Long-term meditation is associated with increased gray matter density in the brain stem

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Extensive practice involving sustained attention can lead to changes in brain structure. Here, we report evidence of structural differences in the lower brainstem of participants engaged in the long-term practice of meditation. Using magnetic resonance imaging, we observed higher gray matter density in lower brain stem regions of experienced meditators compared with age-matched nonmeditators. Our findings show that long-term practitioners of meditation have structural differences in brainstem regions concerned with cardiorespiratory control. This could account for some of the cardiorespiratory parasympathetic effects and traits, as well as the cognitive, emotional, and immunoreactive impact reported in several studies of different meditation practices. ***NeuroReport*** **20:170–174 © 2009 Wolters Kluwer Health | Lippincott Williams & Wilkins.**

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Introduction

Investigations using voxel-based morphometry have revealed robust evidence of structural neuroplasticity related to learning in tasks requiring sustained states of attention. Recent cross-sectional studies have shown gray matter changes associated with learning to navigate in large-scale space [1], bilingualism [2], learning to juggle [3], and revising for exams [4].

Meditation designates a wide variety of techniques of mental training, which can be divided into two broad styles. Concentrative practices (CPs) are aimed at sustaining attention upon mental content, for example, particular external or internal objects such as concepts, sounds (mantra), or bodily sensations – frequently those associated with breathing. Open awareness (OA) practices do not have a specific focus, but aim to develop a more panoramic and reflexive monitoring ability in which mental content is registered, but not fixated upon [5].

To our knowledge, only two earlier studies examined the structural correlates of meditation practice [6,7]. In the first study [6], the authors used MRI-based cortical thickness measurements in a cross-sectional study. The finding was that the right prefrontal cortex (Brodmann area 9/10) and the right insula of CP meditation practitioners had thicker cortical layers than the control group. The second study [7] showed that Zen meditators did not show the decreased attentional performance and left putamen gray matter volume as a function of age found in a control group. However, neither study investigated the lower brain stem region.

As attention to breathing is a common element in meditation training in many traditions [8], and as meditation is known to have lasting effects on respiration control [9] and respiration rate, skin conductance, and oxygen consumption are all reduced [10], it is plausible that the long-term practice of such forms of meditation might induce structural changes in brain regions involved in basic autonomic regulation as suggested earlier [8,11].

To study the possible structural effects of long-term meditation practice, we therefore compared groups of highly experienced meditators and normal controls using voxel-based morphometry of whole-brain MRI including the lower brainstem.

Methods

Participants

We compared a group of 10 experienced mixed-sex meditators (mean age 55.0 years ± SD 6.2, six males and four females) with 10 right-handed and age-matched mixed-sex normal controls with no history of meditation practice (mean age 58.0 years ± SD 6.8, five males and five females). All participants were ethnic Danes. All the participants were healthy with no history of neurological disorders, psychological and/or psychiatric, cardiovascular

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diseases, brain injury, cancer, addiction to drugs/alcohol, severe impediment to limb movement, hearing, and vision. The meditating participants have all practiced the same style of meditation following instructions from the same teacher in the Dzogchen tradition of Tibetan Buddhism, accumulating 8000–35 000 h of practice during the past 14–31 years, on an average of 16.5 years (SD = 5.1). Participants practiced meditation in formal sessions for an average of 2.2 h/day (SD = 0.48). The daily practice of our participants combines elements of CP and OA. CPs include a series of four exercises involving conscious attention to breathing. Elements of loving kindness and compassion [11] meditation are also included in each practice session, which serve to create a suitable environment for OA, which is their main practice. The experiment was approved by the local ethics committee. Informed written consent was obtained from all the participants.

Scanning protocol

We used high-resolution MRI scans with standard voxel-based morphometry and statistical mapping methods [12] to identify significant regional differences between the two groups using whole-brain voxel-wise statistics. All experiments were performed on a 3.0 T General Electric Medical Systems Excite MRI scanner (GE Medical Systems, Milwaukee, USA). A high-resolution T_1-weighted three-dimensional inversion recovery spoiled gradient echo sequence was used to image the whole brain and brain stem from vertex to approximately 2.5 cm below the lower rim of the cerebellum covering well below the medulla oblongata. Repetition time/inversion time/echo time/flip angle = 6.2/750/2.8 ms/14°, matrix size = 256 × 256, axial slice thickness = 1.2 mm, and field-of-view = 240 mm giving a voxel size of 0.94 × 0.94 × 1.20 mm. Total acquisition time was 15 min.

To visualize and identify the fine anatomical structures of the lower brainstem, which is not possible with human in-vivo MR images of humans because of time factors, high-resolution images of a formalin-fixed normal human adult postmortem brainstem were coregistered to the common coordinate system of all volunteers (Montreal Neurological Institute, Montreal, Quebec, Canada). using rigid-body transformations (SPM5, University College of London, London, UK). Two-dimensional spin echo imaging was used with repetition time/echo time/matrix size/averages = 8 s/18 ms/512 × 512/53, slice thickness = 0.5 mm, and field-of-view = 5.4 mm giving an in-plane image resolution of 105 × 105 μm. Total acquisition time was 61 h.

Voxel-based morphometry

We applied voxel-based morphometry [12] to analyze both gray matter density and volume differences between the two groups. A unified voxel-based morphometry procedure VBM5 was used (Christian Gaser, Department of Psychiatry, University of Jena, Germany, available as a SPM toolbox at *http://www.fil.ion.ucl.ac.uk/spm/*). The method involved the following procedures: linear and nonlinear spatial normalization into the same stereotaxic space (Montreal Neurological Institute, Canada). Segmentation into gray, white, and cerebrospinal fluid components by using the International Consortium for Brain Mapping probabilistic atlas of European brains (*http://www.loni.ucla.edu/ICBM/ICBM_TissueProb.html*).

General linear model

Normalized gray matter maps of all participants were smoothed with an isotropic Gaussian kernel of full-width-half-maximum equal to 4 and 12 mm to produce two data sets with a different degree of spatial smoothing to sensitize the statistic analysis to spatial differences of these two spatial extents. This was done to obey the normality assumption by the central limit theorem required by the parametric tests (*t*-test and F-test) of SPM5 [12]. The 12-mm kernel was used for the cortical regions, whereas for the reported results of fine structures in the brain stem region a 4-mm kernel was used.

Statistics

Two-group *t*-test of SPM5 was applied to identify gray matter density and volume differences between the meditators and the control group. The resulting statistical images were thresholded at a false-discovery rate of a P value of less than 0.05 (corrected for multiple comparisons across the whole brain). The regions of structural differences in brain stem gray matter could then be mapped onto the ultra high-resolution scan of the normal adult postmortem brain stem (Fig. 1) and histological atlases could then be used to identify the anatomical substructures as illustrated in Fig. 2.

Results

Group comparison of the gray matter density showed significantly increased levels of gray matter density but no volume differences in the meditators in a well-defined region of the lower brain stem – the medulla oblongata (Fig. 1). Moreover, there was complete separation of gray matter densities between the groups in the brain stem region (Fig. 1d).

The region extended bilaterally from the dorsal part of medulla oblongata (left: $x = -5$; $y = -46$; $z = -51$, $Z = 5.49$, right: $x = 5$; $y = -45$; $z = -51$, $Z = 4.89$) to the anterior and more caudal part (with peak in right: $x = 3$; $y = -37$; $z = -58$, $Z = 5.24$). The region includes bilaterally the solitary nucleus and tract (the so-called dorsal respiratory group), the dorsal motor nucleus of the vagus, the nucleus ambiguous, ventrally, and more caudally, the medullar reticular formation and the inferior olivary complex (Fig. 2). The gray matter differences had a slight asymmetric character, with the largest changes in dorsal regions on the left (in the solitary nucleus, the dorsal motor nucleus of the vagus) and in ventral and

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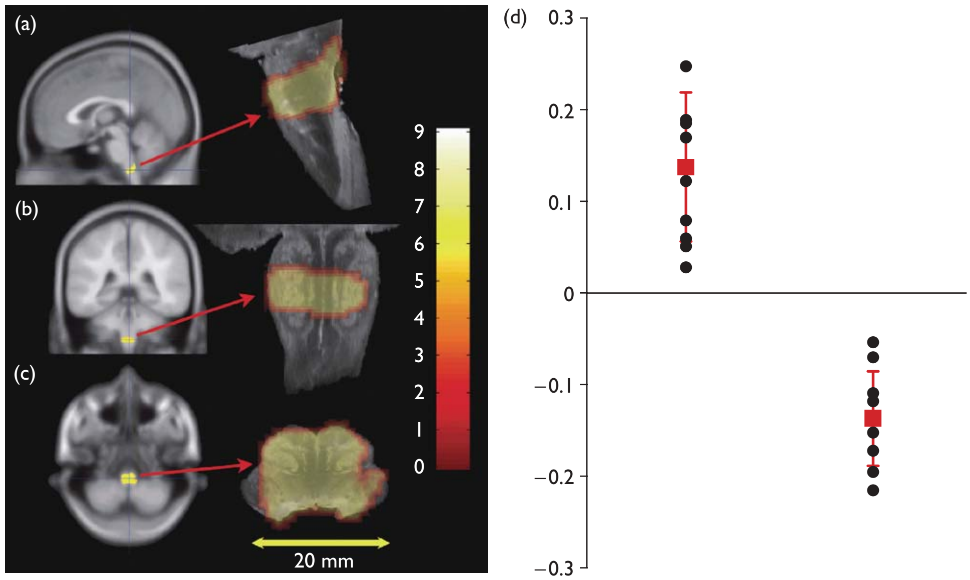
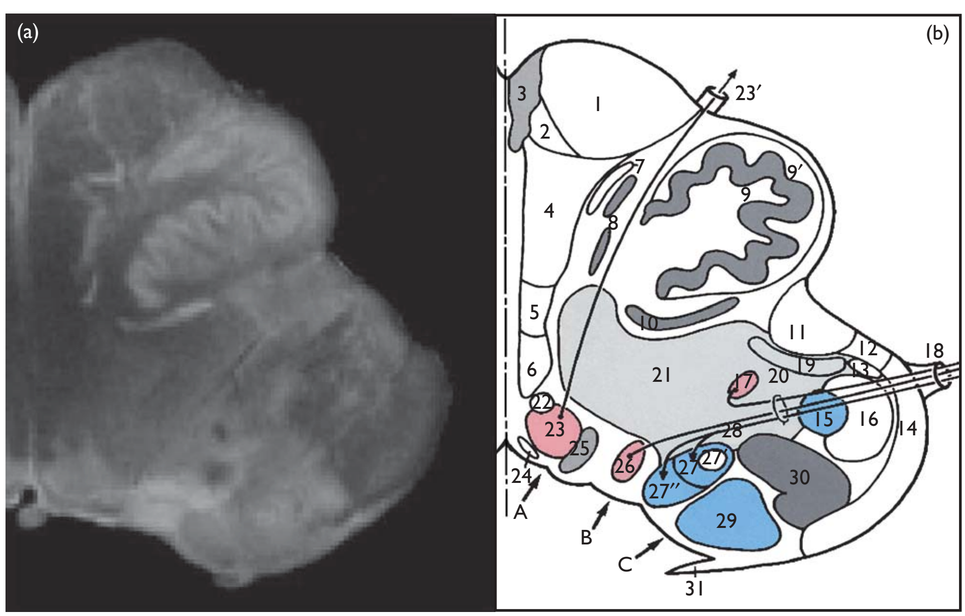
Fig. 1



Increased gray matter density in the brain stem of the meditators. All data were corrected for multiple comparisons, false-discovery rate ($P<0.05$). Degrees of freedom = 18. On the left the region is superimposed on a T1 MR image in the same stereotaxic space. (a) Sagittal view, (b) coronal view, (c) axial view. On the right the region is superimposed on a 105-µm resolution MR image of the human medulla oblongata *in vitro*, which was coregistered to the stereotaxic space of all participants (Montreal Neurological Institute, Canada). Color scale indicates T-score. (d) Relative gray matter density difference in the peak voxel for the groups of meditators (left) and controls (right). Individual data are shown as well as the group mean and standard deviation.

Fig. 2



Identification of regions in the human lower brain stem. (a) Zoomed axial section of the medulla oblongata in Fig. 1c. (b) Axial section of the medulla oblongata at the level of the middle inferior olivary nucleus (drawing modified and reproduced with permission, H.M. Duvernoy, The Human Brain Stem and the Cerebellum, Springer-Verlag 1995). Regions: 9, inferior olivary nucleus; 17, nucleus ambiguous; 18, fibers of the vagus nerve; 21, nucleus reticularis medullae oblongatae centralis; 26, dorsal motor vagal nucleus; 27, nucleus of the solitary tract; 27′, solitary tract; 27′, nucleus gelatinosus of the solitary tract.

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more caudal regions on the right (approximate center in the reticular nucleus of the medulla/nucleus ambiguous).

Increased gray matter densities were also found in the left superior frontal gyrus Brodmann 10 ($x = -24$; $y = 50$; $z = 4$, $Z = 4.29$) and the left inferior frontal gyrus – more specifically the pars triangularis (Brodmann 45A) ($x = -44$; $y = 28$; $z = 6$, $Z = 3.96$). Further, differences were found in the anterior lobe of the cerebellum bilaterally [33, −59, −36, and −29, −58, −37 ($Z = 4.14$ and 4.61) and a small region in the left fusiform gyrus (−52, −19, −29, $Z = 4.75$)]. Correlation analysis showed no significant difference in gray matter density as a function of total practice hours (8000–35 000 h) in any of the regions.

Discussion

This study indicates that there are structural differences associated with long-term meditation in the gray matter of the medulla oblongata, the anterior cerebellum (bilaterally), in the left superior and inferior frontal gyrus as well as the left fusiform gyrus.

The largest differences were detected in the medulla oblongata region of the dorsal brain stem known to contain autonomic nerve system structures, such as the solitary tract nucleus and the dorsal motor nucleus of vagus (Fig. 2), which are involved in relaying sensory inputs from the body and in respiratory and cardiac control. The main function of the dorsal respiratory group (in the solitary tract nucleus) is to control the basic rhythm of respiration. In a functional MRI study, the same section of the dorsal medulla oblongata has been shown to mediate behavioral respiratory control in humans [13]. Parasympathetic fibers from the dorsal motor nucleus of the vagus innervate, through the vagus (X cranial nerve), the heart muscle, and the smooth musculature and glands of the respiratory and intestinal tracts.

The structural differences in the dorsal medulla gray matter in the region of autonomic respiratory control centers are noteworthy, because studies have shown that breathing and heart rate are reduced during meditation [6,14]. Moreover, the finding of a decreased response to progressive hypercapnia [9] indicates that there are lasting effects on respiration control as a trait of meditation practice. Studies have shown that several types of meditation practice are associated with increased vagal tone [15] and related traits such as a lower cortisol level [14] and an increased level of antibodies [16]. Similarly, increased vagal tone has been associated with a higher attentional stability during exposure to stressful stimuli [17]. Together with our finding of structural differences in the medulla oblongata, this suggests a possible mechanism for the finding that regular practice of meditation can induce increased resistance to stressful stimuli, increased attentional skills [18,19], and the increased sense of calmness commonly reported by practitioners [20].

Increased gray matter intensity was also found in the left superior frontal gyrus and midventrolateral prefrontal cortex. Although the left superior frontal gyrus has been shown to be involved in self-relation [21] through introspection related activity and thereby self-awareness, there is substantial evidence that the mid-ventrolateral prefrontal cortex is critically involved in active retrieval of memories [22].

The first report of cortical structural changes associated with meditation [6] demonstrated a cortical thickening of the right anterior insula and of the right prefrontal cortex (BA 9/10). This is different from the finding of this study where the structural differences were found in the left forebrain [left mid-ventrolateral prefrontal cortex (BA 45A) and the superior frontal gyrus (BA 10)]. This difference in laterality may be attributable to a number of differences between the two studies.

First, the method for measuring cortical thickness in the earlier study [6] is a different measure than the voxel-based morphometry gray matter density and volume measures used in this study as well as in a number of recent studies of sustained attention [1,3,4,23].

Although both methods are likely to be sensitive to micro-structural changes in the dendritic tree such as arborization, dendrite density and perhaps spine density, a putative difference in the specificity and sensitivity to the underlying neuroanatomical changes needs further validation.

Second, the type of meditation differs. The participants in the earlier study practiced insight meditation, in which attention to breathing and sensory stimuli is a central element (CP) [6], whereas OA was the central aspect of the practice of the meditators studied here.

Extensive evidence that attentional practice can produce cortical structural changes in the human brain is found [3,4,23]. To our knowledge, this study is the first that specifically looks for and finds structural differences in the human medulla oblongata. The fact that we found structural differences in the brain stem of experienced meditators, indeed complete separation of the medulla oblongata gray matter densities observed in the two groups (Fig. 1), is in itself highly noteworthy because of the numerous projections from these autonomous control centers in the brainstem to the brain.

Despite the efforts to match the groups of normal volunteers by age, handedness, sex distribution, ethnicity,

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and general health status, it cannot be ruled out entirely that some other factor (e.g. socio-economic background) that was not explicitly targeted in the design of this study could have contributed to the differences reported. Further, it cannot be ruled out that the observed anatomical differences predated the long-term meditation practice because of a preference for engaging such a practice in the first place. However, we do not find this likely. The lack of correlation between structural differences and accumulated hours of practice in the highly experienced participants (examined range 8000–35 000 h) suggests a ceiling effect preceding the minimum of 8000 h of accumulated practice reported in this study. This is in accordance with the rapid neuroplasticity observed in respiratory control systems in the brain stem of several studies of nonhuman mammals [24] and longitudinal studies of structural neuroplasticity [3,4], which show that structural brain changes are detectable after only 3 months of attentional practice. Indeed, a recent study [25] has found changes in attention, anxiety, stress-related cortisol, and increased immunoreactivity after as little as 5 days of meditation practice. These findings indicate rapid effects of meditation on cognitive and emotional processing. Longitudinal studies of specific forms of meditation, however, need to be done to see how this is reflected in structural neuroplasticity.

Conclusion

This study indicates that at least one form of long-term meditation practice is associated with structural differences in the lower brain stem and the left forebrain as compared with controls.

Owing to the numerous projections from the lower brain stem to the entire brain this may account for many of the cardiorespiratory parasympathetic effects and traits, as well as the cognitive, emotional, and immunoreactive impact reported in studies of several different meditation practices.

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