, and triceps skinfold thickness after adjustment for possible confounders. The norepinephrine response to mental stress was a weak positive predictor of future waist circumference and did not significantly predict BMI and triceps skinfold thickness.

To assess sympathoadrenal activity, we measured arterial catecholamines, which were previously reported to be a more sensitive marker of overall sympathetic activity than venous sampling (19). There is uncertainty, however, about which part of the sympathetic nervous system may be the best determinant of weight development. Arterial samples reflect better the overall sympathetic activity, including spillover from heart and kidney, whereas venous samples for a larger part reflect muscle sympathetic activity. Release from muscle sympathetic nerves contributes to \( \approx 50\% \) of peripheral venous norepinephrine (20). Thus, if sympathetic activity in muscle tissues is the most important determinant of future body fat, venous measurements would have been preferable and could possibly explain the weak associations between norepinephrine activity and weight development compared with epinephrine. However, there are no indications that muscle sympathetic activity is crucial in this regard. Clinically, fat tends to deposit centrally, and the sympathetic stimulation of visceral areas is better reflected through arterial sampling. In assessing the activity in the adrenal medulla, arterial sampling is far better than venous sampling because \( \approx 50\% \) of epinephrine is cleared by peripheral tissues. Arterial concentrations are thus higher and more precisely determined than are venous concentrations. A limitation of the study was the lack of arterial catecholamine measurements at follow-up, because we were unable to decide whether individuals remained in the same stress quartile throughout the follow-up period.

Our main findings remained significant after adjustment for possible confounders. Heavy exercise was significantly related to both BMI and triceps skinfold thickness in the multiple regression analyses, which supports earlier findings (21). We had no information on total intake of calories or on thyroid status at
sympathetic stimulation of the diet-induced thermogenesis, accomplished by noradrenergic pathways believed to be sensed by the brain, which subsequently triggers weight gain in humans (6). Excessive calorie intake in humans is an important role in the development of obesity. Mice lacking the alpha2-adrenergic receptors (24, 25). Our finding that none of the catecholamine variables during rest or the cold pressor test were significantly related to changes in BMI, waist circumference, and triceps skinfold thickness. These findings will contribute to the ongoing debate concerning the relationship between obesity and sympathoadrenal activity and provide further insight into the pathophysiology of obesity.

We thank Ruth Amundsen for her expert assay of plasma catecholamines and the Joint Norwegian Medical Services, Head Quarters Defense Command, Norway, for their generous cooperation. The authors’ responsibilities were as follows—AF, IKE, SEK, and MR: designed the study; AF, LS, and MR: analyzed the data; and AF: drafted the manuscript. All authors contributed to the revision of the manuscript. None of the authors had any personal or financial conflicts of interest.

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