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BRIEF REPORT

Anxiety, depressive symptoms, and cardiac autonomic function in perimenopausal and postmenopausal women with hot flashes: a brief report

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Abstract

Objective: The aim of the study was to examine whether anxiety and depressive symptoms are associated with an adverse cardiac autonomic profile among midlife women with hot flashes.

Methods: Anxiety and depressive symptoms were evaluated by validated self-administered questionnaires among peri- and postmenopausal women in a randomized trial of slow-paced respiration for hot flashes. Pre-ejection period (PEP), a marker of sympathetic activation, and respiratory sinus arrhythmia (RSA), a marker of parasympathetic activation, were measured at baseline and 12 weeks using impedance cardiography and electocardiography. Multivariable repeated measures linear regression models examined associations between anxiety and depression symptoms and autonomic markers, corrected for multiple comparisons with Benjamini–Hochberg procedure, and adjusted for age and body mass index.

Results: Among the 121 participants, greater state anxiety was associated with shorter PEP, reflecting higher sympathetic activity ($\beta = -0.24$, $P = 0.02$). Greater trait anxiety and cognitive anxiety were associated with lower RSA, reflecting decreased parasympathetic activity ($\beta = -0.03$, $P < 0.01$ for Spielberger Trait Anxiety; $\beta = -0.06$, $P = 0.02$ for Hospital Anxiety and Depression Scale [HADS] Anxiety Subscale). Greater depressive symptoms were associated with lower RSA ($\beta = -0.06$, $P = 0.03$ for HADS Depression Subscale; $\beta = -0.03$, $P = 0.04$ for Beck Depression Inventory).

Conclusions: Among peri- and postmenopausal women with hot flashes, greater self-reported anxiety and depressive symptoms were associated with lower levels of resting cardiac parasympathetic activity, and greater state anxiety was associated with higher levels of cardiac sympathetic activity. Findings suggest that midlife women with increased anxiety and depressive symptoms may have an unfavorable cardiac autonomic profile with potential implications for their overall cardiovascular risk.

Key Words: Anxiety – Autonomic function – Depression – Hot flashes – Menopause.

Menopausal transition is marked by an increased prevalence of mood symptoms in midlife women, including anxiety and depression.[1] Anxiety and depression are in turn associated with vasomotor symptoms such as hot flashes, the most common menopause-related complaint of peri- and postmenopausal women in western countries.[2,3] Despite their increased prevalence during midlife, relatively little is known about the broader clinical implications of anxiety and depressive symptoms during the menopausal transition. Although women may experience transient increases in anxiety or depression in midlife that have no other clinical consequences, concerns have been raised that increased mood symptoms in midlife may be associated with an increased risk of other adverse health outcomes.[4]

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Prior research in men and nonmenopausal women has suggested that both anxiety and depression may be associated with alterations in cardiac autonomic function,[5,6] particularly parasympathetic function as measured by heart rate variability (HRV). Increased sympathetic and decreased parasympathetic activation have, in turn, been associated with adverse cardiovascular outcomes and mortality in patients with cardiovascular disease, such as coronary artery disease and chronic heart failure,[7-9] as well as higher rates of chronic diseases that increase incidence of cardiovascular disease, such as obesity, diabetes, and hypertension.[10,11]

Menopause has been shown to be a critical period with an increased risk for accumulation of cardiovascular risk factors.[12] In particular, a growing literature has linked hot flashes to cardiovascular disease,[13,14] suggesting that hot flashes may be a marker of underlying cardiovascular disease risk, potentially through the decrease in cardiac vagal control.[15] To further elucidate these relationships among women in the menopausal transition, we examined the strength and direction of associations between multiple measures of anxiety and depressive symptoms with cardiac markers of resting sympathetic and parasympathetic activation among peri- and postmenopausal women with hot flashes enrolled in a behavioral relaxation intervention trial. Our goal was to assess whether increased anxiety or depressive symptoms among midlife women with hot flashes may be associated with an unfavorable cardiac autonomic profile, which could, in turn, have potential implications for women's overall cardiovascular risk.

METHODS

Study population and intervention

The Menopausal Treatment Using Relaxation Exercise (MaTURE) trial was a parallel-group, single-blinded, randomized trial of slow-paced respiration for treatment of hot flashes in peri- and postmenopausal women funded by the National Institutes of Health. Details of the eligibility criteria, recruitment procedures, and study design have previously been described.[16] Briefly, participants were recruited from the San Francisco Bay Area between January 2012 and December 2014. Inclusion criteria included being 40 to 59 years old, being peri- or postmenopausal, and reporting at least four hot flashes per day in a validated 7-day hot flash diary.[17] Exclusion criteria included being pregnant or breastfeeding in the past year and using medications with known effects on hot flashes in the previous 3 months, including estrogens and selective serotonin/norepinephrine reuptake inhibitors (SSRIs/SNRIs).

Participants were randomly assigned to either the paced respiration ($N = 62$) or music-listening intervention ($N = 59$) by computer algorithm. They were given either a commercially available, portable-guided breathing device (ReSPERATE; Intercure, Ltd.) to practice slowing their resting breathing rate to less than 10 breaths per minute (paced respiration) or an identical-appearing device reprogrammed to play relaxing music (for the music-listening control). Participants in both groups were instructed to use their assigned portable devices for a minimum of 15 minutes per day for 12 weeks. All participants provided informed consent before randomization, and all procedures were approved by the institutional review board of the University of California, San Francisco.

Anxiety and depression symptom questionnaires

Anxiety and depressive symptoms were assessed using multiple validated self-report questionnaires at baseline and 12 weeks. State anxiety (ie, fluctuating, transitory emotional state to perceived threats) and trait anxiety (ie, stable individual tendency toward perceived threats) were measured using the Spielberger State Trait Anxiety Inventory (STAI),[18] a 20-item questionnaire validated in both psychiatric and clinical populations. Cognitive anxiety (ie, mental component of anxiety associated with fear of future adverse events) was assessed using the 7-item anxiety subscale of the Hospital Anxiety and Depression Scale (HADS).[19] Depressive symptoms were assessed using the 21-item Beck Depression Inventory-II (BDI-II)[20] and the 7-item depression subscale of HADS. In addition, perceived stress (ie, degree to which situations in one's life are appraised as stressful) was measured as another construct closely associated with both anxiety and depression, using the 10-item Perceived Stress Scale (PSS).[21]

Cardiac autonomic function

We assessed participants' cardiac sympathetic and parasympathetic nervous function using impedance cardiography and electrocardiography (ECG), which provided measures of pre-ejection period (PEP) and respiratory sinus arrhythmia (RSA) during a resting period at baseline (before intervention assignment) and after 12 weeks. PEP, the time period from the start of cardiac ventricular depolarization to the opening of the aortic valve, provides a measure of ventricle contractility that has been shown to provide a relatively pure measure of sympathetic activity without parasympathetic influence.[22] Increases in cardiac sympathetic nervous system activity correspond to shortening PEP.[23] RSA, the variability of the heart rate during the typical respiratory cycle, provides an indication of the amount of influence of the cardiac vagus nerve and is an established marker of parasympathetic activity.[22,24] Higher RSA corresponds to greater cardiac parasympathetic nervous system activity.

Details on the protocol and procedure with regard to collection of these resting cardiac autonomic functions have been previously described.[25] Briefly, participants were outfitted with electrodes by trained study staff, and then asked to sit still, relax, and view a neutral nature documentary video for a 10-minute period while resting measurements were obtained. Impedance cardiography measurements were obtained using a band electrode system that completely encircled the neck and chest areas. Electrocardiography measurements were obtained using electrodes in a lead II configuration. All signals were sampled at 1,000 Hz and stored using a Biopac MP150 data acquisition system (Biopac Systems, Inc.). Trained research assistants visually inspected

TABLE 1. *Anxiety and depression symptoms and cardiac autonomic function by intervention group at baseline and 12 weeks*

	Baseline		12 wk	
	Paced respiration Mean (SD)	Music control Mean (SD)	Paced respiration Mean (SD)	Music control Mean (SD)
Anxiety Questionnaire Scores				
STAI State Anxiety (20-80)	34.6 (12.3)	34.9 (12.4)	31.7 (12.1)	31.1 (11.6)
STAI Trait Anxiety (20-80)	39.2 (11.6)	38.9 (11.7)	34.6 (11.5)	35.3 (11.4)
HADS Anxiety Subscale (0-21)	7.6 (4.1)	7.3 (4.4)	6.9 (4.5)	5.9 (4.1)
Depression Questionnaire Scores				
Beck Depression Inventory (0-63)	11.0 (8.0)	9.7 (7.5)	7.7 (8.8)	6.2 (5.8)
HADS Depression Subscale (0-21)	4.6 (3.9)	4.3 (3.7)	3.7 (3.6)	3.6 (3.6)
Perceived Stress Questionnaire Score				
Perceived Stress Scale (0-40)	16.8 (6.6)	16.3 (8.2)	14.5 (7.4)	14.0 (6.9)
Cardiac Autonomic Function				
PEP at rest (ms)	121.7 (13.4)	121.3 (13.5)	121.3 (13.8)	122.8 (13.4)
RSA at rest (ms^2)	5.2 (1.3)	5.4 (1.3)	5.6 (1.3)	5.3 (1.2)

HADS, Hospital Anxiety and Depression Scale; PEP, pre-ejection period; RSA, respiratory sinus arrhythmia; STAI, Spielberger State Trait Anxiety Inventory.

all waveforms off-line, then edited and scored the data in 1-minute bins for each of the minutes of the experiment using Mindware software (HRV 3.0, IMP 3.0). Average PEP (in milliseconds) was calculated by examining the time between ventricular depolarization (assessed by electrocardiogram output) and the B point of the dZ/dt wave indicating the opening of the aortic valve (measured by impedance cardiography) using Mindware software (Mindware Technologies, Ltd.).[26,27] RSA was estimated with an algorithm that uses a 4 Hz time series to interpolate the interbeat interval,[28] and a second-order polynomial was applied to minimize nonstationary trends.

Statistical analysis

Baseline demographic and clinical characteristics of participants who provided cardiac autonomic data were examined using descriptive statistics. To assess for between group differences, χ^2 test was used for categorical variables and Kruskal–Wallis one-way ANOVA for continuous variables. Multivariable models using linear regression with repeated measures were then developed to assess relationships between anxiety, depression symptoms, and perceived stress (modeled as continuous questionnaire scores) with autonomic function outcomes (modeled as continuous PEP and RSA parameters). As no significant between-group differences in the above variables were detected at baseline or 12 weeks,[16,25] data from both groups were combined in these multivariable analyses. The Benjamini–Hochberg procedure[29,30] was used to correct for multiple comparisons. For our main models, Benjamini–Hochberg adjustments were made in each predictor model to account for testing of associations with multiple autonomic outcomes (ie, PEP and RSA). In additional sensitivity analyses, Benjamini–Hochberg adjustments were made separately for each autonomic outcome to account for multiple predictors (ie, anxiety, depression, and stress questionnaires). All models were adjusted for age and body mass index (BMI) measured at baseline, given past evidence that both of these variables are associated with resting cardiac autonomic function, hot flashes, anxiety, and depression.[31-35] All statistical analyses were performed using SAS 9.4 (Cary, NC).

RESULTS

Of the 123 randomized participants, 121 provided data on anxiety symptoms, depressive symptoms, and perceived stress as well as cardiac autonomic function at either baseline or 12 weeks. Details on baseline demographic and clinic characteristics of participants have been previously published.[16,25] Briefly, the average age was 53 years (SD 3.4), 17% were racial or ethnic minorities, and most women were naturally peri- or postmenopausal (see Table, Supplemental Digital Content 1, http://links.lww.com/MENO/A322, which presents the baseline characteristics for the total sample and by intervention group). Average BMI was 26.5 (SD 5.5) with less than 20% of women taking medications for hypertension, hyperlipidemia, diabetes, or thyroid disease. Participants experienced a high hot flash burden at baseline, with a mean of 8 hot flashes per day and 5 moderate-to-severe hot flashes per day.

No significant differences in the mean scores on questionnaire measures evaluating anxiety, depressive symptoms, or perceived stress were detected between the paced respiration and the music control groups at baseline (Table 1). On average, questionnaire scores were within the normal ranges (ie, below the commonly used thresholds of 39-40 for STAI-State[36]; 45 for STAI-Trait; 14 for BDI-II[37]; and 8 for HADS that are used to detect clinically significant anxiety or depression). For PSS, no specific cutoff score available but norm for women is mean 13.7 (SD 6.6).[38] There were also no significant between-group differences in the resting cardiac autonomic function, as measured by PEP or RSA, at baseline. Cardiac autonomic parameters were similar at baseline and at 12 weeks. Compared with the general US population, on average, participants' PEP was within normal range (117.0; SD 10.5)[27] and RSA was slightly lower (6.8; SD 1.2) though RSA differed across age, sex, and race.

In our main analyses combining data from both intervention groups, using repeated observations from baseline and 12 weeks, with statistical correction for assessment of two types of autonomic outcomes, and adjusting for age and BMI, we found that greater state anxiety (ie, fluctuating, transitory

TABLE 2. *Multivariable-adjusted associations between anxiety or depression symptoms and cardiac autonomic function*

	Pre-ejection period (PEP)		Respiratory sinus arrhythmia (RSA)	
	β (SE)	P^a	β (SE)	P^a
Anxiety Questionnaire Scores				
STAI State Anxiety	−0.24 (0.09)	0.02	−0.16 (0.01)	0.06
STAI Trait Anxiety	−0.11 (0.10)	0.24	−0.03 (0.01)	0.006
HADS Anxiety Subscale	−0.31 (0.27)	0.25	−0.06 (0.02)	0.02
Depression Questionnaire Scores				
Beck Depression Inventory	−0.09 (0.17)	0.60	−0.03 (0.01)	0.04
HADS Depression Subscale	−0.40 (0.36)	0.26	−0.07 (0.03)	0.03
Perceived Stress Questionnaire Score				
Perceived Stress Scale	−0.23 (0.15)	0.14	−0.03 (0.01)	0.06

HADS, Hospital Anxiety and Depression Scale; STAI, Spielberger State Trait Anxiety Inventory.
[a]Beta-coefficients and *P* values were derived from repeated measure multivariable linear regression models using data from both baseline and 12 weeks, adjusting for age and body mass index, and using the Benjamini–Hochberg procedure to correct for testing for two different autonomic outcomes (PEP and RSA).

emotional state to perceived threats) as measured by STAI-State score was associated with shorter PEP, reflecting higher sympathetic activity (Table 2). No other significant associations between other anxiety measures and PEP were, however, detected. Similarly, no significant associations between depression measures and PEP were observed.

Also in our main analyses, greater trait anxiety (ie, stable individual tendency toward perceived threats) as measured by STAI-Trait score and cognitive anxiety (ie, mental component of anxiety associated with fear of future adverse events) as measured by HADS Anxiety Subscale were associated with lower RSA, reflecting decreased parasympathetic activity (Table 2). Depressive symptoms as measured by HADS Depression Subscale and BDI-II were also associated with lower RSA. Although there was a trend toward lower RSA with greater state anxiety as measured by STAI-State score and perceived stress as measure by PSS, these associations did not reach statistical significance.

In additional sensitivity analyses that corrected for assessment of multiple anxiety/depression measures despite overlap in the content of these measures, greater trait anxiety continued to be associated with lower RSA (see Table, Supplemental Digital Content 2, http://links.lww.com/MENO/A322); however, no other statistically significant associations were seen in these additional models, although there were nonsignificant trends toward lower RSA with higher scores on other depression and anxiety measures.

DISCUSSION

We examined relationships between anxiety and depression symptoms and cardiac autonomic function in peri- and postmenopausal women with hot flashes, a group at increased risk for the development of cardiovascular diseases. In this sample, greater self-reported anxiety and depressive symptoms as measured by multiple validated questionnaires were associated with decreased cardiac parasympathetic activation as reflected by lower RSA. In addition, greater state anxiety was associated with increased resting sympathetic nervous system activation as reflected by lower PEP, although no significant associations between other measures of anxiety or depression and sympathetic activation were detected.

Our findings are consistent with prior studies that included men and nonmenopausal women, indicating that those with clinical anxiety or depressive disorders may be associated with low resting parasympathetic activity.[39,40] Specifically, in several past studies, RSA was shown to be lower in patients with clinical anxiety disorders and major depression disorder compared with healthy controls.[41,42] From the cardiovascular risk standpoint, lower RSA has also been strongly linked to hypertension, hyperlipidemia, hyperglycemia, and obesity[10,11]; and in patients with known cardiac disease, lower RSA was a strong predictor of sudden cardiac death after myocardial infarction.[43] Taken as a whole, these findings suggest that increased psychological symptom burden may be associated with a more unfavorable cardiac autonomic profile and higher cardiovascular risk.

With regard to sympathetic nervous system activation, several studies in nonmenopausal samples have reported that depression may be associated with attenuated PEP reactivity to stressful stimuli.[44,45] In the present study, PEP was assessed only at rest rather than in response to an evoked stressful stimulus, which may have limited our ability to assess for meaningful relationships. Nevertheless, we did observe an association between high state anxiety and low resting PEP, although no associations between other types of anxiety or depressive symptoms with resting PEP were detected. Among peri- and postmenopausal women, state anxiety may be a better reflection of transient mood fluctuations during the menopausal transition as compared with trait anxiety, which has been described as a more general tendency and stable characteristic by Spielberger.[18]

Our study had several strengths, including an ethnically diverse group of women in various stages of the menopausal transition, multiple well-validated and sensitive measures of anxiety, depressive symptoms, and perceived stress, and specific cardiac markers of sympathetic and parasympathetic function that were administered in a rigorously controlled setting with a high retention rate. Several limitations of this study should, however, be mentioned.

First, all participants were women practicing either paced respirations or music listening for frequent hot flashes, and these findings may not be generalizable to women in noninterventional settings or those without hot flashes. Second, our study did not collect information about psychological symptoms earlier in life; thus, it is unclear if anxiety or depressive symptoms and perceived stress reported during the 12-week trial were reflective of long-standing psychological functioning or changes related to the menopausal transition. In addition, psychological symptom scores on average were within normal ranges, and these findings may not be generalizable to women with clinically significant mood disorders. Third, given that we performed multiple comparisons, it is possible that some associations were significant by chance. For example, the association between state anxiety and lower PEP may be a chance association, given that no other significant associations between other anxiety measures and PEP were detected. Based on the stronger pattern of association between psychological measures and lower RSA even after correction for multiple outcomes with the Benjamini–Hochberg procedure, it is, however, less likely that all associations with RSA were due to chance alone. In addition, our findings are consistent with prior studies in men and nonmenopausal women showing association between anxiety and depression with lower resting parasympathetic activity.[39,40]

CONCLUSIONS

In summary, among peri- and postmenopausal women with hot flashes enrolled in a behavioral paced respiration intervention trial, women with a higher burden of anxiety and depression symptoms demonstrated lower cardiac vagal tone. Given the literature supporting associations between reduced vagal tone and cardiovascular disease, these findings suggest that women with increased anxiety and depressive symptoms in midlife may have an unfavorable cardiac autonomic profile that could, in turn, have potential implications for their overall cardiovascular risk. Our findings may provide support for future long-term studies examining the impact of psychological functioning in midlife women's cardiovascular health and raise the possibility that therapeutic interventions targeting control of anxiety and depression may be helpful in decreasing cardiovascular risk.

REFERENCES

1. Bromberger JT, Kravitz HM. Mood and menopause: findings from the Study of Women's Health Across the Nation (SWAN) over 10 years. *Obstet Gynecol Clin North Am* 2011;38:609-625.
2. Gold EB, Colvin A, Avis N, et al. Longitudinal analysis of the association between vasomotor symptoms and race/ethnicity across the menopausal transition: study of women's health across the nation. *Am J Public Health* 2006;96:1226-1235.
3. Juang KD, Wang SJ, Lu SR, Lee SJ, Fuh JL. Hot flashes are associated with psychological symptoms of anxiety and depression in peri- and postbut not premenopausal women. *Maturitas* 2005;52:119-126.
4. Muka T, Oliver-Williams C, Colpani V, et al. Association of vasomotor and other menopausal symptoms with risk of cardiovascular disease: a systematic review and meta-analysis. *PLoS One* 2016;11:e0157417.
5. Friedman BH. An autonomic flexibility-neurovisceral integration model of anxiety and cardiac vagal tone. *Biol Psychol* 2007;74:185-199.
6. Rottenberg J. Cardiac vagal control in depression: a critical analysis. *Biol Psychol* 2007;74:200-211.
7. Nolan J, Batin PD, Andrews R, et al. Prospective study of heart rate variability and mortality in chronic heart failure: results of the United Kingdom heart failure evaluation and assessment of risk trial (UK-heart). *Circulation* 1998;98:1510-1516.
8. Liao D, Carnethon M, Evans GW, Cascio WE, Heiss G. Lower heart rate variability is associated with the development of coronary heart disease in individuals with diabetes: the atherosclerosis risk in communities (ARIC) study. *Diabetes* 2002;51:3524-3531.
9. Thayer JF, Lane RD. The role of vagal function in the risk for cardiovascular disease and mortality. *Biol Psychol* 2007;74:224-242.
10. Licht CM, Vreeburg SA, van Reedt Dortland AK, et al. Increased sympathetic and decreased parasympathetic activity rather than changes in hypothalamic-pituitary-adrenal axis activity is associated with metabolic abnormalities. *J Clin Endocrinol Metab* 2010;95:2458-2466.
11. Masi CM, Hawkley LC, Rickett EM, Cacioppo JT. Respiratory sinus arrhythmia and diseases of aging: obesity, diabetes mellitus, and hypertension. *Biol Psychol* 2007;74:212-223.
12. Cagnacci A, Cannoletta M, Palma F, Zanin R, Xholli A, Volpe A. Menopausal symptoms and risk factors for cardiovascular disease in postmenopause. *Climacteric* 2012;15:157-162.
13. Thurston RC, Sutton-Tyrrell K, Everson-Rose SA, Hess R, Matthews KA. Hot flashes and subclinical cardiovascular disease: findings from the Study of Women's Health Across the Nation Heart Study. *Circulation* 2008;118:1234-1240.
14. Thurston RC, Chang Y, Barinas-Mitchell E, et al. Menopausal hot flashes and carotid intima media thickness among midlife women. *Stroke* 2016;47:2910-2915.
15. Thurston RC, Christie IC, Matthews KA. Hot flashes and cardiac vagal control: a link to cardiovascular risk? *Menopause* 2010;17:456-461.
16. Huang AJ, Phillips S, Schembri M, Vittinghoff E, Grady D. Device-guided slow-paced respiration for menopausal hot flushes: a randomized controlled trial. *Obstet Gynecol* 2015;125:1130-1138.
17. Sloan JA, Loprinzi CL, Novotny PJ, Barton DL, Lavasseur BI, Windschitl H. Methodologic lessons learned from hot flash studies. *J Clin Oncol* 2001;19:4280-4290.
18. Spielberger CD. *Manual for the State-Trait Anxiety Inventory.*. Palo Alto, CA: Consulting Psychologists Press; 1983.
19. Zigmond AS, Snaith RP. The hospital anxiety and depression scale. *Acta Psychiatr Scand* 1983;67:361-370.
20. Beck AT, Ward CH, Mendelson M, Mock J, Erbaugh J. An inventory for measuring depression. *Arch Gen Psychiatry* 1961;4:561-571.
21. Cohen S, Kamarck T, Mermelstein R. A global measure of perceived stress. *J Health Soc Behav* 1983;24:385-396.
22. Berntson GG, Cacioppo JT, Binkley PF, Uchino BN, Quigley KS, Fieldstone A. Autonomic cardiac control. III. Psychological stress and cardiac response in autonomic space as revealed by pharmacological blockades. *Psychophysiology* 1994;31:599-608.
23. Sherwood A, Allen MT, Obrist PA, Langer AW. Evaluation of beta-adrenergic influences on cardiovascular and metabolic adjustments to physical and psychological stress. *Psychophysiology* 1986;23:89-104.
24. Berntson GG, Bigger JT Jr, Eckberg DL, et al. Heart rate variability: origins, methods, and interpretive caveats. *Psychophysiology* 1997;34:623-648.
25. Gibson CJ, Mendes WB, Schembri M, Grady D, Huang AJ. Cardiac autonomic function and hot flashes among perimenopausal and postmenopausal women. *Menopause* 2017;24:756-761.
26. Mendes WB. Assessing autonomic nervous system activity. In: Harmon-Jones E, Beer JS, editors. *Methods in Social Neuroscience*. New York, NY: The Guilford Press; 2009. pp. 118-147.
27. Blascovich J, Vanman EJ, Mendes WB, Dickerson S. *Social Psychophysiology for Social and Personality Psychology*. Los Angeles, CA: SAGE; 2011.
28. Berntson GG, Cacioppo JT, Quigley KS. Respiratory sinus arrhythmia: autonomic origins, physiological mechanisms, and psychophysiological implications. *Psychophysiology* 1993;30:183-196.
29. Benjamini Y, Hochberg Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J R Stat Soc* 1995;57:2035-2041.
30. Huque MF. Validity of the Hochberg procedure revisited for clinical trial applications. *Stat Med* 2016;35:5-20.

31. Thurston RC, Chang Y, Mancuso P, Matthews KA. Adipokines, adiposity, and vasomotor symptoms during the menopause transition: findings from the Study of Women's Health Across the Nation. *Fertil Steril* 2013;100:793-800.
32. Karason K, Molgaard H, Wikstrand J, Sjostrom L. Heart rate variability in obesity and the effect of weight loss. *Am J Cardiol* 1999;83:1242-1247.
33. De Meersman RE, Stein PK. Vagal modulation and aging. *Biol Psychol* 2007;74:165-173.
34. Vink D, Aartsen MJ, Comijs HC, et al. Onset of anxiety and depression in the aging population: comparison of risk factors in a 9-year prospective study. *Am J Geriatr Psychiatry* 2009;17:642-652.
35. Lykouras L, Michopoulos J. Anxiety disorders and obesity. *Psychiatriki* 2011;22:307-313.
36. Knight RG, Waal-Manning HJ, Spears GF. Some norms and reliability data for the State–Trait Anxiety Inventory and the Zung Self-Rating Depression scale. *Br J Clin Psychol* 1983;22 (Pt 4):245-249.
37. Smarr KL, Keefer AL. Measures of depression and depressive symptoms: Beck Depression Inventory-II (BDI-II), Center for Epidemiologic Studies Depression Scale (CES-D), Geriatric Depression Scale (GDS), Hospital Anxiety and Depression Scale (HADS), and Patient Health Questionnaire-9 (PHQ-9). *Arthritis Care Res (Hoboken)* 2011;63 (Suppl. 11): S454-S466.
38. Cohen S, Williamson G. Perceived stress in a probability sample of the United States. In: Spacapan S, Oskamp S, editors. *The Social Psychology of Health*. Newbury Park, CA: Sage; 1988.
39. Gorman JM, Sloan RP. Heart rate variability in depressive and anxiety disorders. *Am Heart J* 2000;140 (4 Suppl.):77-83.
40. Dishman RK, Nakamura Y, Garcia ME, Thompson RW, Dunn AL, Blair SN. Heart rate variability, trait anxiety, and perceived stress among physically fit men and women. *Int J Psychophysiol* 2000; 37:121-133.
41. Licht CM, de Geus EJ, van Dyck R, Penninx BW. Association between anxiety disorders and heart rate variability in The Netherlands Study of Depression and Anxiety (NESDA). *Psychosom Med* 2009;71:508-518.
42. Licht CM, de Geus EJ, Zitman FG, Hoogendijk WJ, van Dyck R, Penninx BW. Association between major depressive disorder and heart rate variability in the Netherlands Study of Depression and Anxiety (NESDA). *Arch Gen Psychiatry* 2008;65:1358-1367.
43. Peltola M, Tulppo MP, Kiviniemi A, et al. Respiratory sinus arrhythmia as a predictor of sudden cardiac death after myocardial infarction. *Ann Med* 2008;40:376-382.
44. Salomon K, Bylsma LM, White KE, Panaite V, Rottenberg J. Is blunted cardiovascular reactivity in depression mood-state dependent? A comparison of major depressive disorder remitted depression and healthy controls. *Int J Psychophysiol* 2013;90:50-57.
45. Hu MX, Lamers F, de Geus EJ, Penninx BW. Differential autonomic nervous system reactivity in depression and anxiety during stress depending on type of stressor. *Psychosom Med* 2016;78: 562-572.

