Identifying cardiovascular neurocircuitry involved in the exercise pressor reflex in humans using functional neurosurgery

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In 1937, Alam and Smirk (2) reported the first experimental evidence in humans to support the hypothesis that a component of the cardiovascular responses to exercise was caused by a reflex response from working muscle. They showed that arresting the circulation from the exercising muscle resulted in maintained elevated arterial blood pressure in the recovery period from exercise, suggesting that the "byproducts" of muscle contraction accumulated within the muscle and somehow stimulated the afferent fibers. Direct evidence for the muscle pressor reflex came from studies in both anesthetized and decerebrate cats (14). Here contraction of the hindlimb muscles via stimulation of ventral roots caused a rise in arterial blood pressure that was abolished by sectioning the dorsal roots from the muscle. McCloskey and Mitchell (35) then went on to establish that the reflex was mediated by groups III and IV afferent fibers. Group III fibers were predominantly stimulated by the mechanical effects of muscular contraction, whereas group IV fibers were stimulated by the metabolic products of muscular contraction (25).

The afferent pathways of the exercise pressor reflex are well documented in animal studies. Specifically, the ventrolateral medulla (both the rostral and caudal areas) is known to have a role in modulating the reflex during static muscle contraction (3, 6). However, the involvement of higher central circuits has not been firmly established; although, the hypothalamus (1), the prefrontal cortex (5), the insular cortex (20), the cingulate cortex (8), and the thalamus (40) have either themselves been implicated in cardiovascular regulation or have been shown to receive projections from previously identified sites of cardiovascular regulation. Moreover, the subthalamic locomotor region of the posterior hypothalamus is a key area that integrates the cardiovascular response to muscle contraction and stimulated exercise (15, 43). There is also evidence from cats for a role of the periaqueductal gray (PAG) in the exercise pressor reflex (30, 46). Muscle contraction increases enkephalin release (46) and c-Fos activity in the PAG (30); moreover, C-Fos activity was identified in PAG neurons during dynamic treadmill exercise in rats (22).

The heterogeneous structure of the PAG is known to project to the ventrolateral medullary regions that control blood pressure (11, 12, 32), including the subretrofacial nucleus and the nucleus paragigantocellularis lateralis. In rats, the PAG also projects to higher centers involved in cardiac regulation, including the hypothalamus (21). Direct stimulation of the dorsal and lateral PAG in animal models increases arterial blood pressure and heart rate (10, 33). Similarly, in humans, stimulation of the dorsal PAG increases arterial blood pressure, whereas stimulation of the ventral PAG leads to hypotension (18). Substantial increases in neural activity in the PAG also occur during both anticipation of exercise and actual exercise in humans, suggesting that the PAG might be important in integration of the “central command” component of the cardiovascular response to exercise (17, 19).

The aim of this study was to investigate which sub-cortical structures are likely candidates that might integrate the exercise pressor reflex. We made use of the unique opportunity afforded by functional neurosurgery in humans involving the implantation of deep brain stimulating (DBS) electrodes in several deep brain nuclei that have previously been identified to be cardiovascular active in both animals and humans. Importantly, DBS allowed us to make local field potential (LFP) recordings from these nuclei to test the hypothesis that neural activity in...
subcortical structures is directly related to increases in arterial blood pressure (ABP) when evoked by actual exercise, and activation of the exercise pressor response following muscle occlusion.

MATERIALS AND METHODS

Ethical approval. Informed consent was obtained from patients, and the study was approved by the local hospital ethics committee (Oxfordshire REC C: 05/Q1605/47). The study conformed with the Declaration of Helsinki.

Patients. Fifty-one patients [30 men, 21 women, mean age 54.5 ± 10.2 yr (means ± SD)] who had undergone DBS were selected for this study (see online supplementary Table 1). Twelve patients had electrodes inserted in the PAG for the treatment of chronic pain (mean age 53.7 ± 12.4 yr). A further 13 patients had electrodes in the sensory thalamus, also for the treatment of chronic pain (mean age 50.8 ± 12.1 yr), either in the ventroposterolateral or ventroposteromedial nucleus for treatment of limb pain or facial pain, respectively. Another 13 patients had electrodes in the subthalamic nucleus (STN) for the treatment of Parkinson’s disease (mean age 60.2 ± 5.9 yr). One patient had an electrode in the inferior posterior hypothalamus for the treatment of cluster headaches (age 60.0 yr). Eighty patients had electrodes in the globus pallidus interna (GPi) for the treatment of generalized dystonia (mean age 56.8 ± 5.1 yr). Finally, four patients had electrodes in the anterior cingulate cortex for the treatment of chronic pain (mean age 45.0 ± 4.6 yr).

Surgical procedure. Our surgical procedure for localization of electrodes in the PAG has been described previously (18). In brief, preoperative magnetic resonance (MR) images of the patient’s brain were fused to a stereotactic computed tomography (CT) scan on the day of surgery using Image Plan software (Integra, Radionics, Burlington, MA; Ref. 38). Electrodes (Medtronic 3387, Medtronic, Minneapolis, MN) consisted of four cylindrical contacts of 1.5 mm, spaced at 1.5 mm. The electrodes were tunneled laterally and externalized for a week of testing and, if clinical effect was shown, were internalized and connected to the Kineta (Medtronic) implantable pulse generator that was positioned in a subcutaneous pocket in the chest wall 1 wk later under general anesthetic.

Postoperative localization of electrodes. The position of each electrode was mapped onto a brain atlas (34) using the postoperative MRI scans and the FMRIB Software Library. The scans were rotated so that the anterior and posterior commissures were on the same slice, and the mid-commissural point was calculated, followed by the relative coordinates of the electrode contacts in all three planes.

Experimental protocol. Experiments were performed 2–5 days after the first surgical procedure while the electrodes were externalized. Protocol 1 (Fig. 1A) was performed with 36 patients. Each patient had a 2-kg weight attached to their right wrist and were asked to sit comfortably for a rest period of 3 min. After the rest period, the patient was alerted to exercise by an oral cue. The patient then performed “light exercise” (bicep curls at a rate of 30–60/min) for 2 min, following which the patient was alerted to stop exercising and a blood pressure cuff was inflated around the exercised arm to above systolic pressure (~200 mmHg). The cuff remained inflated for a period of 2 min and was then released. This protocol was repeated three times.

We also wanted to examine the difference in neural activity between a normal exercise pressor reflex and an ischemic exercise pressor reflex, so we used five patients as single case studies and asked them to continue exercising during the period of cuff inflation (protocol 2; Fig. 1B). To establish whether increasing exercise intensity provides a direct increase in neural activity, we performed a third protocol (protocol 3; Fig. 1C) with 10 patients. This adhered to the same procedure as protocol 1, followed by repetition of the whole protocol with double the weight (4 kg). All patients were also asked to perform a set of control experiments during which the cuff was inflated without prior exercise; this was to ensure that neural activity observed during cuff inflation was not due to the sensation of the cuff inflation itself (protocol 4, Fig. 1D). Patients having treatment for pain disorders were asked to rate their pain levels using the Visual Analog Pain Severity Scale (VAPSS) to determine whether any changes in brain activity might actually be due to the patients’ pathology rather than exercise responses.

Experimental measurements. Noninvasive continuous finger arterial blood pressure (ABP) was measured with a Portapres (Finapres Medical Systems, Amsterdam, The Netherlands). Although we used the height correction unit, the pressure transducer and finger cuff were positioned at heart level to increase accuracy. Lead II electrocardiogram was recorded using disposable adhesive Ag/AgCl electrodes (H207PT, Kendall-LTP) and amplified ×1,000 (CED 1902, Cambridge Electronic Design, Cambridge, UK). The finger pressure and ECG were digitized at 4 kHz with 16-bit resolution (CED 1401 Mark II, Cambridge Electronic Design) using Spike II software (version 5.0, Cambridge Electronic Design).

LFPs were simultaneously recorded with bipolar configuration from the adjacent four circumferential contacts of each DBS macroelectrode; recordings were only analyzed from those contacts that provided relief from the symptoms of each patient’s respective disease. Signals were filtered at 0.5–500 Hz and amplified (>10,000) using isolated CED (1902) amplifiers and digitized using CED 1401 Mark II at a rate of 4 kHz (Cambridge Electronic Design). LFPs were then displayed online and saved onto hard disk using Spike II, as previously discussed by Green et al. (19).

Data analysis. The postoperative MRI or CT scan (fused to the preoperative MR) for each patient was used to determine the electrode location in each nucleus. For each of the trials for each patient, LFP data were broken into epochs of 30-s duration from each of “rest,” “2-kg exercise,” “4-kg exercise,” “occlusion,” “occlusion with exercise,” and “recovery.” To identify the fundamental spectral frequencies, a transformation of each of the 30-s epochs was performed from the time domain into the frequency domain by applying a fast Fourier Transform (FFT) algorithm offline using a digital spectrum analyzer (Matlab, version 6.5 MathWorks, Natick, MA). Signals were resampled at a rate of 2,500 Hz, and 50-Hz frequencies were filtered to avoid the inclusion of mains/line frequency artifact. A Hann window of 2 s in width was selected so that the signal could be carefully examined. The area under each power spectrum for each condition (e.g., rest, exercise, etc.) was calculated for the following frequency bands: 4–8 Hz, 8–12 Hz, 12–25 Hz, and 25–60 Hz. Local field potentials are traditionally divided into these bands, which were originally defined by prominent surface features of the spectrum and by spectral factor analysis and are thought to correlate with distinct behavioral states (7, 41). Mean arterial pressure (MAP) was calculated from the blood pressure trace and heart rate from the ECG trace for the same time data segments used in the LFP analysis. For statistical purposes, n refers to the total number of patients; separate trials were averaged in each patient and the obtained average was used as a single value. Any significant differences between the conditions were assessed by one-way ANOVA in SPSS (v 16.0, SPSS, Chicago, IL.). PSD data are presented as means ± SE.

RESULTS

Cardiovascular parameters. Figure 2 summarizes the changes for protocol 1 in cardiovascular parameters during exercise, occlusion, and recovery. Mean arterial pressure (MAP) increased during exercise by 13.8 ± 3.6% (P < 0.001, n = 25, t-test) and dropped slightly during occlusion, but was still significantly higher than resting levels, showing an increase from rest of 11.4 ± 1.9% (P < 0.001, n = 25, t-test). Recovery showed MAP returning to resting parameters. The R-R interval
decreased significantly during exercise \((P < 0.001, n = 36)\) and returned to resting parameters on occlusion.

**PAG area LFPs for protocol 1.** Figure 3 reveals the precise electrode locations of all six PAG patients who performed protocol 1, to give an idea of interpatient variability in electrode placement. This small variability in electrode placement probably accounts for the variability seen in the LFPs among patients. The PAG data presented in this study were recorded from the contacts that provided the patients with pain relief; these contacts were all located within the dorsal aspect of this nucleus. Figure 4 shows a typical electrode placement in a patient with a unilateral PAG electrode and changes in power spectral density (PSD) averaged across all six patients who performed protocol 1. ANOVA revealed that there was a difference among the conditions (rest, exercise, occlusion, and recovery, \(P < 0.05\)) for all patients individually. The major changes in the PAG occurred in the lower frequency bands. In the 4- to 8-Hz frequency band, occlusion was associated with an increase in PSD of 20.8 \pm 8.6\% \((P = 0.002, n = 6)\). In the 8- to 12-Hz frequency band, occlusion was associated with an increase in PSD of 29.4 \pm 15.7\% \((P < 0.001, n = 6)\). In the 12- to 25-Hz (beta) frequency band, occlusion was associated with a 29.5 \pm 10.2\% increase \((P < 0.001, n = 6)\).

**LFPs in other nuclei (thalamus, subthalamic nucleus, hypothalamus, globus pallidus interna, and anterior cingulate cortex) for protocol 1.** Figure 5 shows the PSD changes for all the other nuclei in patients following protocol 1 and typical electrode placements for some of the nuclei. ANOVA revealed no significant differences among any of the conditions for any of the patients, except for the 12- to 25-Hz frequency band in the STN patients. However, instead of an increase in PSD as observed in the PAG, there was a decrease in PSD; exercise
was associated with a reduction of $-38.6 \pm 3.1\%$ ($P < 0.001, n = 11$) and occlusion was associated with a reduction of $-12.7 \pm 3.2\%$ ($P < 0.001, n = 11$). There also appeared to be a peak in the power spectral density graphs for the GPi patients at $\sim 25$ Hz, but this peak was consistent among all conditions and all patients. It is likely that these were simply the resting activity levels of the nucleus.

**PAG area LFPs for protocol 2.** Figure 6 shows the changes in the three PAG patients who performed protocol 2 (the ischemic protocol), expressed as single case studies. In the first patient (Fig. 6, A–E), ABP increased during exercise by 24.5% and increased a further 4.4% during occlusion. With regards to PAG activity, significant changes were seen in the 4- to 8-Hz frequency band, where occlusion was associated with a 75.8% $P < 0.001$ increase. There was also an increase in the 8- to 12-Hz frequency band of 25.4% ($P = 0.018$) and in the 12- to 25-Hz band of 20.3% $P = 0.042$). In the second patient (Fig. 6, F–J), ABP increased during exercise by 9.8% and increased a further 6.5% during occlusion. In terms of PAG activity, significant changes were seen during occlusion in both the 4- to 8-Hz frequency band and the 8- to 12-Hz frequency band, an increase of 19.8% $P = 0.047$ and 72.8% $P < 0.001$, respectively. In the third patient (Fig. 6, K–O), ABP increased during exercise by 7.6% and increased a further 3.3% during occlusion. Significant changes in PAG activity were seen only in the 12- to 25-Hz frequency band, where exercise was associated with a 42.7% $P < 0.001$ increase, occlusion with a 27.7% $P < 0.001$ increase, and recovery with a 14.7% $P < 0.018$ increase in PSD ($P < 0.001$, and $P = 0.029$, respectively).

**PAG area local field potentials for protocol 3.** Figure 7 shows the changes in the three PAG patients who performed protocol 3 (the varied intensity protocol), again expressed as single case studies. In the first patient (Fig. 7, A–F), ABP increased by 22.7% during 2-kg exercise and remained elevated during occlusion, with an increase from rest of 13.5%. ABP also increased with 4-kg exercise by 25.5% and once more remained elevated during occlusion, with an increase from rest of 14.0%. With occlusion following 2-kg exercise, there was a significant increase of PAG activity in the 8- to 12-Hz frequency band of 21.8% ($P < 0.001$) and a decrease during recovery by 15.6% $P = 0.036)$. With occlusion following 4-kg exercise, there was an increase of 20.9% $P = 0.009$) in the 4- to 8-Hz band, and in the 8- to 12-Hz band, there was an increase of 67.7% $P < 0.001$. For 4-kg exercise itself, there was a significant increase in PAG activity by 27.8% $P = 0.001$ in the 12- to 25-Hz frequency band.

In the second patient (Fig. 7, G–L), ABP increased by 23.9% during 2-kg exercise and remained elevated in the subsequent period of occlusion, with an increase from rest of 18.7%. ABP also increased with 4-kg exercise by 23.8% and increased a further 7.4% during the subsequent period of occlusion. With occlusion following 2-kg exercise, there was an increase of 16.0% $P = 0.004$ in the 8- to 12-Hz frequency band. With occlusion following 4-kg exercise, there was an increase of 19.3% $P < 0.001$ in the 4- to 8-Hz frequency band, an increase of 8.1% $P < 0.001$ in the 8- to 12-Hz band ($P = 0.044$), and an increase of 17.9% $P < 0.001$ in the 12- to 25-Hz frequency band.

**Fig. 2. Changes in cardiovascular parameters with exercise and occlusion.** A: blood pressure changes for protocol 1 (normal protocol; $n = 25$). It should be noted that the $n$ value here is not 36, due to failure of the Portapres with 11 of the patients. B: changes in R-R interval for protocol 1 (normal protocol; $n = 36$). ***$P < 0.001$.

**Fig. 3. Electrode locations for periaqueductal gray (PAG) patients who performed protocol 1 (normal protocol).** Schematic diagram showing the contacts recorded only from those contacts that provided pain relief—these are all within the dorsal aspect of the PVG/PAG (highlighted in red). AC, anterior commissure; MCP, mid-commissural point; PC, posterior commissure; PVG, periventricular grey; SC, superior colliculus; RN, red nucleus.
25-Hz band. During 4-kg exercise itself, there was a significant increase in PAG activity of 9.9 \%/0.3% (P \%/0.002).

In the third patient (Fig. 7, M-R), ABP increased by 13.6% on 2-kg exercise and remained elevated on occlusion, with an increase from rest of 11.4%. ABP also increased during 4-kg exercise by 17.7% and again remained elevated on occlusion, with an increase from rest of 12.3%. For the period of occlusion following 2-kg exercise, there was a significant increase of PAG activity in both the 4- to 8-Hz and 8- to 12-Hz frequency bands, of 38.8 \%/7.9% (P \%/0.001) and 61.9 \%/15.4% (P \%/0.001), respectively. For the period of occlusion following 4-kg exercise, there was an increase of 32.3 \%/10.7% (P \%/0.001) in the 4- to 8-Hz band, an increase of 106.0 \%/21.4% (P \%/0.001) in the 8- to 12-Hz band, and an increase of 29.3 \%/21.7% (P \%/0.043) in the 12- to 25-Hz band. With 4-kg exercise itself, there was a significant increase in PAG activity of 23.9 \%/5.3% (P \%/0.014) in the 8- to 12-Hz band and of 65.0 \%/23.7% (P \%/0.001) in the 12- to 25-Hz band.

Table 2 (see online supplementary data) shows the quantitative data for all the other patients who performed protocols 2 and 3. There were no significant differences between any of the conditions for the thalamus, GPi, and one of the two STN patients performing protocols 2 and 3. The other STN patient performing protocol 3 exhibited a significant decrease in PSD of -20.4 \%/5.3% (P \%/0.001) and -17.4 \%/3.7% (P \%/0.001) in the 4- to 8-Hz frequency band during the 2- and 4-kg exercise periods, respectively. There were no significant differences between the conditions of rest, occlusion without prior exercise, and recovery in the patients performing protocol 4, which was the control protocol (Fig. 8), in neither the cardiovascular parameters nor the neural activity.

It should be noted that patients suffering from chronic pain were asked to rate their pain levels during all protocols, using the VAPSS, and there were no changes from resting level pain in any of the patients during the protocols.

DISCUSSION

The new findings presented here are that 1) the exercise pressor reflex is associated with a significant increase in dorsal PAG activity. This response is graded to exercise intensity and is consistent with the hypothesis that the PAG is directly involved in the neurocircuitry of the reflex. 2) The other nuclei tested, some of which are known cardiovascular areas, appear not to be involved in the integration of this reflex. 3) The sensation of the cuff inflation alone does not result in increases in PAG activity.

Role of the PAG during exercise. Our findings revealed a marked increase in PAG neural activity during occlusion of the exercised limb across the 4- to 8-Hz, 8- to 12-Hz, and 12- to 25-Hz frequency bands, and this was also seen during ischemic exercise (protocol 2). When the exercise drive was increased using double the weight (protocol 3), neural activity also increased; it was not only more widespread across the frequency bands, but also significantly more elevated in terms of absolute PSD values. This trend is similar to the findings of...
Green et al. (19), who had their patients exercising at a much higher intensity during cycle ergometry compared with the small muscle mass exercise in our patients and saw significant PAG activity even in the 25- to 60-Hz and 60- to 90-Hz bands. Higher drive and effort appears to be associated with increased PAG activity over a wider range of frequency bands. It should be noted that in some of our experiments, particularly when only 2 kg was used during the exercise periods, the pressor reflex may not have been activated, as the exercise was very light. This manifests itself in the fact that PAG neural activity did not usually increase during this “light” exercise, but only during the following period of occlusion. In contrast, the heavier exercise with a 4-kg weight resulted in a clear increase in dorsal PAG activity that was associated with higher ABP during the period of occlusion.

The PAG is a major neural structure for autonomic regulation, in particular cardiovascular changes associated with integrated behavioral “defense” responses. It is a heterogeneous structure, divided into five columns—dorsomedial, dorsolateral, lateral, ventrolateral, and ventromedial—with each area eliciting a distinct autonomic response. In rats, stimulation of the ventral PAG is associated with a reduction in arterial blood pressure (ABP) and analgesia (23). In humans, the degree of analgesia induced by stimulation of the rostral PAG is related to the magnitude of reduction in ABP. Microinjection of excitatory amino acids to the dorsolateral and lateral columns in cats and rats evokes tachycardia and hypertension (13), whereas excitation of the ventrolateral column evokes bradycardia and hypotension (9). Similarly, in humans, electrical stimulation of the dorsal periventricular gray (PVG; continuous with the PAG) causes an increase in systolic blood pressure. In contrast, stimulation of the ventral PVG causes a reduction in systolic blood pressure (18). When our results are taken together with the known effects of electrical stimulation of the dorsal PVG-PAG in humans and that of excitation of the lateral PAG in animals, they provide a framework that strongly suggests this part of the PAG is a key aspect of the neurocircuity that integrates the exercise pressor reflex. When the
Fig. 6. Local field potential changes in the PAG for protocol 2 (ischemic protocol). A: raw data trace for first patient. B: postoperative image (postoperative CT fused to preoperative MR) for first patient showing a unilateral electrode in the right PAG. C: blood pressure changes for first patient. D: mean power spectral density for first patient. E: normalized spectral changes (rest = 1.0) divided into frequency bands. F: raw data trace for second patient. G: postoperative image for second patient showing a unilateral electrode in the left PAG. H: blood pressure changes for second patient. I: mean power spectral density for second patient. J: normalized spectral changes divided into frequency bands. K: raw data trace for third patient. L: postoperative image for third patient. M: blood pressure changes for third patient. N: mean power spectral density for third patient. O: normalized spectral changes divided into frequency bands. *P < 0.05, **P < 0.01, ***P < 0.001 (significance was calculated using ANOVA on several data points collected from each individual patient.)
reflex is evoked in exercising animal models, c-Fos expression increases in PAG neurons (22). Moreover, direct electrical stimulation of the ventral roots supplying the hindlimb muscles of the cat increases PAG activity (27).

What is the signaling pathway that links the muscle afferent pathway to the PAG? Williams et al. (44–46) have suggested that catecholaminergic-opioidergic interaction in the PAG may integrate the peripheral sensory traffic from working muscle.
Clonidine (an α2 agonist) attenuates the muscle pressor reflex when injected into the dorsolateral PAG (44, 45); an effect that was prevented by naloxone, which suggests an interplay with the adrenergic-opioidergic system (45). There is also convincing evidence that immunoreactive enkephalinergic substances are released in the dorsolateral PAG in the cat during isometric contractions (46). This may provide a transmitter link to excitation, since it is well established that the PAG projects to the classical cardiovascular sites in the medulla (12, 32). Importantly, lesions in the PAG abolish the rise in blood pressure caused by muscle contraction (48). Similarly, ventrolateral medullary lesions block the cardiovascular responses that are evoked by stimulating the dorsal PAG in rats (32).

**Role of the subthalamic nucleus and other nuclei during exercise.** The observation that the activity of the subthalamic nucleus (STN) decreased during exercise and occlusion in the 12- to 25-Hz (beta) frequency band is consistent with much evidence that showed STN suppression following warning and “go” cues (19, 29, 49). The STN is a known cardiovascular active area. Stimulation results in increased MAP and heart rate and also facilitation of movement (42); however, our data suggest that it is not a major integrating site of the exercise pressor reflex itself. Other sites we tested that are known to be cardiovascular active (42) were also not activated during the pressor reflex.

**Central command and muscle pressor reflex integration.** The idea of linking the central and peripheral nervous systems during exercise has been around for a long time since the classical studies of Krogh and Lindhard (28), Alam and Smirk (2), Coote et al. (14), McCloskey and Mitchell (35), and Goodwin et al. (16). When all of the detailed neurocircuitry in small animal models and measurements of neuropeptides from these circuits during simulated exercise are viewed together, the PAG appears to be a key site of integration, in particular the dorsolateral PAG in animals (4, 21, 32) and the dorsal PVG/PAG in humans (19). Although this is not the “command” area itself, it appears to have all the hallmarks of being a major integrating site, since destruction of the PAG prevents the rise in arterial blood pressure caused by muscle activation (48).

**Limitations.** Do changes in spectral power directly translate into changes in neural activity? Since local field potentials (LFPs) are likely to be a consequence of local synchronous neuronal population activity (36, 39), as opposed to isolated spikes (asynchronous activity), the increase in PAG activity as measured by LFPs during the pressor reflex probably reflects increases in network synchrony or network size (24, 26, 31). However, the physiological significance of the various frequencies remains to be established and is beyond the scope of this study.

It could be argued that it cannot be determined from the data whether the increase in PAG activity is causal to the exercise pressor reflex, is compensatory to the pressor reflex, or is simply coincidental to perceptual aspects of the exercise regimen. First, it is highly unlikely that the activity is simply coincidental, as the increase in neural activity was only observed during occlusion following exercise, since there was no increase in PAG activity with occlusion alone. Second, there is evidence to reject the idea that the increase in activity was compensatory to the pressor response; increasing ABP in cats with phenylephrine does not change c-Fos immunoreactivity in the PAG, whereas it did increase immunoreactivity in other cardiovascular brain stem areas (47). Others, however, re-

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**Fig. 8.** Local field potential changes in the PAG for protocol 4 (control protocol). A: blood pressure changes for protocol 4 (control protocol; n = 12). B: mean power spectral density for all 12 patients. C: normalized spectral changes (rest = 1.0) divided into frequency bands. n = 12.
ported that both phenylephrine and nitroprusside produced extensive Fos expression in the PAG (37). However, the increase in PAG activity was only seen during the occlusion period. It is therefore highly unlikely that it is secondary to the increase in ABP, since there was no difference in neural activity during the increase in ABP accompanying the exercise period. Moreover, in humans, direct stimulation of the dorsal PAG causes an increase in arterial blood pressure (18), and we have shown here that the magnitude of the pressor reflex and PAG activity is graded to exercise intensity.

Another limitation of this study is that we are dealing with a patient model that has an underlying pathology. Although PAG patients have an idiopathic presentation for pain, there is no evidence that the PAG, or the autonomic nervous system, are dysfunctional in these patients. The nature of this study also limits the specific nuclei that can be recorded from, since sites were targeted for best therapeutic outcome given each patient’s pathology. However, the areas we chose have been well described in the animal literature as being cardiovascular active. The size of the electrode is rather large given some of the areas we targeted, and it is difficult to be completely precise about the location without histology. However, the neurosurgeons placed them using precise stereotactic coordinates, and for the PAG electrodes, we performed postoperative electrode localization analysis to further confirm their positions within the dorsal aspect of the PAG. Finally, the particular type of exercise undertaken and its low intensity probably underestimated the full range of the power increase that is potentially available from the PAG. This was due to the fact that the patients did not feel capable of performing very high-intensity exercise so soon after surgery.

In conclusion, the direct neurophysiological recordings from this study suggest that the dorsal PAG is a key site for integrating the exercise pressor reflex in humans.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

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