Taiji practice attenuates psychobiological stress reactivity - a randomized controlled trial in healthy subjects

Running Title: Taiji practice attenuates psychobiological stress reactivity

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Abstract

**Background:** Stress reducing effects of Taiji, a mindful and gentle form of body movement, have been reported in previous studies, but standardized and controlled experimental studies are scarce. The present study investigates the effect of regular Taiji practice on psychobiological stress response in healthy men and women.

**Methods:** 70 participants were randomly assigned to either Taiji classes or a waiting list. After 3 months, 26 (8 men, 18 women) persons in the Taiji group and 23 (9 men, 14 women) in the waiting control group underwent a standardized psychosocial stress test combining public speaking and mental arithmetic in front of an audience. Salivary cortisol and α-amylase, heart rate, and psychological responses to psychosocial stress were compared between the study groups. (ClinicalTrials.gov number, NCT01122706.)

**Results:** Stress induced characteristic changes in all psychological and physiological measures. Compared to controls, Taiji participants exhibited a significantly lower stress reactivity of cortisol (p = .028) and heart rate (p = .028), as well as lower α-amylase levels (p = .049). They reported a lower increase in perceived stressfulness (p = .006) and maintained a higher level of calmness (p = .019) in response to psychosocial stress.

**Conclusion:** Our results consistently suggest that practicing Taiji attenuates psychobiological stress reactivity in healthy subjects. This may underline the role of Taiji as a useful mind-body practice for stress prevention.

**Keywords**
Taiji, psychosocial stress, salivary cortisol, salivary α-amylase, heart rate
Introduction

The harmful impact of stress on health has been documented repeatedly (Brotman et al., 2007; Ehlert et al., 2001; Rozanski et al., 1999; Shonkoff et al., 2009). Stress reactivity research suggests that adverse consequences of psychosocial stress on physical and mental health may relate to stress-induced activation of different stress-responsive physiological systems (Brotman et al., 2007; Ehlert et al., 2001; Lovallo and Gerin, 2003; Raison and Miller, 2003). Large-magnitude physiological reactions to acute stressors in particular, often combined with delayed recovery, could be identified as stress-related risk factors for cardiovascular disease (Chida and Hamer, 2008; Chida and Steptoe, 2010; Steptoe et al., 2006). More precisely, stress-induced hyperreactivity of the two main human stress systems, the hypothalamus-pituitary-adrenal (HPA) axis with its end-product cortisol and the sympathetic nervous system (SNS) may increase cardiovascular risk, either alone and / or by inducing adverse changes in intermediate biological risk factors for cardiovascular disease such as coagulation activity, inflammation, or lipids (Brotman et al., 2007; Chida and Steptoe, 2010; Hamer et al., 2010; Lovallo and Gerin, 2003; Rosmond and Bjorntorp, 2000; Rozanski et al., 2005; Steptoe et al., 2007; von Kanel et al., 2001).

These findings underline the importance of investigating stress preventive interventions and their effects on psychobiological stress reactivity. By now, cognitive behavioural stress management has been repeatedly shown to markedly decrease psychological and biological reactivity towards psychosocial stress in healthy subjects (Gaab et al., 2003; Hammerfald et al., 2006). In contrast, the effectiveness of mind-body interventions for reducing psychosocial stress reactivity so far has only been examined for compassion meditation, showing a dose dependent effect on immune and psychological stress responses (Pace et al., 2009).

Mind-body practices are characterized as methods focusing on the interactions among the brain, mind, body, and behaviour, with the intent of using the mind to affect physical
functioning and promoting health (U.S. National Institutes of Health, 2010). In fact, an increasing amount of scientific evidence suggests that mind-body practices, such as Taiji, might contribute to improvements in physical and mental health (Jahnke et al., 2010; Klein and Adams, 2004; Wang et al., 2010a; Wang et al., 2010b). Taiji - variably spelled Taijiquan, Tai Chi or Tai Chi Chuan - is a mindful and gentle form of slow body movements with roots in ancient Chinese martial arts. Because of its integration of numerous physical, cognitive, and contextual components which potentially have independent as well as synergistic therapeutic value, Taiji has been described as a complex multi-component mind-body practice (Wayne and Kaptchuk, 2008).

A few studies suggest that Taiji may have stress reducing effects. Two studies suggest that practicing Taiji has short- and long-term effects on the basal activity of the HPA-axis. In his pioneering research work, Jin (1989) found that practicing Taiji for 60 min reduced cortisol levels after Taiji as compared to before. Similarly, results from a non-controlled pilot study found reduced salivary cortisol levels in healthy subjects, both immediately and four weeks after they completed a Taiji beginners course (Esch et al., 2007). Hitherto, studies exploring the suitability of Taiji as a stress management intervention are scarce. In terms of stress as measured by psychological measures only, a decrease of self-reported stress was observed in healthy young adults as well as in elderly subjects with cardiovascular disease risk factors and persons with HIV disease (Esch et al., 2007; Robins et al., 2006; Taylor-Piliae et al., 2006). To date, one randomized-controlled study assessed the effect of 60 min of Taiji practice on the psychobiological recovery of subjects after they were exposed to a non-validated stressor intended to induce mental and emotional stress by having them watch stressful movies and perform mental arithmetic under time-pressure and noise (Jin, 1992). A decrease of salivary cortisol was measured after Taiji as well as after three different interventions (reading, brisk walking, meditation). However, due to limited saliva sampling and missing pre-stress baseline measurement, findings from this study remain inconclusive.
Taken together, studies measuring self-reported psychometric parameters consistently suggest that Taiji may serve as an effective stress management intervention technique, but its effects on physiological reactivity to acute stress remain unclear.

To the best of our knowledge, randomized controlled trials examining the effects of Taiji on physiological and psychological reactivity to standardized psychosocial stressors have not yet been reported. We thus set out to investigate the effects of Taiji on psychobiological reactivity to a standardized and well-validated stressor, the Trier Social Stress Test (TSST). We repeatedly assessed different measures of independent stress responsive systems such as self-reported stressfulness, mood, and calmness, as well as the physiological stress indicators salivary cortisol, salivary α-amylase levels and heart rate. We hypothesized that practicing Taiji would be associated with lower psychobiological stress reactivity.

Methods

Participants and design

The ethics committee of the Canton of Bern, Switzerland formally approved the research protocol. Recruitment was carried out from April 2010 to August 2010 through advertisement of the study on pin boards and on the websites of the University of Bern and the University Hospital in Bern.

Through telephone screening, healthy subjects aged from 18 to 50 years and fluent in German were included if the following exclusion criteria did not apply within the six months prior to the screening (yes/no): regular or occasional intake of any medication, any self-reported acute or chronic somatic or mental disorders, smoking more than five cigarettes/day, consumption of more than two alcoholic drinks/day, consuming any kind of addictive substances, any previous participation in stress research projects (in order to ensure that
subjects included were naïve to the TSST protocol), more than one week of predictable absence during the intervention period, any previous practical experience with Taiji exercises. Women who were using hormonal contraceptives, were pregnant or planning to become pregnant during the study were also excluded. The included subjects received complete written and oral descriptions of the study. Informed written consent was obtained before participating. After baseline examination was completed, the participants were randomly assigned to either the Taiji group or the waiting control group. The allocation ratio was 1:1. Allocation concealment was achieved by using sequentially numbered, opaque and sealed envelopes. An independent research assistant generated the random allocation sequence by sealing, mixing and subsequently numbering 80 opaque envelopes. They were opened individually by the primary investigator (MN) for each eligible subject who had agreed to participate in the study and completed baseline examination. TSST examination was completed only on subjects with compliance to start and test instructions. The participant inclusion process is depicted in Figure 1.

Taiji intervention

The Taiji course being offered to the intervention group started in September 2010 and lasted for 12 weeks. The training sessions of 60 minutes took place twice a week. Taiji classes differed in composition (participants chose 2 of 6 potential training time points per week) and size (5 to 15 participants per session). Participants who missed a class were asked to attend a make-up class. The intervention group was encouraged to practice Taiji at home in addition to the classes. The average number of home practice sessions was assessed retrospectively using a brief self-report questionnaire at the end of the course. Participants’ class attendance was journalized by the Taiji teacher. All classes were held by the same Taiji teacher. He was trained in China as well as in Europe, has 10 years of Taiji experience, and is a certified Taiji
teacher as awarded by the Swiss Society for Qigong and Taijiquan (Schweizerische Gesellschaft für Qigong und Taijiquan – SGQT).

In the Taiji course, participants were taught the first 18 sequences of the 37 Chen Man-Ch’ing Yang-Style Taiji short form. An adaptation of five simplified Taiji movements from this form has been previously used in Taiji trials on patients with chronic heart failure (Yeh et al., 2004; Yeh et al., 2011). As our study participants were all healthy, we decided not to simplify the form but to teach the first 18 movements consecutively, as recommended by Robinson (2006). The main reasons for choosing this form are the following: (1) inclusion of the basic Taiji principles such as extension, relaxation and alignment of the body, as well as holistic and mindful body movements (Wolf et al., 1997), (2) feasibility given the moderate teaching and practicing time of two hours per week for three months, (3) enhanced embodiment of basic Taiji principles thanks to frequent repetitions enabled by the shortness of the 18 sequences. Moreover, the Cheng Man-Ch’ing form is widely taught in Switzerland and subjects interested in learning the remaining part of this form after the study would easily find a suitable Taiji-school. Each Taiji session began with warm-up exercises (15 min) followed by practicing Taiji movements and reviewing the underlying principles (35 min) and concluded with Taiji related breathing and relaxation exercises (10 min).

Prior to group allocation participants of both study groups were requested not to take part in any new physical exercise or mind-body program during their study participation. All participants agreed with this request. After the termination of the study, an equivalent Taiji training was offered to all subjects participating in the control group.

Assessment of potentially confounding variables

Potential Taiji-related confounding variable. We assessed participants’ previous regular practical experience (in months) with self-applicable mind-body practices (i.e. meditation, Feldenkrais, Alexander Technique, Qigong, Yoga, Pilates, guided imagery, deep breathing
exercises, progressive muscle relaxation and Reiki) at baseline to rule out a non-Taiji related influence of prior mind-body practice experience on the parameters under study.

*Potential confounders of physiological stress reactivity.* We controlled for age (Kudielka et al., 2004), as well as for menstrual cycle phase (luteal vs. follicular phase, see below) and gender as salivary cortisol reactivity in hormonal contraceptive-free female subjects is blunted during the follicular phase and differs from cortisol reactivity in male subjects (Kirschbaum et al., 1999). We asked all female participants to fill out a questionnaire assessing duration (days) and regularity of the menstrual cycle phase (yes/no), as well as the dates of onset of menstruation before and after the stress test examination. Luteal phase was defined as the time span of 14 days before onset of menstruation (Lenton et al., 1984). Additionally, we controlled for the cardiovascular risk factors smoking (number of cigarettes smoked per day) and body mass index (BMI, kg/m², see Table 1), as well as for regular physical activity (average hours per week) during the intervention period (Benson et al., 2009; Rimmemele et al., 2007; Rohleder and Kirschbaum, 2006).

*Procedure of the Trier social stress test (TSST) examination*

The experimental sessions were conducted during the first 3 weeks after termination of the 12 week Taiji intervention between 1300 h and 1800 h. The timing of the stress test performance was balanced between males and females and between participants in the two study groups. Participants were told to refrain from eating and drinking anything but water for 2 h and from intense physical activity, caffeine, nicotine, and alcohol during the 24 h before the experiment. Participants’ compliance to preparatory instructions and absence of the exclusion criteria was verified; non-compliant participants were excluded from the TSST. Next, the ECG recording equipment was attached and the recording was started. We used the Trier Social Stress Test (TSST) combining a 10 min preparation phase followed by a 5 min mock job interview, and a 5 min mental arithmetic exercise (Kirschbaum et al., 1993). Both tasks
were performed two meters in front of two evaluative panel members dressed in white laboratory coats, and a conspicuous video camera and microphone. The socio-evaluative character of this performance task was further underlined by presenting the panel members (a retired male finance manager and a female psychologist) as experts in evaluation of nonverbal behaviour. The TSST reliably activates HPA-axis and the sympathetic nervous system (Dickerson and Kemeny, 2004). During recovery, subjects remained seated in a quiet room for 60 min.

**Outcome measures**

All outcomes of interest were measured during the TSST-examination sessions. Physiological as well as psychometric measures were evaluated. Stress reactivity of repeated salivary cortisol levels (i.e. the interaction group-by-stress) was defined as the main outcome measure. Secondary measures included repeated α-amylase, heart rate, and different psychometric assessment tools.

**Physiological Measures.** Saliva samples (Salivette ®; Sarstedt AG, Sevelen, Switzerland) were obtained for determination of salivary cortisol (10 min (-20 min) and 1 min (-10min) prior to the TSST and immediately (+1 min) as well as 10, 20, 30, 45 and 60 min after stress cessation) and α-amylase levels (-20 min, -10 min, +1 min, +10 min, +20 min, +45 min). Samples were stored at -20 °C until assaying. After thawing, saliva samples were prepared for biochemical analysis by centrifuging at 3000 rpm for 5 min to produce a clear supernatant of low viscosity. Estimation of salivary free cortisol was performed using a chemiluminescence immunoassay with high sensitivity of 0.16 ng/mL (IBL Hamburg, Germany). Levels of α-amylase were determined following previously described methods (Rohleder and Nater, 2009). Both salivary cortisol and salivary α-amylase were analyzed in a commercial laboratory (Dresden LabService GmbH, Dresden, Germany). Inter- and intra-assay coefficients of variation were both below 8% (cortisol), and 10% (α-amylase),
respectively. A single-channel electrocardiogram (ECG, standard lead) was recorded continuously at 4036 Hz throughout the experimental session using a portable heart rate (HR) monitoring device (Medikorder MK3, TOM-Medical, Graz, Austria). HR data was aggregated to 5 min HR segments. The first 5 min HR segment (-10 min) was defined as baseline. HR segments measured before (-5 min), during (+5 min, +10 min) and after the stress task (+15 min, +20 min) were considered in statistical analyses.

_Psychometric measures._ Baseline group characteristics included assessment of perceived stress and depression symptoms. Perceived stress was assessed by the German version of the Perceived Stress Scale (PSS) (Cohen and Williamson, 1988). This 10-item self-report questionnaire measures subjects’ evaluation of the stressfulness of the situations experienced in the past month of their lives. Items in the PSS were designed to assess how predictable, uncontrollable and overloading participants consider their lives. Good internal consistency is reported (Cronbach’s $\alpha = .78$). Depressive affect and negative thought patterns were measured by using the “Allgemeine Depressionsskala-Kurzform” (ADS-K) questionnaire (Hautzinger and Bailer, 1993), the German version of the “Center for Epidemiological Studies Depression Scale” (CES-D) (Radloff, 1977). This questionnaire was developed for research in the general population and has shown good internal consistency (Cronbach’s $\alpha = .90$). We measured psychological TSST stress reactivity at baseline and immediately after stress cessation: the Multidimensional Mood Questionnaire (MDMQ) assesses self-reported mood and calmness with good internal consistencies (“mood” - Cronbach’s $\alpha = .75$ to .87; “calmness” - Cronbach’s $\alpha = .77$ to .83) (Steyer et al., 1997). Psychological evaluation of perceived stressfulness during the TSST examination was obtained by completion of a visual analogue scale (VAS) ranging from 0 to 10 with 0 indicating no stress experienced at all.

*Statistical analysis*
Data were analysed using SPSS (version 18) statistical software package for Macintosh (IBM SPSS Statistics. Somers, NY, USA). The calculation of the optimal total sample size has been conducted using the statistical software G*Power (Buchner et al., 1997). Based on prior research on cortisol stress responses reporting effect sizes ranging from $f^2 = .28$ to .35 (Gaab et al., 2003; Storch et al., 2007), the optimal total sample size of $N = 64$ was calculated a priori to detect an expected medium to large effect size of $f^2 = .25$ with a power $\geq .85$ and $\alpha = .05$ (effect size conventions: $f^2$: .02 = small, .15 = medium, .35 = large; see Cohen, 1988).

Effect size parameters ($f$) were calculated from partial $\eta^2$-values and are reported where appropriate (effect size conventions: $f$: .10 = small, .25 = medium, .40 = large; see Cohen, 1988). All analyses were two-tailed, with the level of significance set at $p < .05$ and the level of borderline significance at $p < .10$. Unless indicated, all results are presented as mean ± standard error of means (S.E.M.). Prior to statistical analyses all data were tested for normal distribution and homogeneity of variance using a Kolmogorov-Smirnov and Levene test. As cortisol levels were skewed we log-transformed (log10) cortisol data and obtained a normal distribution. Log-transformed cortisol data were used in statistical analyses but for reasons of clarity untransformed data are depicted in Figure 2a.

Group characteristics were analyzed by $\chi^2$-analysis for categorical data, and independent samples $t$-test for continuous data. Group differences in TSST related baseline values were also tested by $t$-tests.

To reveal possible time and condition effects, repeatedly measured physiological and psychological data were analyzed by using two way ANCOVAs with repeated measurements with group as the independent factor (Taiji group vs. control group) and cortisol, heart rate, $\alpha$-amylase, perceived stressfulness, mood, and calmness data as repeated dependent factors. We applied Huynh-Feldt correction where appropriate.

To prevent overcontrolling given our sample size (Babyak, 2004), we performed a two-step procedure for analyses of physiological parameters. In the first step, representing the
main analysis for the primary outcome measure cortisol, we calculated repeated cortisol ANCOVAs while controlling for cortisol baseline levels, prior experience with self-applicable mind-body practices, age, menstrual cycle phase, and gender as a priori defined covariates. Significant effects were further tested in a second step, where we additionally controlled for smoking and BMI, as well as regular physical activity during the intervention period. Analyses for α-amylase and heart rate were calculated accordingly. In analyses of repeated psychological data, we controlled for prior mind-body practice experience as a covariate.

Post-hoc testing of significant effects in the main analyses included separate recalculation of the previously described ANCOVA analyses for each of the repeated time points.

Results
Of the 112 subjects who underwent a telephone screening, 40 subjects did not fulfil selection criteria. Reasons for exclusion and drop-out are documented in Figure 1. Of the remaining 74 subjects, 70 successfully underwent baseline examination and were randomly assigned to either the Taiji group (N = 35) or to the waiting control group (N = 35). TSST examination was completed by 26 subjects from the Taiji group (mean age 35.77 ±1.61; 69% female) and by 23 subjects from the control group (mean age 35.74 ±1.31; 61% female) as 21 subjects dropped out before the TSST or did not fulfil inclusion criteria for the TSST (Fig. 1). Since 95% of all dropouts have not attended the TSST examination, an intention-to-treat approach is not applicable to this study design. Therefore only subjects completing the TSST were included in data analysis. We had no missing data. No adverse effects of the Taiji training were observed or reported.

Group characteristics
The two study groups did not significantly differ in group characteristic (Table 1) and drop-out rate (p = .603). Drop-out subjects did not significantly differ from the subjects completing the study in any group characteristic (p’s > .415), except BMI (21.17 ± .49 (drop-out group) vs. 23.49 ± .51 (final study group); p < .001).

**Physiological stress reactivity**

At baseline, the study groups did not differ in cortisol, α-amylase, or heart rate. The TSST induced significant increases in all physiological measures under study (main effects of stress: p’s < .001). When controlling for confounders considered in the main analyses, a significant main effect of stress was observed for cortisol (p < .001) and heart rate (p = .027), but not for α-amylase (p = .91).

**Cortisol.** The Taiji group showed an attenuated cortisol stress reactivity as compared to the control group while controlling for the first set of confounders (i.e. physiological baseline level, age, gender, menstrual cycle phase, and prior mind-body practice experience) [interaction group-by-stress: F(2.92/122.50) = 3.16, p = .028, partial η² = .07, f = .27; main effect of group: F(1/0.79) = 2.99, p = 0.091; Fig. 2a]. Additional controlling for the second set of confounders (i.e. smoking status, BMI, and physical activity) did not significantly change results (p = .044, resp. p = .122). Post-hoc tests revealed a trend towards lower cortisol levels 10 min [F(1/0.22) = 3.80, p = .058], 30 min [F(1/0.20) = 2.96, p = .093], 45 min [F(1/0.20) = 3.71, p = .061] and 60 min after stress cessation [F(1/0.20) = 3.80, p = .058] in the Taiji group, suggesting a lower increase and a faster recovery of salivary cortisol in the intervention group (see Fig. 2a).

**Alpha-amylase.** Compared to controls, participants of the Taiji group showed significant lower α-amylase activity before and after stress while controlling for the first set of
confounders [main effect of group: F(1/100795.10) = 4.12, p = .049, partial η² = .089, f = .31; Fig. 2b]. No significant group difference was found for α-amylase stress reactivity [interaction group-by-stress: p = .16]. After additional consideration of the second set of confounders the main effect of group remained significant (p = .040) and a trend towards reduced α-amylase stress reactivity in participants of the Taiji group was revealed [interaction group-by-stress: p = .086]. Post-hoc testing showed significantly lower α-amylase levels 10 min [F(1/40874.38) = 6.63, p = .014], 20 min [F(1/23952.02) = 4.03, p = .051] and 45 min after stress cessation [F(1/41612.24) = 8.66, p = .005] in the Taiji group as compared to the control group, indicating a faster recovery (see Fig. 2b).

Heart rate. The main analysis revealed a significantly blunted heart rate stress reactivity in the Taiji group compared to the control group [interaction group-by-stress: F(2.55/173.76) = 3.34, p = .028, partial η² = .087, f = .31; Fig. 2c]. Furthermore, the participants of the Taiji group showed a trend towards lower heart rate levels before and after the stress protocol [main effect of group: F(1/750.01) = 3.15, p = .083; Fig. 2c]. Additional controlling for the second set of confounders did not significantly change results. Post-hoc tests revealed that participants of the Taiji group exhibited significantly lower heart rate levels during the first 5 min [F(1/559.40) = 5.93, p = .019], and the second 5 min [F(1/569.63) = 4.33, p = .044] of the TSST as compared to controls (see Fig. 2c).

Psychological stress reactivity

At baseline subjective measures of perceived stressfulness, mood, and calmness did not differ between study groups. The TSST significantly increased perceived stressfulness, worsened mood, and reduced calmness in all study participants (main effects of stress: p’s < .001). Controlling for prior experience with self-applicable mind-body practices did not significantly change findings.
Compared to the controls, participants in the Taiji group reported significantly less stressfulness [F(1/16.37) = 8.48, p = .006, partial η² = .156, f = .43], maintained a higher level of calmness [F(1/21.79) = 5.87, p = .019, partial η² = .113, effect size f = .36] and tended towards a lower decrease of mood [F(1/17.78) = 3.43, p = .070] in reaction to the TSST (see Table 2).

Discussion
This is the first randomized-controlled study to explore effects of Taiji on measures of adrenocortical, autonomic, and psychological responses to a standardized and validated psychosocial stress task in healthy Taiji beginners. We found for the first time markedly reduced psychobiological stress responses in Taiji practitioners as compared to a non-Taiji control group, i.e. attenuated cortisol and heart rate stress reactivity, lower α-amylase levels, as well as lower perceived stressfulness and better maintenance of calmness in response to the stress task. Baseline values did not differ between groups and stress induction proved to be successful, as indicated by the expected significant increases in all physiological measures under study in the total sample.

The present results extend previous research by suggesting an overall stress-buffering effect of Taiji practice on a broad array of measures representing different stress-responsive systems with effect sizes ranging from medium to large. Notably, we recruited Taiji beginners and our Taiji intervention lasted for 12 weeks. As Taiji is thought to improve its beneficial effects with increasing practice skills over years (Cheng, 1982), it can be speculated that the observed effects may be even more pronounced in advanced Taiji practitioners.

In contrast to stress management interventions (Gaab et al., 2003; Hammerfald et al., 2006; Storch et al., 2007), the Taiji course in our study was neither designed nor taught as a form of stress management. It was conceptualized to convey an embodiment of basic Taiji
principles by applying a guided introspective teaching approach. We did not train any specific coping strategy (e.g., cognitive restructuring) nor did we use role-plays or psychodrama course elements as often used in cognitive behavioral (Gaab et al., 2003; Hammerfald et al., 2006) and resource activating stress management programs (Storch et al., 2007). In contrast to our Taiji course, training elements of such stress management programs may, in addition to their specific effects on stress appraisal, have more similarities to the TSST situation and therefore might additionally prepare for the stress test itself. Considering the lacking emphasis on stress management in the Taiji intervention, the incongruence between the training environment and the TSST setting, and the focus on developing Taiji related body awareness and body mechanics, we feel that the stress protective effects of Taiji observed in our study are likely to result from a mindful embodiment of effortless stability and calmness in motion. This reasoning is further supported by our finding that, similarly to the Taiji effect, the control variable “prior experience with mind-body practices” (other than Taiji) was significantly associated with blunted cortisol stress reactivity, lower α-amylase levels, as well as lower perceived stressfulness and better maintenance of mood in response to the stress task (p’s < .040).

Prior research further supports that the observed attenuation of psychobiological stress reactivity in our Taiji group may relate to mind-body interaction effects. An increased body awareness induced by regular Taiji practice has been reported in previous studies (Gyllensten et al., 2010; Uhlig et al., 2010) and is likely to enhance a resource activating embodiment. Maintaining resource activating embodiment in turn has been shown to reduce cortisol levels under resting conditions (Carney et al., 2010). Moreover, coping strategies including embodiment were an integrated part of a resource activating stress management program found to attenuate the reactivity of the HPA-axis in response to the TSST (Storch et al., 2007). As participants did not report any Taiji-participation induced increase in social contacts (data not shown), it seems unlikely that the observed stress-buffering in the Taiji
group relates to a training-induced increase in social support. However, it may be speculated that subjects in the Taiji group, because of their participation in the active study group, might have expected to be better prepared for their upcoming performance task and thus achieved greater emotion regulation during the TSST. To clarify the potential contribution of such an expectancy effect, future research is needed, preferably by including an additional active control group with an intervention raising similar expectations.

Our study has several strengths. First, we used a well-validated standardized acute psychosocial stress task (Dickerson and Kemeny, 2004; Kirschbaum et al., 1993). Second, we used a non-Taiji control group with randomized assignment. Third, we assessed multiple parameters indicating reactivity of different independent stress responsive systems. Fourth, baseline characteristics were thoroughly collected and both study groups were homogenous regarding their demographic and psychometric parameters, indicating a successful randomization of subjects. Finally, both groups had moderate scores in questionnaires assessing baseline levels of perceived stress and depressive affect. It therefore seems unlikely that the reported results are influenced by pre-existing group differences or selection bias related to increased proneness to stress.

The following limitations need to be considered. First, our results are restricted to a group of healthy and well-educated young to middle-aged men and women. They cannot be generalized to other groups with less advantageous health conditions or social backgrounds. Second, the retrospectively assessed average number of Taiji home practice sessions per week, the average time spent on sportive activities per week during the intervention period, and the determination of the menstrual cycle phase were based on self-report. Third, our results are restricted to Taiji novices. The effects of long-term Taiji practice on psychobiological stress-reactivity still need to be investigated. Fourth, our psychobiological assessment approach does not include assessment of further stress-responsive physiological systems such as the immune, the lipid, or the coagulation system. Also, HPA axis measures
other than cortisol such as corticotropin releasing hormone (CRH), or adrenocorticotrophic hormone (ACTH) still need to be examined. Fifth, because of habituation of cortisol responses in the majority of people repeating the TSST (Schommer et al., 2003), it was not possible to assess cortisol stress responses before and after the intervention, nor in the control group after completion of their Taiji course. Sixth, Our non-significant effects of Taiji on α-amylase stress reactivity should be interpreted with care as we cannot rule out a type II error. Future studies, preferably with a higher power, are needed to confirm our non-significant effects of Taiji on α-amylase stress reactivity as well as the overall stress-reducing effects on the other measures under study. Finally, due to our restrictive exclusion criteria we had a drop-out rate of 30%. However, this rate is comparable to a prior TSST study examining mind body practices (Pace et al., 2009) and does not seem unusual in studies examining Taiji effects on psychological well-being (Wang et al., 2010a).

In conclusion, our results consistently suggest that practicing Taiji attenuates psychobiological stress reactivity. This may underline the role of Taiji as a useful mind-body practice for stress prevention which may contribute to enhance health in the general population. Clinical implications remain to be elucidated.

Conflict of interest

All authors declare that they have no conflicts of interest.

Role of funding source

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Contributors

MN was involved in study design, writing of the proposal for the ethics committee, enrolment of participants, supervising the intervention, data collection, data analysis, and writing of the manuscript. BA was involved in study design, writing of the proposal for the ethics committee, and manuscript reviewing; KS and RS were involved in study design, and manuscript reviewing; PW was involved in study design, data analysis, and writing of the manuscript. All authors contributed to and have approved the final manuscript.
References


# Tables

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<th>Group characteristics</th>
<th>Taiji group (n = 26)</th>
<th>Control group (n = 23)</th>
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<td>Age 1 (years)</td>
<td>35.77 ±1.61</td>
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<td>.99</td>
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<td>Gender (male / female)</td>
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<td>9 / 14</td>
<td>.56</td>
</tr>
<tr>
<td>Menstrual cycle phase at TSST examination day</td>
<td>9 / 9</td>
<td>4 / 10</td>
<td>.29</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>23.47 ±.67</td>
<td>23.51 ±.79</td>
<td>.97</td>
</tr>
<tr>
<td>Education (with / without high school degree i.e. Swiss “Matura”)</td>
<td>20 / 6</td>
<td>17 / 6</td>
<td>1.00</td>
</tr>
<tr>
<td>Occupational status (full or part time workers / students)</td>
<td>24 / 2</td>
<td>23 / 0</td>
<td>.49</td>
</tr>
<tr>
<td>Smoking (non smokers / light smokers 2)</td>
<td>21 / 5</td>
<td>18 / 5</td>
<td>1.00</td>
</tr>
<tr>
<td>Sportive activity – during the intervention (hrs/week)</td>
<td>2.40 ±.37</td>
<td>2.98 ±.52</td>
<td>.37</td>
</tr>
<tr>
<td>Previous experience with mind-body practices (months of regular practice; pre intervention)</td>
<td>15.62 ±6.03</td>
<td>29.13 ±10.68</td>
<td>.26</td>
</tr>
<tr>
<td>Depressive affect (ADS-K score)</td>
<td>10.88 ±1.35</td>
<td>11.04 ±1.45</td>
<td>.94</td>
</tr>
<tr>
<td>Perceived stress (PSS score)</td>
<td>17.46 ±1.03</td>
<td>17.91 ±1.22</td>
<td>.78</td>
</tr>
<tr>
<td>Taiji classes attended (incl. % -value)</td>
<td>20.65 ±.59 (86%)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Taiji practice at home (number of sessions/week)</td>
<td>1.69 ±.33</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

1 Continuous data are expressed as mean ± S.E.M.

2 smoking less than 5 cigarettes per day
<table>
<thead>
<tr>
<th>Variables</th>
<th>Taiji Group (n = 26)</th>
<th>Control Group (n = 23)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre TSST</td>
<td>Post TSST</td>
<td>Stress change $^2$</td>
</tr>
<tr>
<td>Self-reported stressfulness (VAS) $^4$</td>
<td>1.22 ±.19</td>
<td>3.47 ±.45</td>
<td>2.25 ±.39</td>
</tr>
<tr>
<td>Calmness (MDMS) $^5$</td>
<td>16.47 ±.46</td>
<td>12.78 ±.62</td>
<td>-3.69 ±.54</td>
</tr>
<tr>
<td>Mood (MDMS) $^5$</td>
<td>17.30 ±.44</td>
<td>14.23 ±.74</td>
<td>-3.07 ±.64</td>
</tr>
</tbody>
</table>

1 All data are expressed as mean ± S.E.M.

2 Stress change = post TSST value minus pre TSST value

3 p-values refer to repeated measures ANCOVAs with prior mind-body practice as a covariate

4 VAS = visual analogue scale ranging from 0 = ‘not stressful at all’ to 10 = ‘extremely stressful’

5 MDMS = multidimensional mood scale ranging from 5 to 20 with higher scores indicating a higher degree of calmness and a more positive mood
Figure Legends

Legend to Figure 1

Flow diagram for the progress through the phases of the randomized trial (based on the consolidated standards of reporting trials [CONSORT] recommendations).

Legend to Figure 2

Values are means ±S.E.M. We calculated ANCOVAs with repeated measures of physiological stress parameters as dependent variables and group (Taiji vs. Control) as the independent variable. We controlled for physiological baseline level, age, gender, menstrual cycle phase and prior experience with mind-body practices as covariates. The Taiji group showed attenuated cortisol stress reactivity (p = .028; Figure 2a), α-amylase levels (p = .049; Figure 2b), as well as lower heart rate stress responses (p = .028; Figure 2c). Significance levels are: ° = p < .1;  * = p < .05;  ** = p < .01).
Figure 1 Documentation of the subject inclusion process

112 subjects underwent telephone screening
- 38 were excluded
  - 18 used hormonal contraceptives
  - 7 were older than 50 years
  - 7 had a physical illness
  - 3 had scheduling conflicts
  - 1 had a mental disorder
  - 1 had practiced Taiji before
  - 1 lost interest in participation

74 subjects underwent baseline examination
- 2 had scheduling conflicts
- 2 lost interest in participation

70 subjects underwent randomization

35 were assigned to the Taiji group
- 2 drop-outs prior to Taiji intervention due to scheduling conflicts
- 5 drop-outs during the Taiji intervention
  - 1 had a physical injury (not related to the intervention)
  - 4 had scheduling conflicts
- 2 drop-outs prior to TSST appointment
  - 1 was due to sickness unable to attend TSST session
  - 1 took hormonal contraceptives prior to TSST

25 completed TSST examination

35 were assigned to the waiting control group
- 3 lost interest in participation right after group assignment
- 2 were due to sickness unable to attend TSST session
- 2 took antidepressive prior to TSST
- 2 took hormonal contraceptives prior to TSST
- 1 was unwilling to complete TSST
- 1 was pregnant
- 1 moved out of Bern area

23 completed TSST examination
Figure 2a

The graph illustrates the changes in salivary cortisol levels (nmol/L) over time for two groups: the control group and the Taiji group. The x-axis represents time in minutes, ranging from -10 to 60 minutes, while the y-axis shows salivary cortisol levels from 0 to 20 nmol/L. The control group's cortisol levels increase sharply during the TSST (Trier Social Stress Test) phase and then decline, whereas the Taiji group shows a more gradual increase with a lower peak compared to the control group.
Figure 2b

The graph shows the salivary alpha-amylase levels over time for two groups: control group and Taiji group. The x-axis represents time in minutes, ranging from -10 to 45, and the y-axis represents salivary alpha-amylase levels in U/ml, ranging from 0 to 300. The shaded area indicates the TSST event, and the graph illustrates a significant increase in salivary alpha-amylase levels in both groups during and immediately after TSST, with the control group showing a more pronounced response. Statistical significance is indicated by the symbols: *, **, and ***.
Figure 2c

![Graph showing heart rate changes during TSST (Trier Social Stress Test) for control and Taiji groups. The x-axis indicates time points: pre TSST, TSST 5min, TSST 10min, post TSST 5min, post TSST 10min. The y-axis represents heart rate (beats/minute) ranging from 60 to 110. The control group shows a significant increase in heart rate during the TSST, with a peak around TSST 10min, followed by a gradual decrease post TSST. The Taiji group maintains a lower heart rate throughout the test period.]