RESEARCH ARTICLE

Cortisol Level Modulated by Integrative Meditation in a Dose-dependent Fashion

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Abstract

Prior research has shown that an additional training session immediately after acute stress decreases release of salivary cortisol in a college student group trained with 5-day integrative body–mind training (IBMT) in comparison with a control group given the same amount of relaxation training. However, 5 days of training does not influence the basal secretion of cortisol. The current study seeks to extend this finding and determine whether increasing amounts of IBMT will decrease the basal cortisol level, suggesting reduced stress to daily activities. Thirty-four Chinese undergraduates were randomly assigned either to 4 weeks of IBMT or a relaxation control. Salivary cortisol levels at baseline before training and the three stages of a stress intervention test (i.e. rest, stress and additional 20-min practice) after 2 and 4 weeks of training were assessed. The basal cortisol level decreased significantly in the IBMT but not in relaxation group after 2 and 4 weeks of training. An additional IBMT practice session immediately after acute stress produced significantly lower cortisol release for the IBMT group in comparison with relaxation at weeks 2 and 4. The results indicate that IBMT produces a change in the basal endocrine system and larger acute effects as the dose of training increases. Copyright © 2013 John Wiley & Sons, Ltd.

Received 1 January 2013; Revised 25 April 2013; Accepted 27 April 2013

Keywords

integrative body–mind training (IBMT); relaxation training (RT); stress interventions; cortisol; HPA axis

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Published online in Wiley Online Library (wileyonlinelibrary.com) DOI: 10.1002/smi.2497

Introduction

It is now widely accepted that psychological stress increases the activity of the hypothalamus–pituitary–adrenal (HPA) axis with subsequent rise in cortisol level (Dickerson & Kemeny, 2004; Roy, 2004). Although neuroendocrine stress responses are essential for the maintenance of homeostasis, exaggerated secretion of cortisol can suppress aspects of immune function and have negative effects on health, particularly when acute responses to stress become chronic (Glaser, 2005; McEwen, 1998).

A number of studies have demonstrated the effectiveness of stress reduction interventions in influencing psychological and physiological parameters and health outcomes in health and disease (Carlson, Speca, Faris & Patel, 2007; Pace et al., 2009). Given that cortisol is a hormone secreted in response to stress, researchers have chosen to evaluate the potential role of this hormonal mediator as an objective physiological marker for improvement in stress management (Hammerfald et al., 2006; MacLean et al., 1997; Matousek, Dobkin & Pruessner, 2010; Pawlow & Jones, 2005; Scholey et al., 2009). The assessment of cortisol in saliva has been proved to be a valid and reliable reflection of the unbound hormone in blood (Aardal-Eriksson, Karlberg & Holm, 1998). Because of several advantages over blood cortisol analyses (e.g. stress-free sampling, laboratory independence and non-invasive collection procedure), salivary cortisol is a more appropriate measure for the assessment of adrenocortical function than serum cortisol (Gozansky, Lynn, Laudenslager & Kohrt, 2005).

There is accumulating evidence indicating that cortisol levels decrease following participation in stress reduction programs, such as yoga (West, Otte, Geher, Johnson & Mohr, 2004), Qi training (Lee, Kang, Ryu & Moon, 2004), progressive relaxation training (RT; Pawlow & Jones, 2005), mindfulness-based stress reduction (MBSR) (Carlson et al., 2007) and cognitive-behavioural stress management (Gaab et al., 2003). However, not all researchers have reported beneficial effects of participation in these programs on the HPA axis. Galantino, Baime, Maguire, Szapary, and Farrar (2005) found no significant changes in salivary cortisol levels from pre-intervention to post-intervention in 42 healthcare professionals who completed a MBSR program. Similarly, several studies have failed to find group differences in basal cortisol levels between controls and participants following MBSR program (Klatt, Buckworth & Malarkey, 2009; Robert McComb, Tacon, Randolph, & Caldera, 2004; Robinson, Mathews, & Witek-Janusek, 2003). Plausible explanations for the lack of findings regarding basal cortisol levels may include lack of control over confounding variables, such as diet and physical activity prior to cortisol sampling (Klatt et al., 2009) and small sample sizes, with nine women per group in Robert McComb et al. (2004).

Integrative body–mind training (IBMT) or integrative meditation integrates several key components of body–mind techniques including body relaxation, mental imagery and mindfulness training, which can help accelerate practitioner access to meditative states (Tang & Posner, 2009; Tang, Rothbart & Posner, 2012; Tang et al., 2007, 2009). Our previous study had shown that short-term IBMT practice could improve self-regulation ability and control of stress (Tang et al., 2007). An additional training session immediately after acute stress induced decreased release of salivary cortisol in a group trained with 5 days IBMT in comparison with a control group given the same amount of RT. However, 5 days' training did not influence the basal cortisol level. The current study set out to extend our previous finding. We hypothesized that increasing amounts of integrative meditation training will decrease the basal cortisol level and produce further improvements in adrenocortical function in response to stress.

Materials and methods

Subjects

Subjects were recruited for a study of stress management through a notice on the bulletin board of Dalian University of Technology. The notice briefly described the study. Interested subjects had the opportunity to apply. They then filled out a questionnaire, containing exclusion criteria designed to reduce confounding factors that have been shown to affect physiological dependent measures. Subjects were excluded if they reported having a past or present history of psychiatric, immune, metabolic or endocrine disease, such as depression, anxiety, psychosis, allergies or autoimmune disease, diabetes and acquired immunodeficiency syndrome. Thirty-four healthy Chinese undergraduates [16 men and 18 women, mean age ($\pm$standard deviation) = 20.87 ± 0.26 years] without any previous training experiences participated in this study. The human experiment was approved by the local Institutional Review Board, and informed consent was obtained from each participant.

Subjects were randomly assigned to an IBMT group and an RT group. Seventeen experimental subjects (eight men and nine women) continuously attended practice of IBMT for 4 weeks, whereas 17 control subjects (eight men and nine women) received an equal period of RT.

Training methods

Integrative body–mind training involves several body–mind techniques including body relaxation, mental imagery and mindfulness training, accompanied with selected background music (Tang et al., 2007). The method emphasizes no effort to control thoughts and achievement of a state of restful alertness that allows a high degree of awareness of body, breathing and external instructions (Tang & Posner, 2009; Tang et al., 2007, 2009, 2012). It includes a pre-session, a practice session and a post-session. RT is a form of a muscle relaxation technique very popular in the west (Bernstein & Borkovec, 1973). RT involves the relaxing of different muscle groups over the face, head, shoulders, arms, legs, chest, back and abdomen, guided by a tutor and a compact disk (CD). With eyes closed and in a sequential pattern, one is forced to concentrate on the sensation of relaxation, such as the feelings of warmth and heaviness. This progressive training helps the participant achieve physical and mental relaxation and calmness.

Experimental procedure

Before training, the qualified IBMT and RT coaches separately gathered the subjects to introduce the structure of the program and the training method, answer questions and also set up the exact time, training room and discipline for the group practice. Saliva was collected to obtain baseline samples.

All subjects completed group-training sessions for each of the four successive weeks (Monday to Friday). Subjects separately followed the IBMT-CD and RT-CD guided by the coaches in different rooms every night for 20–30 min. Subjects sit in a chair and closed their eyes while listening to the CD in a dark room. Following each training session, every subject filled out a self-report questionnaire and evaluated the practice. The coaches gave brief and immediate responses to questions from the participants as required. The qualified coach has completed IBMT or RT training and passed associated certification tests, demonstrating the ability to lead the group training.

All subjects performed a stress intervention test after 2 and 4 weeks of training. The stress intervention test included three stages (i.e. rest, stress and additional 20-min practice). After a 5-min rest, subjects performed 3 min of mental arithmetic (described in the succeeding texts) to induce stress. Then the experimental subjects practised additional IBMT for 20-min RT, whereas the controls practised additional 20-min RT.

Mental arithmetic task

Mental arithmetic was used as an acute laboratory stressor (Tang et al., 2007). Subjects read the introduction writing on a computer screen and performed serial subtraction of 47 from a four-digit number. During the

3 min of mental arithmetic task, participants were prompted to respond verbally as fast and accurate as possible. The investigator checked the correct answers printed in a paper. If the participants did not give the correct answer in 5 s, the computer produced a harsh sound to remind the subjects, who were required to restart the task and do it again.

Sampling methods and biochemical analysis

Before training, saliva samples were collected to obtain baseline data. After 2 and 4 weeks of training, saliva samples were collected repeatedly before the stress, immediately after the stress and immediately after additional 20-min practice. To control for variations of cortisol levels over the circadian rhythm, saliva sample collection was performed from 14.00 to 18.00 hours (Hucklebridge, Clow & Evans, 1998).

Subjects were required to abstain from smoking and alcohol on the day of the test and to not eat a large meal or engage in physical activity for 4 h prior to the test. They were also required to rinse their mouths before collecting samples. Subjects collected about 1 mL of saliva samples by using the disposable syringe by themselves. The investigator put the saliva into the test tubes, labelled and placed the test tubes into a refrigerator under −20°C and then thawed 24 h later for analysis. The concentration of cortisol was analysed by radioimmunoassay at the Dalian Medical University. Intra-assay and inter-assay coefficients of variation were below 10%. To reduce error variance caused by imprecision of the intra-assay, all samples of each subject were analysed in the same run.

Statistical analyses

All statistical analyses were performed by using the SPSS 13.0 for Windows. Data were analysed using the repeated measures analyses of variance (ANOVAs). Pairwise comparisons were performed for each cortisol sample.

Results

Basal (resting) measures

Means and standard deviations of basal (resting) salivary cortisol levels before training and after 2 and 4 weeks of training are shown in Table I.

Since assignment was random, the IBMT group and RT group should not differ prior to training. As expected, there was no significant difference between the two groups in pre-training cortisol level [$F(1, 32) = 0.006$, $p > 0.05$].

After 2 weeks of training, the basal (resting) cortisol level decreased significantly in comparison with pre-training in the IBMT subjects but not in controls [$t(16) = 2.684$, $p = 0.016$]. However, there was no significant difference between the two groups in the basal cortisol level at week 2 ($F(1, 32) = 0.942$, $p > 0.05$).

Table I. The basal salivary cortisol levels (nmol/L) before training and after 2 and 4 weeks of training

	Pre-training	Week 2	Week 4
IBMT	16.48 ± 0.99	$14.75 \pm 1.02^{\dagger}$	$13.07 \pm 0.67^{\dagger\dagger}$
RT	16.37 ± 1.10	16.15 ± 1.03	17.42 ± 1.56

IBMT: integrative body–mind training; RT: relaxation training.
Values are mean ± SE.
*$p < 0.05$ IBMT group versus RT group;
$^{\dagger}p < 0.05$; $^{\dagger\dagger}p < 0.01$ week 4 or week 2 versus pre-training.

After 4 weeks of training, the basal (resting) cortisol level also significantly decreased in comparison with pre-training in the IBMT subjects but not in controls [$t(16) = 3.075$, $p = 0.007$]. Moreover, the basal cortisol level of the IBMT group at week 4 was significantly lower than that of the RT group [$F(1, 32) = 6.539$, $p = 0.015$]. There was no significant difference in the basal cortisol level between week 4 and week 2 for both groups ($p > 0.05$).

Stress response measures

Repeated measures ANOVAs

For the cortisol levels response to a stress intervention test after 2 and 4 weeks of training, repeated measures ANOVAs were respectively conducted with the between-group factor of Group (IBMT and RT) and the within-subjects factor of Condition (rest, stress and additional 20-min practice). For week 2, the analyses revealed significant main effects for Group [$F(1, 32) = 6.484$, $p < 0.05$] and Condition [$F(2, 64) = 24.274$, $p < 0.001$] as well as a significant interaction for Group × Condition [$F(2, 64) = 4.146$, $p < 0.05$]. For week 4, ANOVA showed significant main effects for Group [$F(1, 32) = 11.057$, $p < 0.01$] and Condition [$F(2, 64) = 23.038$, $p < 0.001$] as well as a significant interaction for Group × Condition [$F(2, 64) = 5.687$, $p < 0.05$].

Post-training cortisol measures (week 2)

Salivary cortisol levels at the three stages of a stress intervention test after 2 weeks of training are shown in Figure 1.

Following the acute mental arithmetic challenge, the cortisol level significantly increased compared with the resting level for both groups. The cortisol level immediately after stress of the IBMT group was significantly lower than that of the RT group ($p < 0.05$).

After stress, the IBMT group received an additional 20-min practice, and the control group relaxed for 20-min practice. In comparison with the level immediately after stress, the cortisol level following the additional practice significantly increased in the control group [$t(16) = -3.873$, $p < 0.01$]. However, no significant change was found in the IBMT group ($p > 0.05$). There was a significant difference between the two

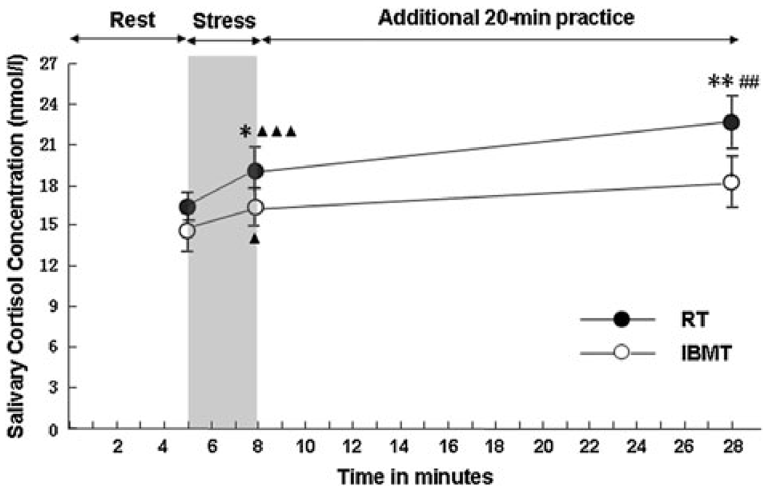

Figure 1. Salivary cortisol levels at the three stages of a stress intervention test after 2 weeks of training. *$p < 0.05$, **$p < 0.01$, integrative body–mind training (IBMT) group versus relaxation training (RT) group. ▲$p < 0.05$, ▲▲$p < 0.01$, ▲▲▲$p < 0.001$, immediately after stress versus rest. #$p < 0.05$, ##$p < 0.01$, immediately after additional practice versus immediately after stress

groups in the cortisol level following the additional practice [$F(1, 32) = 9.430$, $p < 0.01$].

Post-training cortisol measures (week 4)

Salivary cortisol levels at the three stages of a stress intervention test after 4 weeks of training are shown in Figure 2.

Following the acute stress, the cortisol level also significantly increased compared with the resting level for both groups, and there was significant difference between the two groups in the cortisol level immediately after stress ($p < 0.05$).

Following the additional 20-min practice, no significant changes were found from the cortisol level immediately after stress for both groups ($p > 0.05$). However, the cortisol level immediately after additional practice of the IBMT group was significantly lower than that of the control group [$F(1, 32) = 13.285$, $p < 0.01$]. Moreover, the cortisol level following the additional practice at week 4 was significantly lower than week 2 only for the IBMT group [$t(16) = 2.918$, $p < 0.05$].

Discussion

Cortisol elevation induced by long-term stress may contribute to the onset of various stress-related diseases, such as hypertension, diabetes, depression and cancer (Rabin, 2005; Reiche, Nunes & Morimoto, 2004). Many stress management studies have chosen salivary cortisol as a reliable physiological index to evaluate the efficiency of intervention (Hammerfald et al., 2006; MacLean et al., 1997; Matousek et al., 2010; Pawlow & Jones, 2005; Scholey et al., 2009). The

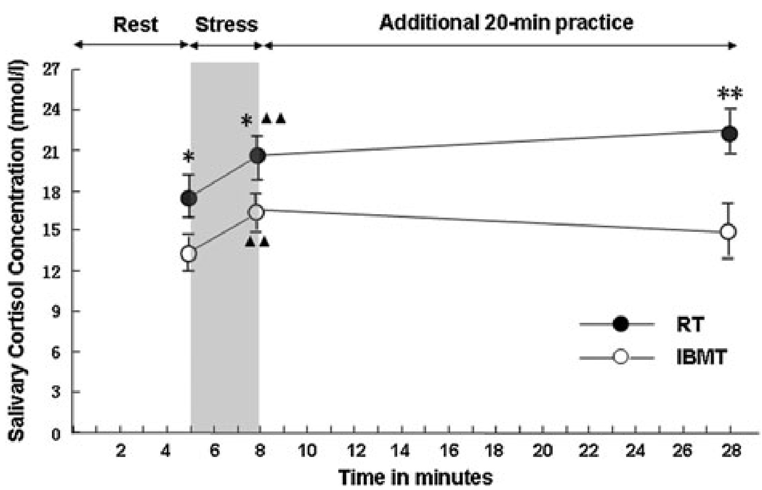

Figure 2. Salivary cortisol levels at the three stages of a stress intervention test after 4 weeks of training. *$p < 0.05$, **$p < 0.01$, integrative body–mind training (IBMT) group versus relaxation training (RT) group. ▲$p < 0.05$, ▲▲$p < 0.01$, immediately after stress versus rest

current study set out to determine if 2 or 4 weeks of IBMT practice will decrease the basal (resting) cortisol level and produce further improvements in endocrine function in response to stress.

Our results showed that the basal cortisol level decreased significantly in the IBMT group but not in the RT group after 2 and 4 weeks of training, and the basal cortisol level of the IBMT group was significantly lower than that of the controls following 4 weeks of training. Some studies have failed to find group differences in basal cortisol levels between controls and participants following an 8-week MBSR program (Robert McComb et al., 2004; Robinson et al., 2003). Volkmann and Weekes (2006) reported that basal secretory immunoglobin A (sIgA) (an index of mucosal immunity) and cortisol levels could predict stress-related health outcome. Participants with low basal sIgA levels and high basal cortisol levels had poorer health outcomes during the examination session than did participants with high basal sIgA levels and low basal cortisol levels. Recently, we reported that the basal sIgA levels increased significantly in a group trained with 4-week IBMT but not in a control group given the same amount of RT (Fan, Tang, Ma & Posner, 2010). These results indicate that IBMT would be a good endocrine and immune function modulator, which is helpful for health and stress management.

Stress leads to the secretion of cortisol. Salivary cortisol is a delayed peripheral response to acute stress (Wang et al., 2005). Studies showed that the level of salivary cortisol reached its maximum about 20 min after acute stress reaction (Gaab et al., 2003). The peripheral manifestations may have lagged because of the greater latency of HPA cascade. Exaggerated release of cortisol can suppress aspects of immune function and have negative effects on health (Segerstrom & Miller, 2004). Our previous study examined the dynamic changes in salivary cortisol and sIgA response to acute stress; the sIgA fall was significantly correlated with the cortisol rise during the 20 min after stress (Fan et al., 2009). We also demonstrated that 5 days of IBMT improves self-regulation and control of stress (Tang et al., 2007). Compared with the control group, 20 min of IBMT practice immediately after acute stress rendered the participants lower salivary cortisol and higher sIgA. In the present study, the IBMT group received an additional 20-min practice after stress, and the control group relaxed for 20 min. The results showed that no significant changes were found in the IBMT group from the level immediately after stress over the succeeding 20 min at both weeks 2 and 4, whereas there was a significant elevation in the control group at week 2, not at week 4. The cortisol levels immediately after stress and immediately after additional practice of the IBMT group were significantly lower than those of the control group. These results indicated that IBMT may assist in modulating endocrine function in response to stress.

People need to cope with high day-by-day stressful life events. IBMT provides a method to modulate endocrine and immune response to stress in time and to attenuate the negative effects of cortisol elevation induced by long-term stress on immune system and health. The present findings may indicate that IBMT produces persistent changes in the endocrine function and larger acute effects as the dose of training increases. It is suggested that the current results provide one mechanism through which long-term practice of IBMT could achieve improved health and stress management.

There were two limitations to this study. One was the small size of the sample (34 subjects), which consisted only of Chinese undergraduates. The other was the short study period (4 weeks), with no examination of whether the effects of IBMT would persist if no further practice occurred. Further studies could explore the effects of long-term training on the endocrine function both when practice continues and following its end and with a larger and more diverse sample. We also do not know whether two different coaches for IBMT or RT might have slight effects on the outcomes, although such effects (if any at all) would probably be minor because of the randomized design; a future study should address this.

Conflict of interest

All authors declare no conflict of interest.

Acknowledgments

We thank the radioimmunoassay division in Dalian Medical University for technical assistance and Institute of Neuroinformatics for data collection. This study was funded by grants from the 973 Project 2012CB518200 and the Office of Naval Research.

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