

# Neural activation during acute capsaicin-evoked pain and allodynia assessed with PET

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## Summary

The PET H<sub>2</sub><sup>15</sup>O-bolus method was used to image regional brain activity in normal human subjects during intense pain induced by intradermal injection of capsaicin and during post-capsaicin mechanical allodynia (the perception of pain from a normally non-painful stimulus). Images of regional cerebral blood flow were acquired during six conditions: (i) rest; (ii) light brushing of the forearm; (iii) forearm intradermal injection of capsaicin, (iv) and (v) the waning phases of capsaicin pain; and (vi) allodynia. Allodynia was produced by light brushing adjacent to the capsaicin injection site after ongoing pain from the capsaicin injection had completely subsided. Capsaicin treatment produced activation in many discrete brain regions which we classified as subserving four main functions: sensation–perception (primary somatosensory cortex, thalamus and insula); attention (anterior cingulate cortex); descending pain control (periaqueductal grey); and an extensive network related to sensory–motor integration (supplementary motor cortex, bilateral putamen

and insula, anterior lobe and vermis of the cerebellum and superior colliculus). Comparison of the noxious and non-noxious stimuli yielded several new insights into neural organization of pain and tactile sensations. Capsaicin pain, which had no concomitant tactile component, produced little or no activation in secondary somatosensory cortex (SII), whereas light brushing produced a prominent activation of SII, suggesting a differential sensitivity of SII to tactile versus painful stimuli. The cerebellar vermis was strongly activated by capsaicin, whereas light brush and experimental allodynia produced little or no activation, suggesting a selective association with C-fibre stimulation and nociceptive second-order spinal neurons. The experimental allodynia activated a network that partially overlapped those activated by both pain and light brush alone. Unlike capsaicin-induced pain, allodynia was characterized by bilateral activation of inferior prefrontal cortex, suggesting that prefrontal responses to pain are context dependent.

**Keywords:** nociception; neuropathic pain; capsaicin; regional cerebral blood flow; neuroimaging

**Abbreviations:** BA = Brodmann area; SI = primary somatosensory cortex; SII = secondary somatosensory cortex

## Introduction

Pain, in its most elementary representation as a nociceptive spinal reflex, may be considered a fairly uncomplicated system (Sherrington, 1947). However, within the brain, pain is a complex, multi-dimensional phenomenon that influences a wide variety of nervous system functions. These range from sensory-discriminative and affective-motivational components to motor integratory responses and may extend to influences on neuro-immune function in chronic pain conditions. Thus, identifying the multiple neural networks that subserve these functional responses and harnessing this knowledge to manipulate the pain response in new and beneficial ways are challenging tasks. The objectives of the

present study were to examine in normal volunteers (i) the neural networks activated during a strong acute pain stimulus via selective stimulation of primary afferent C-fibres by capsaicin and (ii) potential alterations in these pain network(s) during experimentally induced abnormal pain processing.

The various dimensions of the pain experience can be influenced by the intensity of the stimulus, its duration and the location of the pain on, or within, the body (cutaneous, muscular or visceral). These quantitative and qualitative factors have only been partially explored in previous PET studies of experimental pain in normal volunteers (Jones *et al.*, 1991; Talbot *et al.*, 1991; Casey *et al.*, 1994; Coghill

*et al.*, 1994; Hsieh *et al.*, 1995) which evoked pain via noxious heat delivered repetitively to the surface of the skin by a contact thermode. We used a different pain stimulus, the intradermal injection of a pain-producing chemical (capsaicin), which has no concomitant repetitive tactile component and only one injection is given. Capsaicin, which is extracted from hot chili peppers, directly activates nociceptive primary afferent C-fibres (Schmidt *et al.*, 1995; Baumann *et al.*, 1991). It is an undeniably effective nociceptive agent, yet, as an experimental pain stimulus, it circumvents the potential for tissue damage inherent with painfully hot thermal stimuli. Thus, capsaicin provided a pain stimulus with minimal contributions from other somatosensory modalities and allowed us to compare painful and tactile stimuli with no overlap between the primary afferents subserving these modalities. In the CNS, however, we hypothesized that there would be some degree of overlap between the regions activated by the two somatosensory stimuli.

In addition to providing a strong acute pain stimulus, capsaicin transiently induces a variety of sensory abnormalities including hyperalgesia and allodynia (Simone *et al.*, 1989, 1991; LaMotte *et al.*, 1991). Allodynia is a very distressing painful syndrome frequently encountered in patients suffering from a variety of chronic pain conditions such as postherpetic neuralgia, peripheral neuropathies and reflex sympathetic dystrophy (Gracely *et al.*, 1992; Bennett, 1994). The painful sensory abnormalities that occur in the post-capsaicin period have been used to investigate peripheral and spinal components contributing to hyperalgesia and allodynia, and form the basis of an acute, reversible human model for neuropathic pain (Park *et al.*, 1995). In this model, large secondary zones of hypersensitivity occur in the skin around the focal injection site. Within these zones, subjects report abnormally strong pain to light pinpricks, painful sensations to light brushing and non-noxious von Frey hair stimuli, and hyperalgesia to thermal stimulation. The period of hyperalgesia and allodynia (0.5–2 h) is dose-dependent and the effect is completely reversible. The ability of capsaicin to induce this altered sensitivity provided an opportunity to examine higher CNS correlates of both acute pain and abnormal pain processing in normal subjects. The present PET data indicate that the capsaicin stimulus immediately activated a widespread network of brain regions. A large, but only partially overlapping, set of regional activations occurred during the subsequent capsaicin-induced allodynia.

## Methods

### Subjects

The study was performed in accordance with a clinical protocol approved by the Institutional Review Board of the National Institute of Dental Research and within the guidelines set by the National Institutes of Health Radiation Safety Branch. All subjects gave their written informed consent for the procedure. Thirteen healthy subjects, aged 24–50 years

(five females and eight males) were studied. All were right hand dominant. None were taking any medication at the time of the study. Subjects were given a test dose of capsaicin (250 µg in 20 µl, intradermal) ~1 week prior to the PET scan to familiarize the subjects with the stimulus and to screen out subjects who might find it too uncomfortable. At this time the subjects were also tested for the presence of hyperalgesia and allodynia, although not with the same prolonged time interval as in the PET portion of the study. All subjects remained in the study after the screening dose.

All subjects received conventional MRI scans consisting of a set of 6.5 mm thick images co-planar with the PET slices and a volumetric scan of 1.2 mm thick slices. The MRI scans were used to screen for neurostructural abnormalities and for positioning the subjects in the scanner such that the arm area of primary somatosensory cortex (SI), which is located superficially and superiorly in the brain, was within the field of view of the PET scanner. Final positioning was made by comparison with the reconstructed Ge/Ga transmission scan obtained in the PET scanner.

### Activation stimuli and scan sequence

Each subject underwent six to seven scans. All scans were conducted with the eyes closed, the room lights dimmed, and with minimal noise in the room. All stimuli were administered to the left volar forearm and the bolus of radioactive H<sub>2</sub><sup>15</sup>O was administered into the right antecubital vein at the same time as the stimuli were begun. Brushing stimuli were continued throughout the 1 min scan. The scans consisted of, in order: (i) an eyes-closed, resting state; (ii) light brushing to the left volar forearm; (iii) acute capsaicin pain via intradermal injection of 250 µg in 20 µl vehicle using a 27 gauge needle; (iv) waning phase of capsaicin when some residual ongoing pain was present (Wane1); (v) little or no residual ongoing pain (Wane2); and (vi) mechanical allodynia induced by light brushing adjacent to the site of capsaicin injection. Following the Wane2 scan, scans were paused until spontaneous pain had subsided completely or nearly so (i.e. no report of consistent pain) before beginning the allodynia scan. Six of the subjects received an extra resting state scan which was administered between the light brushing and the capsaicin scan.

### Instructions

Subjects were instructed to hold still for the entire period and told to close their eyes ~2 min prior to the scan and to keep them closed until the 1-min scan was finished. At the time they were told to close their eyes, subjects also were told what the stimulus would be for the upcoming scan, and they were reminded to remain still. With one exception, subjects were motionless following the capsaicin injection. The data from this subject were not included in the analysis.

### **Pain rating**

Pain was rated in the scanner using a proportional rating method in which the magnitude of the initial capsaicin pain was set at 100 and proportional comparisons were made thereafter. Ratings were obtained immediately after the capsaicin scan and before and after each of the remaining scans.

### **Image acquisition**

Scanning was performed with the use of a Scanditronix PC2048-15B dedicated head PET scanner which collected 15 contiguous planes with a slice thickness and in-plane resolution of 6–6.5 mm full-width, half-maximum after reconstruction. The emission images were corrected for attenuation with a transmission image collected using a rotating Ge<sup>68</sup> source. Subjects were placed in a comfortable supine position for the duration of the experiment and head motion was reduced with the use of an individually fitted thermoplastic mask that was attached to the scanner bed. The lowest scan plane was aligned parallel to, and between 16 and 22.5 mm above, the canthomeatal line. Measurements of changes in CBF were obtained using previously described techniques with minor modifications (Raichle *et al.*, 1987; Herscovitch, 1993). Images of CBF were generated by summing the activity during the 60-s period following the first detection of an increase in cerebral radioactivity after the intravenous bolus injection of 42.5–50 mCi of H<sub>2</sub><sup>15</sup>O. No arterial blood sampling was performed and, thus, the images collected are those of tissue activity. Tissue activity recorded by this method has been shown to be linearly related to regional cerebral blood flow (Fox *et al.*, 1984; Fox and Mintun 1989) and a large literature attests to the fact that, in the absence of major pathology, such increases in regional blood flow reflect increases in neuronal metabolism and neuronal activity. Mean tissue activity for the group was adjusted to a value of 50. Results are therefore reported as adjusted blood flow.

### **Image analysis**

Functional localization and assessment of stimulus-related activations were determined with stereotactic normalization followed by statistical parametric map generation (Friston *et al.*, 1990, 1991). This approach allows inter-subject averaging and quantitative assessment of change significance. In the stereotactic normalization process, the scans were individually re-oriented, linearly re-scaled and re-formatted to generate a stereotactically normalized image of 26 planes parallel to the intercommissural plane with an interslice separation of 4 mm (Friston *et al.*, 1991). This set of planes corresponds to the stereotactic atlas of Talairach and Tournoux (1988). Analysis of change significance was performed on the planes common to all subjects, covering a region extending from ~16 mm above the canthomeatal line in the most inferior slice, to the most superficial slice obtainable in

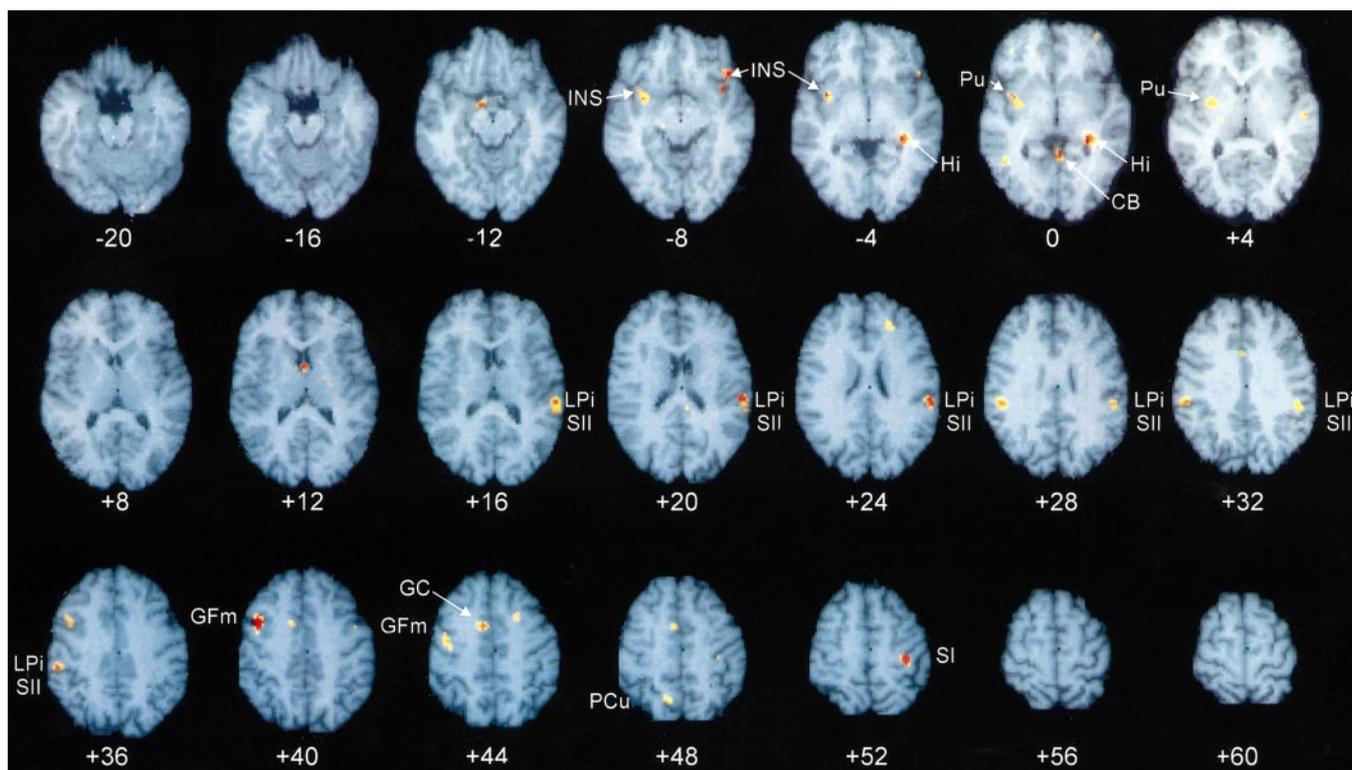
cerebral cortex. The latter was the main factor we considered in positioning the subjects, since the arm area in SI is located very superiorly. The analysis of these planes consisted of a pixel-by-pixel analysis of covariance to remove the effects of differences in global cerebral activity (Friston *et al.*, 1990), followed by planned linear comparisons of the adjusted mean images. The four comparisons were: capsaicin versus resting state; brushing versus resting; allodynia versus resting; and allodynia versus brushing (i.e. to remove the 'brush component' from the allodynia). The value of *t* for each pixel in each comparison was calculated and then transformed to a normal standard distribution. All regions reported as being significantly activated exceeded the *P* < 0.005 level of significance. The individual contrasts (e.g. capsaicin versus resting) were then searched for local maxima and minima (Figs 1–4). The present study focuses on increases in relative regional cerebral blood flow because at present no consensus exists on the exact neurophysiological interpretation of relative decreases in regional cerebral blood flow. This is best examined by determination of absolute regional cerebral blood flow (measured with arterial lines) to ascertain whether such decreases are not an artefact of the normalization (to a mean value of 50) of the count data.

A region-of-interest analysis was performed to examine regional activity in the Wane1 and Wane2 scans in comparison with the capsaicin scan using the local maxima from the capsaicin scan as the point of reference. Each local maximum formed the centre of an 11-voxel volume (nine in-plane and one above and one below the centre voxel). Data from the Wane1 scan for several representative regions are shown in Fig. 5. Data visualization and map generation were performed with MEDx (Sensor Systems, Sterling, Va., USA). The significant increases in relative blood flow are displayed on a individual MRI that had been spatially normalized into standard stereotaxic space (Figs 1–4). The stereotaxic coordinates of the local maxima are presented in tabular form (Tables 1–4).

## **Results**

### **Pain rating**

All of the subjects stated that the capsaicin injection was painful. The subjects were told to assign the capsaicin pain as 100% and subsequent pain was rated as a proportion of the acute capsaicin pain. Capsaicin pain decreased exponentially: from 1 to 2 min, 2 to 4 min and 8 to 12 min post-capsaicin. The mean ( $\pm$  SEM) pain rating progressively diminished to  $53 \pm 9$ ,  $44 \pm 10$  and  $22 \pm 7$  out of 100, respectively. The rating after the Wane1 scan, from 13.5 to 15 min after capsaicin injection, was  $14 \pm 2$ . After the Wane2 scan, ~25 min after capsaicin injection, the rating was  $4.6 \pm 1.1$ . This time course, of an initial rapid decrement and slow resolution for pain after intradermal capsaicin, is similar to published descriptions (Simone *et al.*, 1989). Thus, the allodynia scan was performed between 36 and 53 min after



**Fig. 1** Light brushing compared with the resting state. Twenty-one axial brain slices are shown, arranged from inferior (upper right, slice -20) to superior (lower left, slice +60); the slice numbering refers to the horizontal stereotaxic zero in the atlas of Talairach and Tournoux (1988). Abbreviations are from the same source. Activations are superimposed on an MRI that has been stereotaxically normalized. Z-values  $>2.33$  are displayed in yellow; higher scores are shaded from yellow to red-orange; the highest Z-score in this data set was 3.49 ( $P < 0.0005$ ). Activations (inferior to superior) were detected in: insula (INS, bilaterally, slices -8 and -4) hippocampus, (Hi, contralateral, slices -4 and 0); putamen (Pu, ipsilateral, slices 0 and +4); SII/inferior parietal lobule, BA 40 (LPi/SII bilaterally, slices +16 to +36); middle frontal gyrus (GFm, BAs 8 and 6, ipsilateral, slices +36 to +44); cingulate gyrus (GC, BA 24, midline/ipsilateral, slices +40 to +48); posterior parietal cortex (PCu, precuneus, BA 7, ipsilateral, slice +48); and contralateral SI (slice +52).

the injection of capsaicin and occurred upon a background of little or no spontaneous pain. The average allodynia pain rating was  $18 \pm 4.7$  (see graph inset in Fig. 5B).

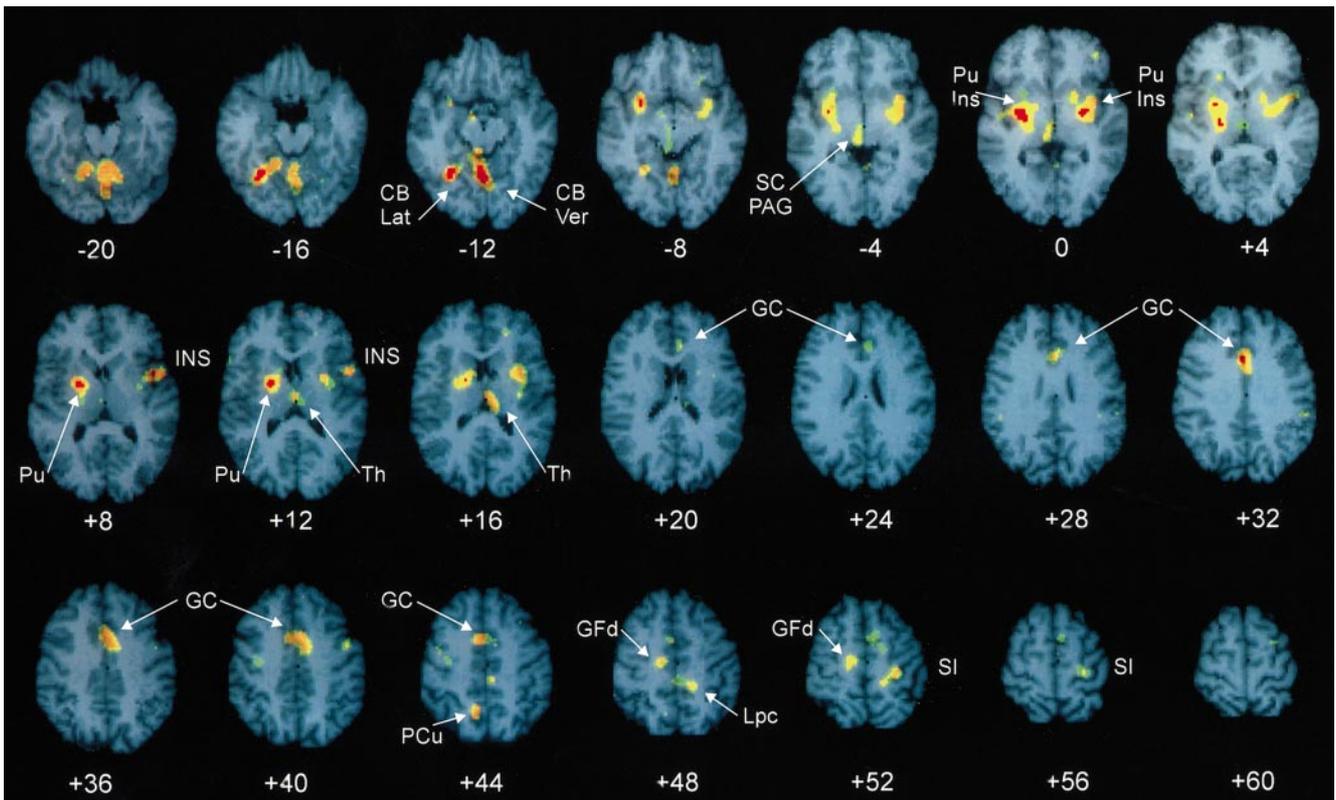
### Light brush activation

Light brushing of the left forearm caused increases in relative blood flow in sensory-related regions and other areas such as the hippocampus (Table 1, Fig. 1). Each brain slice is referred to by the superior-inferior level in mm relative to the intercommissural line. Somatosensory regions that displayed activation included the contralateral SI (slice +52) and secondary somatosensory cortex (SII) both ipsi- and contralaterally, extending from the parietal operculum into the inferior parietal lobule (slices +16 to +36). Five of the remaining regions showed prominent activation in at least two contiguous slices: contralateral hippocampus (slices -4 and 0), anterior cingulate gyrus [Brodmann area (BA) 24, slices +44 and +48], ipsilateral middle frontal gyrus, overlapping both lateral supplementary motor cortex and BA 8 which is the frontal oculomotor field (slices +36 to +44), ipsilateral putamen (slices 0 and +4) and ipsilateral insular cortex (slices -8, -4 and 0). We did not see activation in

contralateral ventroposterolateral thalamus. Nevertheless, the contralateral cortical projection field in SI was clearly activated (slice +52).

### Capsaicin pain activation

Capsaicin produced extensive regional activations in areas ranging from the mesencephalon to the cerebral cortex (Table 2 and Fig. 2). In Fig. 2, note that areas containing contiguous activity at multiple levels may be labelled only in representative slices. The most statistically robust activation was detected in the cerebellar vermis (slices -20 to -8). Activation also occurred at a more lateral focus in the ipsilateral cerebellum and medially within the deep cerebellar nuclei (Fig. 2, levels -20 to -8). The next set of prominent activations were bilateral foci within the putamen/globus pallidus (levels -4 to +8). A third region of prominent activation was the anterior cingulate gyrus in which activity was detected at multiple loci bilaterally, starting from the genu of the corpus callosum and continuing superiorly (slices +16 to +44). These regions encompass BAs 23, 24, 32 and 33. Insular cortex was activated bilaterally (slices -8 to +16). While the activations in the insula and putamen appear to



**Fig. 2** Capsaicin injection compared with the resting state. Conventions as in Fig. 1. Z-values  $>2.33$  are displayed in yellow; higher scores are shaded from yellow to red. The highest Z-score in this data set was 4.83 ( $P \leq 0.0001$ ). Activations were detected in: cerebellar vermis and ipsilateral anterior lobe (CB Ver and CB Lat, respectively, slices  $-20$  to  $-8$ ); midbrain tectum and tegmentum coincident with the superior colliculus, periaqueductal grey and red nucleus (SC, PAG, midline/ipsilateral, slices  $-12$  to  $0$ ); insular cortex (INS, bilaterally, slices  $-4$  to  $+8$ ); putamen/globus pallidus [Pu, bilaterally, slices  $-4$  to  $+16$ ; note that, in this display, the putamen and insular cortex appear to merge (e.g. slices  $-4$  to  $+4$ ) but are spatially distinct (see Table 2)]; thalamus (Th, midline/contralateral, slices  $+12$  to  $+16$ ); cingulate gyrus (GC, activation occupied several foci, starting in slice  $+20$  and extending to slice  $+48$ ); posterior parietal cortex (precuneus, PCu, BA 7, ipsilateral slice  $+44$ ) and lobulus paracentralis (Lpc, BA 5 contralateral, slice  $+48$ ); contralateral SI (slices  $+52$  and  $+56$ ), supplementary motor cortex (gyrus frontalis medialis, GFd, BA 6, ipsilateral, slices  $+48$  to  $+52$ ). The capsaicin pain stimulus did not induce substantial activity in SII except for two small foci in slices  $+28$  and  $+32$  ipsi- and contralaterally.

merge in several slices (Fig. 2), the coordinates for activation in each region are distinct (Table 2).

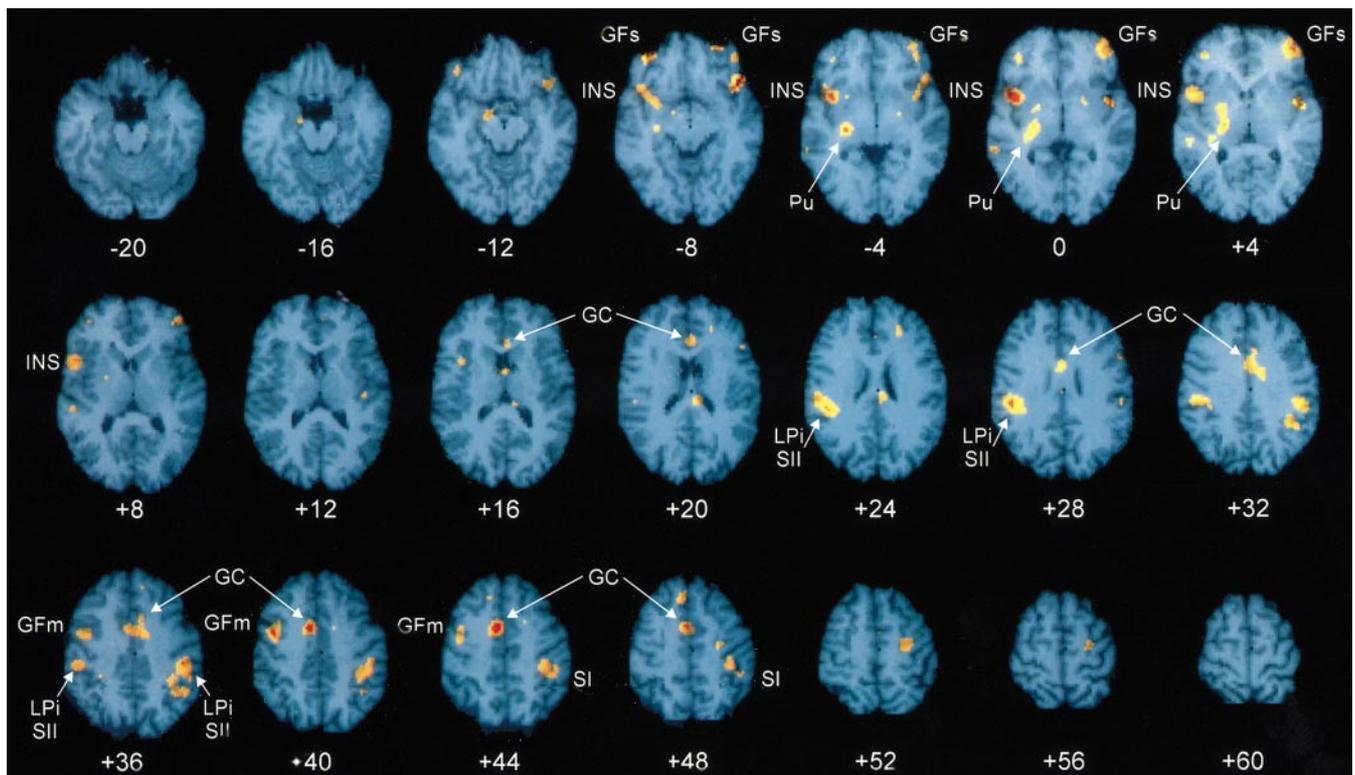
The cerebral cortical regions activated included: contralateral SI (two slices at  $+52$  and  $+56$ ); two sites in posterior parietal cortex, precuneus (ipsilateral, BA 7, slice  $+44$ ) and lobulus paracentralis (BA 5, contralateral medial surface, slices  $+44$  and  $+48$ ); and the supplementary motor cortex (BA 6, ipsilateral middle frontal gyrus, slices  $+48$  and  $+52$ ). Thalamic activity was found mainly contralaterally and at the midline, and somewhat superiorly within the thalamus (slices  $+16$  and  $+12$ ), corresponding to multiple thalamic nuclei, including the dorsomedial nucleus, lateral dorsal nucleus, anterior group, midline and dorsomedial pulvinar. As with the brush condition, a distinct focus in ventrobasal complex (e.g. slices  $+8$  and  $+4$ ) was not detected with the capsaicin stimulus. Lastly, at the level of the mesencephalon, activity was detected at locations in both the periaqueductal grey and superior colliculi (slices  $-12$  to  $0$ ). A third focus in slice  $0$  was assigned to the ipsilateral red nucleus; however, in the Talairach and Tournoux (1988) atlas the transition

from mesencephalon to diencephalon is fairly abrupt, and a location in ipsilateral thalamus is also possible.

The regional map of capsaicin-induced pain activation was notable for the near absence of activation in SII/inferior parietal lobule. Close inspection of slices  $+28$  and  $+32$  (Fig. 2) disclosed several small foci of activity bilaterally in this region with the contralateral side predominating. However, when compared with the activity obtained with the light brush stimulus, they are much smaller in size, extend over fewer axial slices and have a lower Z-score (Tables 1 and 2).

### Experimental allodynia

Comparison of allodynia with the resting state (Table 3 and Fig. 3) revealed several areas in common with either the capsaicin or light brush conditions. These include the bilateral activations in inferior parietal lobule/SII (BA 40) (slices  $+16$  to  $+36$ , see same slices for brush in Fig. 1), more superiorly located activations in SI (also with brushing and capsaicin, slices  $+52$  and  $+56$ ), more superiorly located regions in



**Fig. 3** Allodynia compared with the resting state. Conventions as in Fig. 1, legend. Z-scores  $>2.33$  are displayed in yellow; higher scores are shaded from yellow to red-orange; the highest Z-score in this data set was 4.54 ( $P < 0.0001$ ). Activations were detected in: superior frontal gyrus (GFs, BA 10, bilaterally, slices  $-8$  to  $+4$ ); insula (INS, bilaterally, slices  $-8$  to  $+8$ ; this also includes portions of the inferior frontal gyrus, BA 47, contralaterally in slice  $-8$ ); putamen/globus pallidus (Pu, ipsilateral, slices  $-8$  to  $+4$ ); SII/inferior parietal lobule (SII, LPI, BA 40, bilaterally, slices  $+24$  to  $+36$ ); middle frontal gyrus (GFm, BAs 6, 8, and 10 ipsi- or contralaterally, slices  $+36$  to  $+48$ ); cingulate gyrus (GC, BA 24, midline/ipsilateral, slices  $+40$  to  $+48$ ); and contralateral SI (slices  $+36$  to  $+52$ ).

anterior cingulate gyrus (also with brushing and capsaicin, levels  $+40$ ,  $+44$ ,  $+48$ ) and activations in ipsilateral anterior insula and ipsilateral putamen (also with brushing and capsaicin, slices  $-8$  to  $+4$ ). The prefrontal cortical activation can be seen in slice  $-8$  bilaterally and continuing contralaterally and superiorly.

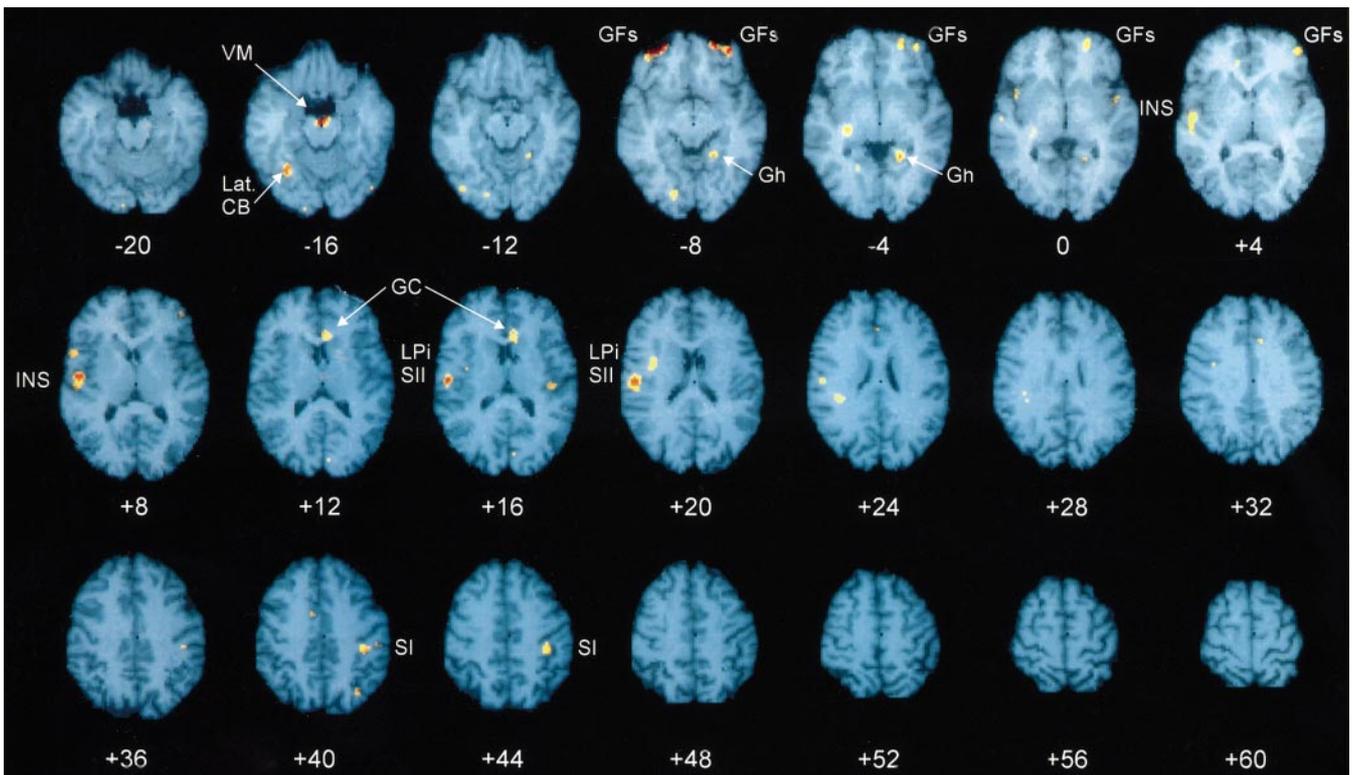
Comparing allodynia with the light brush condition, rather than the resting state, allows the pain components of the allodynia to be more specifically isolated from those related to the brushing that is used to elicit the allodynia (Table 4 and Fig. 4). Several distinct regional activations were obtained: superior frontal gyrus (BA 10, bilaterally and inferiorly, in slice  $-8$  and continuing contralaterally up through slices  $-4$ ,  $0$  and  $+4$ ; this was also seen in the allodynia minus rest comparison); ipsilateral inferior parietal lobule/SII (slices  $+16$ ,  $+20$  and  $+24$ , with the main focus in slice  $+20$ ); and posterior ipsilateral insula (slices  $+4$  and  $+8$ ). Activation was detected in the identical region of lateral cerebellum as obtained in the capsaicin scan (slice  $-16$ ). In slices  $-4$  and  $-8$ , activity was seen in the parahippocampal gyrus, which may represent a response similar to that seen with light brush versus rest, although in that comparison the activation centred on the hippocampus proper (see Fig. 1). Activity in SI was retained, but was more ventrally located in slices  $+40$  and

$+44$  than that seen with either capsaicin or light brush ( $+52$  and  $+56$ ). Three regions of activation appeared to be affected by partial voluming since they exhibited adjusted blood flow values between 39 and 32 ml/100 g/min. These were the anterior activations seen around the corpus callosum in slices  $+12$  and  $+16$ , the anterior activity in slice  $+20$  and posterior activity in slice  $+24$ .

The allodynia condition (versus either rest or brush) is also notable for regions not activated in comparison with acute capsaicin. These included all of the mesencephalic areas (periaqueductal grey, superior colliculus and red nucleus), and the midline cerebellar activations in vermis and the deep nuclei.

### Region-of-interest analysis

The most significant voxel in the regions activated by capsaicin injection were used to designate an 11-voxel volume-of-interest and the activity in this volume was compared across the six scan conditions. Data for all six conditions are shown for two regions, the cerebellar vermis and SI (Fig. 5A and B), and for 18 additional regions for the rest, pain and Wane1 conditions (Fig. 5C). In all volumes-of-interest sampled, the largest adjusted blood flow value was



**Fig. 4** Allodynia compared with light brushing. Conventions as in Fig. 1, legend. Z-scores  $>2.33$  are displayed in yellow; higher scores are shaded from yellow to red-orange; the highest Z-score in this data set was 3.70 ( $P < 0.0001$ ). Subtraction of the light brush component removed many of the activations associated with the raw allodynia condition (i.e. compared with rest). Activation was detected in: ventral midbrain, midline (VM, slice -16); lateral cerebellum (CB lat, ipsilateral, slice -16); superior frontal gyrus (GFs, BA 10, bilaterally in slice 8 and contralaterally up to slice +4); parahippocampal gyrus (Gh, contralaterally, slices -8 to -4); insula (INS, mainly ipsilateral, slices +4 to +8, contralateral activation in slices +16 and +20); cingulate gyrus (GC, midline/contralateral, slices +12 to +16); inferior parietal lobule/SII (LPi, SII, ipsilateral, slices +16 to +24); SI (slices +40 to +44).

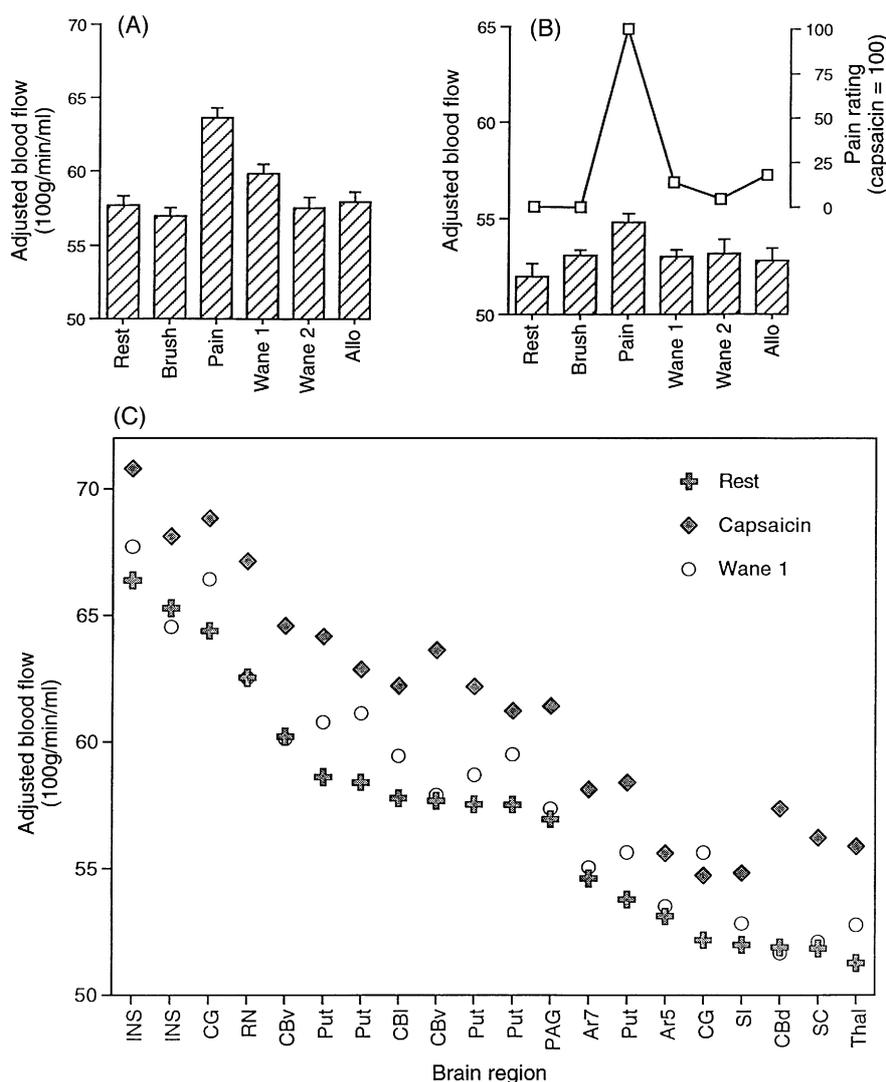
seen with the capsaicin scan, and most of the regions showed a pattern similar to that in the cerebellum (Fig. 5A). The pain rating for the capsaicin, Wane1, Wane2 and allodynia conditions tended to parallel the regional activity (inset, Fig. 5B). The regional activity at the time of the Wane1 scan was either similar to resting state or intermediate between the resting state and capsaicin for nearly every region examined (Fig. 5C). This intermediate Wane1 activity is consistent with the pronounced decrement in spontaneous pain that occurs between the capsaicin injection (rating of 100) and the Wane1 scan (rating of 14.5). The intermediate level of activity was seen at multiple loci within several brain regions (e.g. the multiple sites in putamen or cingulate gyrus illustrated in Fig. 5D). Although not shown, the activity in the Wane2 scan was at, or below, that for Wane1 for 17 of the 20 regions shown in Fig. 5C and in no case did the Wane2 activity exceed the values for capsaicin or fall below those for the resting state.

## Discussion

The set of experiments reported here delineate several new observations and concepts regarding noxious and non-noxious

somatosensory responsiveness at higher levels of the CNS, and reveal several distinct regional responses during experimental allodynia. The data obtained with capsaicin injection demonstrate that robust, temporally acute and spatially localized stimulation of nociceptive C-fibres produced activation in characteristic sets of brain regions that we classify as subserving four main interrelated functional roles. The first is sensory-perceptual, the second is attentional, the third is sensory-motor integration and the fourth is descending control of pain as represented by the mesencephalic periaqueductal grey. This organization provides a conceptual framework. However, the assignments to separate networks are not meant to rigidly compartmentalize the results, since many of the regions are interconnected and multi-sensory.

The data indicate that the non-painful light brush stimulus activates many of the same brain regions represented in the pain-activated networks, but generally less robustly. For example, light brush activation is observed in anterior cingulate gyrus but over a more circumscribed spatial extent (fewer slices show activation) than with capsaicin; in the putamen, activation occurs unilaterally with brush rather than bilaterally. This general tendency for partial overlap also



**Fig. 5** Region-of-interest analysis. Adjusted blood flow (ml/100 g/min) values were normalized to the whole brain mean which was set at 50 ml/100 g tissue/min. **(A)** and **(B)** Each bar represents the average of an 11-voxel volume-of-interest (nine in-plane and two above and below) centred on the most active voxel in the capsaicin scan for each of the scans. **(A)** Cerebellar vermis; **(B)** primary somatosensory cortex. The line graph in panel **B** shows the average proportional pain ratings obtained immediately at the end of the capsaicin, Wane1, Wane2 and allodynia scans. **(C)** Regional activity in the Wane1 scan. Twenty discrete volumes-of-interest are arranged according to their adjusted blood flow during the rest condition. The Wane1 scan was performed 12 min after capsaicin injection. Although not shown, the activity in the Wane2 scan (24 min after injection) was at or below that seen with Wane1 in 17 of the 20 regions-of-interest shown. Abbreviations: INS = insula; CG = cingulate gyrus; RN = red nucleus; CBv = cerebellar vermis; Put = putamen; CBl = cerebellum lateral; PAG = periaqueductal grey; Ar7 = precuneus; Ar5 = lobulus paracentralis; SI = primary somatosensory cortex; CBd = cerebellum deep nuclei; SC = superior colliculus; Thal = thalamus. Most of the regions showed a similar pattern across the six-scan condition as that seen in the cerebellum.

applies to the allodynia condition which contains regional activations common to both the pain and light touch conditions. While some degree of similarity in the activation patterns was expected, two of the most striking differences occurred in SII (seen with light brush but nearly absent with capsaicin) and the cerebellar vermis (seen with capsaicin but not with light brush or allodynia). In the discussion below, pain and light brush are treated together with some reference to the experimental allodynia condition. The latter 'abnormal pain' condition is treated separately at the end.

### Activation produced by capsaicin and/or light brushing

#### Sensory network

**SI.** Capsaicin produced activation in the thalamus and SI, two regions classically associated with somatosensory processing in the CNS. The capsaicin activation in SI mapped to the identical location as that for light brush and the location is consistent with the somatosensory homunculus of Penfield

**Table 1** Regional activation: light brushing versus rest

Brain region	Z-score	x	y	z	Side
Hippocampus	3.49	+26	-34	0	Contralateral
BAs 8/6	3.16	-38	+6	+40	Ipsilateral
(medial frontal gyrus)	2.62	+16	+32	+24	Contralateral
	2.72	-38	-10	+44	Ipsilateral
SI, postcentral gyrus	3.12	+28	-28	+52	Contralateral
Cingulate gyrus	3.10	-10	+2	+44	Ipsilateral
SII/inferior parietal lobule	3.07	+48	-26	+20	Contralateral
	2.89	-44	-30	+28	Ipsilateral
	2.82	-48	-30	+36	Ipsilateral
Insular cortex	2.86	+38	+20	-8	Contralateral
	2.84	-34	+2	-4	Ipsilateral
	2.65	+34	+8	-8	Contralateral
Putamen	2.64	-26	-4	+4	Ipsilateral
BA 7, precuneus	2.58	-14	-58	-48	Ipsilateral

Brushing was applied to the left volar forearm. The table begins with the hippocampus, which had the highest Z-score, all other regions are then listed in descending order according to their Z-score; multiple foci in a region are grouped together. Stereotaxic coordinates and BAs are according to Talairach and Tournoux (1988) ( $n = 11$  and  $P < 0.005$  for all areas listed).  $x$  is mediolateral with +/- for right and left,  $y$  is anterior-posterior and  $z$  is superior-inferior. A region of activation can encompass additional tissue in the slices above or below the indicated slice (see Fig. 1).

**Table 2** Regional activation: capsaicin versus rest

Brain region	Z-score	x	y	z	Side
Cerebellum					
Vermis	4.83	-2	-54	-12	Ipsilateral
Vermis	3.54	0	-64	-20	Midline
Anterior lobe (lateral)	4.45	-26	-52	-12	Ipsilateral
Deep nuclei	3.56	-14	-46	-20	Ipsilateral
Deep nuclei	3.37	+6	-46	-20	Contralateral
Striatum					
Putamen	4.36	-20	-4	+8	Ipsilateral
Putamen	4.29	-26	-10	0	Ipsilateral
Putamen/globus pallidus	4.10	+22	-8	0	Contralateral
Putamen, ventral	3.99	-26	+2	-8	Ipsilateral
Putamen/globus pallidus	3.01	+14	+6	0	Contralateral
Cingulate gyrus					
BA 23/24	3.91	-2	+12	+32	Ipsilateral
BA 24/32	3.48	-10	+2	+44	Ipsilateral
BA 33/24	2.64	+4	+24	+20	Contralateral
Insular cortex	3.76	+38	+6	+8	Contralateral
	2.92	-40	-8	0	Ipsilateral
Parietal cortical areas and SMA					
BA 7, precuneus	3.41	-10	-56	+44	Ipsilateral
BA 5, paracentral lobule	3.11	+10	-36	+48	Contralateral
BA 6, SMA	3.16	-14	-18	+52	Ipsilateral
SI, postcentral gyrus	2.90	+22	-28	+52	Contralateral
SII/inferior parietal lobule	2.64	+46	-34	+32	Contralateral
Mesencephalon					
Superior colliculus	3.20	-6	-36	-12	Ipsilateral
Periaqueductal grey	2.98	-6	-28	-4	Ipsilateral
Red nucleus	2.86	-2	-18	0	Midline*
Thalamus	3.05	+6	-22	+16	Contralateral
	3.04	+2	-14	+16	Contralateral

A 20- $\mu$ l injection containing 250  $\mu$ g capsaicin was made into the left volar forearm. Conventions as in legend to Table 1. A region of activation can encompass additional tissue in the slices above or below the indicated slice (see Fig. 2). \*Coordinates for the red nucleus may, in part, represent thalamic activity since there is a sharp transition from diencephalon to mesencephalon at the AC-PC line (slice 0), and overlap of activity in these areas is possible.

**Table 3** Regional activation: allodynia versus rest

Brain region	Z-score	x	y	z	Side
Cingulate gyrus					
BA 24	4.54	-10	+2	+44	Contralateral
BA 24	3.38	0	+2	+32	Midline
BA 24/33	3.17	0	+24	+20	Midline
BA 24	3.15	+10	-6	+32	Contralateral
BA 29	3.12	+6	-26	+20	Contralateral
BA 24	2.59	+6	+10	+36	Contralateral
Insular cortex	4.32	-40	+4	0	Ipsilateral
	3.52	+38	+20	-8	Contralateral*
	2.85	-36	+8	+16	Ipsilateral
	2.76	+36	+2	0	Contralateral
Striatum					
Putamen (posterior tip)	3.70	-26	-20	-4	Ipsilateral
Globus pallidus	3.45	-20	-18	+4	Ipsilateral
Putamen	2.90	-24	-4	+4	Ipsilateral
Putamen (inferior tip)	2.82	-26	0	-8	Ipsilateral <sup>†</sup>
SII/inferior parietal lobule					
BA 40	3.68	-42	-30	+28	Ipsilateral
BA 44	3.22	-48	+10	+8	Ipsilateral
BA 40	2.95	+34	-50	+36	Contralateral
Frontal cortex					
BA 6	3.54	-38	-2	+40	Ipsilateral
BA 10	3.49	+36	+44	+4	Contralateral
BA 10	3.32	-34	+42	-8	Ipsilateral
BA 6	3.19	+20	-14	+52	Contralateral
BA 10	2.83	+20	+48	-8	Contralateral
BA 10	2.77	-36	+38	0	Ipsilateral
BA 47 (inferior frontal gyrus)	2.76	+38	+38	-8	Contralateral
BA 8 (superior frontal gyrus)	2.74	-8	+26	+48	Ipsilateral
SI, postcentral gyrus	3.47	+38	-26	+36	Contralateral
	3.23	+30	-32	+44	Contralateral

Allodynia was elicited by light brushing of the left volar forearm adjacent to the injection site. Conventions as in legend to Table 1. \*Overlaps with inferior frontal gyrus. <sup>†</sup>Location also is consistent with superior tip of the amygdala.

**Table 4** Regional activation: allodynia versus light brushing

Brain region	Z-score	x	y	z	Side
Prefrontal cortex*					
BA 10	3.70	-30	+48	-8	Ipsilateral
BA 10	2.93	+28	+44	-8	Contralateral
BA 10	2.90	+16	+48	-8	Contralateral
BA 10	2.71	+18	+46	0	Contralateral
BA 10	2.60	+36	+42	+4	Contralateral
SII/inferior parietal lobule	3.37	-48	-18	+20	Ipsilateral
Cerebellum (lateral)	3.33	-28	-54	-16	Ipsilateral <sup>†</sup>
Ventral midbrain	3.14	+2	-14	+16	Midline
Insular cortex	3.08	-46	-10	+8	Ipsilateral
	2.69	-32	-2	+20	Ipsilateral
	2.68	+34	-22	+16	Ipsilateral
Putamen (posterior)	2.89	-26	-22	-4	Ipsilateral <sup>†</sup>
Parahippocampal gyrus	2.87	+16	-46	-4	Contralateral
SI, postcentral gyrus	2.71	+30	-28	+44	Contralateral

The activations due to the light brush component inherent in allodynia have been subtracted out in this comparison. Conventions as in legend to Table 1. \*The atlas (Talairach and Tournoux, 1988) tends to be somewhat inconsistent in all three dimensions in this region and the activation assigned to BA 10 of middle frontal gyrus may also overlap BAs 11 and 9. <sup>†</sup>Very similar loci were found in the capsaicin versus rest comparison.

and Rasmussen (1950) and more recent PET studies of somatotopic mapping of SI with vibration (Fox *et al.*, 1987) and delivery of noxious thermal stimuli to the volar forearm by a contact thermode (Casey *et al.*, 1994; Coghill *et al.*, 1994). While the local maxima of the light brush and capsaicin responses were nearly identical, the pain activation encompassed two axial slices, whereas light brush was detected in only one slice. This suggests that the intensity or magnitude of the response in SI was greater for the nociceptive stimulus than for the non-nociceptive stimulus, despite the fact that the area of skin stimulated by the brush (~20 cm<sup>2</sup>) was greater than the punctate injection site directly exposed to capsaicin (a 20 µl injection). Our data also suggest that different degrees of perceived pain produce a graded response in SI as assessed by the volume-of-interest analysis which showed an intermediate level of activity in the Wanel scan for this region. Moreover, the latter statement also is applicable to other regions which, during the waning phase, exhibited an intermediate level of activity (see Fig. 5D).

Based on the Z-score and percentage increase over resting state, the response of SI ( $Z = 2.90$ , 5.4% increase) to capsaicin was weak relative to regions like the cerebellar vermis ( $Z = 4.83$ , 10.2% increase) and ipsilateral putamen ( $Z = 4.36$ , 8.5% increase). The lower statistical significance probably reflects a lower degree of activation, but other factors may also contribute such as a greater spatial variability of SI compared with putamen and vermis. Activation of SI has not been detected in every PET study of pain. This has caused considerable discussion regarding the role of SI in pain perception (see Duncan *et al.*, 1992; Jones *et al.*, 1992; Apkarian, 1995). Nevertheless, SI activation has been observed in several, more recent PET studies of thermal pain, and unequivocally with imaging modalities such as functional magnetic resonance imaging (Iadarola *et al.*, 1994) such that the direct participation of SI in pain processing is unquestionable. However, precisely what aspects of the pain signal are responded to or analysed by human SI needs further clarification (reviewed in Kenshalo and Willis, 1991).

**SII.** Pain and light brush produced differential degrees of activation in SII. Capsaicin pain caused a barely detectable activation of contralateral SII, whereas light brush provoked a marked, extensive and bilateral activation of SII and the adjacent inferior parietal lobule (BA 40). The very weak activation of SII by capsaicin-induced pain was unexpected since at least three previous PET studies of thermal pain have shown increased regional blood flow in SII. The difference suggests that some distinct feature of the stimulus in the previous thermal pain studies triggers a response in SII that is not solely a function of the painful component. This distinct feature is absent from the capsaicin stimulus, which is basically punctate in terms of space and time. Three of the thermal pain PET studies used a contact thermal probe that was moved repeatedly from one point on the skin to adjacent points during the period of the scan (Talbot *et al.*, 1991; Casey *et al.*, 1994; Coghill *et al.*, 1994). In each of

these studies, the control condition was a non-painful warm stimulus delivered in the same manner. Using this as the baseline for comparison, the activity associated with the repetitive tactile components should have been removed; nevertheless, activation above that in the non-painful control condition occurred in SII. In contrast, a painful stimulus that does not have the repetitive tactile component (e.g. capsaicin injection) does not produce an activation of SII (or yields a very weak signal). Recent results from our laboratory replicate the lack of SII activation with capsaicin and demonstrate that this is not an artefact (Coghill *et al.*, 1995a). These data indicate a differential role for SI and SII in pain processes which needs to be reassessed functionally and anatomically. There is obviously a specific characteristic of the experimental pain stimuli that this region processes and the consequences of certain cortical lesions suggest that loss of SII can produce deficits in pain processing (Greenspan and Winfield, 1992).

**Thalamus.** The third region in the sensory-perceptual network is the thalamus. In the present study, the capsaicin-induced activation within the thalamus occurred at the midline, mainly contralaterally, with a dorsomedial location that extended posteriorly (Fig. 2). The nuclei contained within this region include the anterior nuclear group, the dorsomedial nucleus, the lateral dorsal nucleus and dorsomedial portions of the pulvinar. A distinct locus associated with the contralateral ventroposterolateral nucleus was not detected. Our observation of a more widespread and more medially placed thalamic activation, rather than a distinct focus in the ventroposterolateral nucleus, is consistent with several previous PET studies of pain (Casey *et al.*, 1994; Coghill *et al.*, 1994). The light brush stimulus also failed to produce activity in the medial thalamus or a distinct locus in the ventroposterolateral nucleus, perhaps due to the small size of the area stimulated and the fact that each subject had only one brush and one capsaicin scan. Sufficient repetition of a tactile/proprioceptive finger-to-thumb tapping task (e.g. four resting state and four active state scans in eight subjects) produces a significant, discrete focus of activation in the contralateral ventroposterolateral nucleus (K. F. Berman and J. Van Horn, personal communication). The participation of the ventroposterolateral nucleus may be inferred by the fact that, following capsaicin, light brush or allodynia, activation in the corresponding SI was detected in each case.

The more medially lying thalamic nuclei that are activated by capsaicin have been demonstrated to make connections with several cortical regions that also show activation in either the capsaicin condition or in allodynia. These include the anterior cingulate (discussed below), BAs 5 and 7 in the parietal lobe, and prefrontal cortex (BA 10).

**Insula.** The insula is placed in the sensory-network category because of the demonstrated connections with SI and BAs 5 and 7 in parietal lobe (reviewed in Mesulam and Mufson, 1985) but since insular activation is also commonly observed in PET studies of limb and digit movement, it may also be

relevant to motor integratory processes (Sadato *et al.*, 1995). While direct connections with cortical motor nuclei are comparatively sparse (e.g. little or no projections from primary motor cortex), the insula has direct projections to the spinal cord and pons. The insula has been divided functionally and anatomically into anterior and posterior portions; the posterior insula receives inputs from cortical and thalamic somatosensory regions and the anterior receives inputs from medial thalamic nuclei and cortical limbic, olfactory and paralimbic regions (e.g. cingulate gyrus and amygdala). Capsaicin yielded bilateral activation over a widespread area of insular cortex with a mid-anterior focus contralaterally and a more posterior focus ipsilaterally. Several previous studies of pain or non-noxious somatosensory stimulation have detected insular activation (Burton *et al.*, 1993; Casey *et al.*, 1994; Coghill *et al.*, 1994; Derbyshire *et al.*, 1994). Most of the activations appear to be anteriorly located if pain is the stimulus. Light brush also produced bilateral, mid-anterior insular activation. This appears to suggest an association with limbic circuitry. However, without more definitive mapping using several different types of tasks or stimuli, it seems premature to conclude that these activations reflect a clear segregation into anterior (limbic/motor) and posterior (sensory) divisions.

### Attentional network

**Anterior cingulate.** Several other activated regions were common to both light brush and capsaicin pain conditions. Many of these are multimodal and were categorized into networks subserving attentional or sensory-motor integration functions. The anterior cingulate gyrus activations are placed in the attentional network and both pain and light brush produced significant activations here. However, as seen in SI, pain produced a more extensive activation of anterior cingulate cortex (approximately six slices, mainly BA 24 but overlapping BAs 23, 32, and 33) than did the light brushing (BA 24, mainly in one slice) (Figs 1 and 2). It is interesting to note that, among its many inputs (reviewed in Vogt, 1985; Devinsky *et al.*, 1995), BA 24 receives afferents from thalamic nuclei that are activated by capsaicin; e.g. reciprocal connections have been demonstrated in the macaque between BA 24 and the dorsomedial nucleus of the thalamus and additional afferents are received from the anteromedial, intralaminar and other thalamic nuclei (Musil and Olsen, 1988; Yeterian and Pandya, 1988). Thus, based on the medial location of much of the thalamic activation, this region may also constitute a portion of the attentional network.

Results from numerous PET studies have amply demonstrated that anterior cingulate cortex activation occurs under many conditions and may be subdivided according to efferent connections (Paus *et al.*, 1993). Stimuli as diverse as performing a delayed response alternation task (Gold *et al.*, 1996), the Stroop Test (Pardo *et al.*, 1990), or receiving a painful cutaneous input, produce robust activation of anterior

cingulate gyrus, with BA 24 being the common denominator in most studies. This cortical region is large, receives multiple sensory inputs, and has multiple connections with the motor system (e.g. motor cortex, red nucleus and spinal cord) to implement or inhibit action (Dum and Strick, 1991; Morecraft and Van Hoesen, 1993; Devinsky *et al.*, 1995). In this regard it is important to note that the conditions of the capsaicin scan imposed a strong motoric 'conflict' in that the subjects were instructed not to move (and indeed did not overtly move) and, at the same time, the stimulus imposed a drive to orient to the site of injection and withdraw the limb. Experimental results from higher-order motor control tasks have suggested the participation of the anterior cingulate in the execution of appropriate, and suppression of inappropriate, motor responses (Pardo *et al.*, 1990; Paus *et al.*, 1993).

The light brush stimulus also activated anterior cingulate cortex, although less robustly than the capsaicin pain stimulus, suggesting that activation of this region is common to several modalities of somatosensory stimulation. The context in which the stimulus is delivered may have a strong influence on the degree of activation seen in brain networks not directly involved in the perceptual aspects of sensory processing. In the PET scanner, the light brush stimulus was non-threatening, the subject was forewarned and therefore there was no element of surprise or alarm involved. However, if the 'light brush' stimulus had been delivered by an insect crawling on the skin, activation within the attentional and motor integratory networks might have been much more robust, especially if the visual component of the stimulus was included. Activation of these networks by acute pain may be less dependent on context, since pain provides its own context, and sub-routines such as orientation are executed automatically.

The above circuit was characterized as part of an attentional network (see Posner and Petersen, 1990). However, this same set of regions has also been characterized as constituting the 'medial pain system' with connections being made via the medial and intralaminar thalamic nuclei and cingulate cortex (Price, 1988). The medial pain system has been suggested to be the network subserving the affective component of pain, whereas the lateral spinothalamocortical system is traditionally assigned a role in stimulus localization and discrimination. The affective portion of the stimulus, if present, was not specifically addressed as part of the experimental design in our study. It is interesting that there was no activation detected in brain regions typically associated with affect, such as the caudate nucleus or the amygdala (which receives afferent projections from the anterior cingulate, plus strong input from the insula) (Mesulam and Mufson, 1985; Vogt, 1985; Bhatia and Marsden, 1994) while the anterior cingulate itself was robustly activated. In addition, the light touch stimulus, which was not aversive, also produced activation of the cingulate. It is possible that the brief duration of the scan (the first minute after capsaicin injection) may not have been sufficient to elicit a strong affective component. Current evidence suggests that the anterior cingulate, with its many interconnections, is involved in a number of higher order

cognitive processes, including response selection, affect, aversive conditioning and premotor processing, and that it performs several interrelated functions during painful stimulation. While it would be interesting to target the affective component directly, experimentally, the present data suggest that an attentional–somatomotor integratory role is one of the main functions of cingulate gyrus in the present experimental context (see also discussions in Hsieh *et al.*, 1995; Vogt *et al.*, 1996; Fink *et al.*, 1997).

### Motor network

**Cerebellum.** It is commonly believed that the cerebellum does not play a role in our conscious perception of pain and this region was assigned to the sensory–motor integratory network. The activations in cerebellar areas, especially the vermis, are among the most robust in the entire brain. The vermis had the highest Z-score, was significant in our earliest analysis ( $n = 7$ ) and the coordinates were spatially stable in subsequent analyses. This robust activation is probably due to a high density of direct spinocerebellar inputs to the vermis as shown by retrograde tracing studies with HRP (horse radish peroxidase) in cat and monkey (Snyder *et al.*, 1978). Whether the projections to the vermis represent collaterals of spinothalamic projection neurons or a separate set of neurons has not been established. In contrast to capsaicin pain, activation of cerebellar vermis is not detectable with either light touch or pain associated with the experimental allodynia. While the areas of activation are large and tend to overlap, two additional sites of activation were detected in the cerebellum. A second maximum was seen in the lateral cerebellum which we ascribe to the arm area of anterior lobe (Ekerot *et al.*, 1991a, b) and a third maximum was consistent with a location in the deep nuclei. The lateral site was also seen in the comparison of allodynia to light brush (Fig. 3 and Table 3).

**Putamen/globus pallidus.** Capsaicin produced a prominent, bilateral activation of these striatal nuclei. The putamen, as compared with the caudate, is more involved in the integration and elaboration of movement, rather than affect, as demonstrated by the clinical signs of humans with differential lesions of the putamen or caudate, respectively (Bhatia and Marsden, 1994). We observed bilateral activation with capsaicin and unilateral activation with light brush. Thus, both sensory stimuli elicit an activation which we attribute to initiation of orientation and withdrawal, and voluntary inhibition of these two responses, as mentioned above for the attentional network. Our bilateral activation with a painful stimulus is consistent with the trend towards bilateral activation observed by Jones *et al.* (1991) who used the word ‘priming’ to describe the motoric role of the putamen/globus pallidus activity during noxious stimulation. Consistent with all studies of thermally induced pain was a lack of activation in either the head or tail of the caudate nucleus.

**Premotor cortex and frontal oculomotor fields.** The activation detected in contralateral BA 6 with the capsaicin stimulus is consistent with the engagement of a motoric circuit (but without a movement being executed) and had been seen in previous PET studies of thermal pain (Coghill *et al.*, 1994; Hsieh *et al.*, 1995). Highly significant activity in the light brush condition was detected in the ipsilateral middle frontal gyrus which overlapped both ipsilateral BA 8 (frontal oculomotor fields) and BA 6 (supplementary motor cortex). No similar activation occurred in the BA 8 region with capsaicin. In the comparison of allodynia with the resting state, the BA 8 activation is clearly present. However, this activation was eliminated in the comparison of allodynia with light brush, suggesting a unique association with the light brush stimulus. We tentatively attribute the brush activation of BA 8 to a strong cortical involvement in visual orientation and identification of a moving, light tactile stimulus. Additional overlap with cortical regions activated during motor tasks may also include BA 5. An interplay between sensory systems and motor systems, both primary and premotor, is apparent from the recent study of Fink *et al.* (1997).

**Red nucleus and superior colliculus.** These two regions have well known roles in sensory–motor integration. The superior colliculi are activated bilaterally. The deep layers of the superior colliculus respond to noxious peripheral stimulation (Larson *et al.*, 1987) and receive afferent projections from the anterolateral quadrant of the spinal cord. This region co-ordinates head/neck movement with visual, auditory and somatosensory input. The ipsilateral red nucleus is activated at its superior pole. The red nucleus receives input from the motor cortex and cerebellum and the latter was one of the most strongly activated regions in the CNS following capsaicin. Activity in the red nucleus and superior colliculus may reflect priming of the motor system in response to the painful stimulus. Neither of these areas were activated during the waning phase scans or by light brush, suggesting a dependence on stimulus intensity and/or the painful nature of the stimulus.

### Descending control network

**Periaqueductal grey.** The periaqueductal grey is the third mesencephalic region activated. This region is known to be involved in descending control of pain and to receive input from ascending second order spinal cord nociceptive neurons (Trevino, 1976; see also Willis and Coggeshall, 1978). Electrical stimulation of this region produces analgesia in animals. Activation of the periaqueductal grey has also been reported in two PET studies of thermal pain (Casey *et al.*, 1994; Derbyshire *et al.*, 1994) and by Rosen *et al.* (1994) during experimentally induced angina pectoris. As with the attentional network, we have placed only one region in this ‘descending control network’. However, many regions

interconnect with the periaqueductal grey, including the anterior cingulate (for reviews see Vogt, 1985; Devinsky *et al.*, 1995). No activation of the periaqueductal grey occurred with the light brush stimulus or with the allodynia; the latter may be due to a lower level of pain in comparison with capsaicin.

### ***The waning phase scans***

The waning phase scans were not part of our formal set of statistical comparisons. The region-of-interest analysis suggests that the intermediate levels of spontaneous pain reported by the subjects during these scans is, in part, reflected by an intermediate level of activation in many foci. Nearly every area assessed with the region-of-interest analysis showed either no change or an increased level of relative blood flow in comparison with the resting and brush states. Much of the motor-integratory network, such as the cerebellar vermis or sites in the mesencephalon (including the periaqueductal grey), did not show a sustained increase in blood flow. The putamen was an exception and an intermediate level of residual activity was detected at several loci. Intermediate activity was also seen in the thalamus. One focus in anterior cingulate gyrus exhibited activation in the Wane1 scan equal to that after capsaicin (see Fig. 5C). The sustained activity in some areas versus the shorter duration of activity in others is consistent with the idea of differential sensitivity to aspects of the stimulus such as pain intensity and duration.

### ***Activation produced by allodynia***

For allodynia, the initiating stimulus delivered to the skin is a light brush, and the perception is a painful burning type of sensation. This abnormal sensation is experimentally produced by the prior administration of a strong painful stimulus (in the present experiment, capsaicin injection). This experimental constraint precluded full counterbalancing of the sensory conditions. Order effects in the context of functional brain imaging of pain have not been explored, but it is conceivable that this may have affected our results. Within the context of this constraint, the regional activation pattern seen with allodynia indicates that it is a partial composite of both pain and light brush, and defines additional regions that may participate in CNS processing of abnormal pain sensations. In general, the comparison of allodynia with the resting state yielded a pattern of regional activation similar to light brush, although the extent of activation in the different regions was larger. Since the subtraction of the brush component attenuated or removed many of the regional activations associated with the allodynia, the discussion will mainly focus on this analysis which allows the activations accompanying allodynia *per se* to be discerned.

### ***Comparison of allodynia with capsaicin pain and light brushing***

When comparing allodynia with resting state, activations within the sensory network included primary and secondary

somatosensory cortices but not the medial thalamic regions seen with capsaicin pain; a pattern most similar to that of light brush. However, the inability to detect medial thalamic activation may reflect the lesser degree of pain during the allodynia compared with capsaicin, rather than a selective activation of the light touch pathways. Regardless of the lack of detectable thalamic activation, allodynia did produce a robust activation of SI and SII/inferior parietal lobule and much of the activation in these cortical regions was removed when the light brush component was subtracted from the allodynia. This removal is consistent with the idea that the light touch input is the predominant factor driving SI and SII cortical activation in capsaicin-induced allodynia. Some residual activation in both SI and SII/inferior parietal lobule was seen, although it was more inferiorly located than the loci for normal light brush and capsaicin pain. Thus, some 'non-brush' component appears to be processed in S1 and SII, but it may not necessarily be a pain component. Other levels of the nervous system outside the field of view of the PET scanner may also show an enhanced response to capsaicin-induced changes. For example, recordings from neurons in the dorsal column nuclei, have shown that injection of capsaicin rapidly expands the receptive field size to tactile stimuli (Pettit and Schwark, 1996).

In the motor network, another region that distinguished capsaicin pain from allodynia was the cerebellar vermis: allodynia did not produce a detectable activation of the vermis. While the vermis displayed no detectable activation with allodynia, the lateral cerebellar focus was activated (Fig 4). Extrapolating from electrophysiological observations in the cat, this lateral area is sensitive to a wide range of somatosensory stimuli from light brush to noxious pinch via spino-olivocerebellar inputs (Ekerot *et al.*, 1991*b*). Thus, despite the lesser pain intensity of the experimental allodynia compared with capsaicin, the cerebellum is not completely insensitive to the pain-related aspects of the allodynia stimulus. This selective activation of cerebellar sub-regions may be potentially useful in studies of chronic pain patients exhibiting a prominent allodynia component. The other main region in the motor network was the putamen. Activation of the putamen occurred unilaterally in the allodynia-resting state comparison and was nearly completely removed by subtracting out the brush component. Similarly, the sensitivity to subtraction using the brush condition for comparison also applied to the anterior cingulate gyrus; most of the cingulate activation could be attributed to the brush component; however, the residual activity was more inferiorly located.

Two regional activations were particularly prominent in the allodynia condition. One was a bilateral activation of the inferior frontal lobes which extended further superiorly on the contralateral side. The Talarach and Tournoux (1988) coordinates of the maximum focus of this activation lay within BA 10. However, this large activation also extended to BAs 9 and 11. BAs 9, 10 and 11 all receive input from medial dorsal thalamus and BA 24 of the anterior cingulate gyrus, both of which were activated by pain or brush. In

contrast to the allodynia condition, activation of BA 10 was not seen with capsaicin or in several PET studies using thermal stimuli (Jones *et al.* 1991; Talbot *et al.*, 1991; Casey *et al.*, 1994; Coghill *et al.*, 1994). However, this region has been observed to be more active in patients with chronic neuropathic pain during their spontaneous pain than after pain relief with nerve block, as well as in several other studies of pain induced by thermal or chemical stimuli (Derbyshire *et al.*, 1994; Hsieh *et al.*, 1996), or dobutamine-induced angina (Rosen *et al.*, 1994). The lack of frontal cortical activation with capsaicin in the present study is surprising, given the extensive prefrontal responses with an acute pain stimulus seen in the above studies. This suggests that prefrontal responses to pain may be highly context dependent. One factor contributing to the lack of frontal cortical activation in the present study may be that the capsaicin stimulus has minimal somatosensory complexity. It is unlikely that the fact that the subjects had received a screening injection (thereby familiarizing them with the stimulus) played a role in the differential prefrontal response because, at the time of the screening injection, subjects were familiarized with the subsequent hyperalgesia and allodynia as well.

The second region that was activated in allodynia but not with capsaicin was assigned to the parahippocampal gyrus. Although an exact role for this region in pain processes is not clear, the parahippocampal gyrus projects to, or receives input from, several regions active in both the brush and capsaicin conditions. These include BA 24 of the anterior cingulate gyrus and dorsomedial pulvinar. The activation of prefrontal cortex and parahippocampal gyrus suggests that the allodynia condition contains a cognitive and limbic component, either not present or inconsistently present, during acute pain or light tactile stimulation.

Whether the allodynia model provides a reasonably accurate picture of the regional activation pattern seen in allodynia associated with clinical pathological pain states is presently under investigation (Coghill *et al.*, 1995b). Allodynia in the capsaicin model is established within a matter of minutes whereas that in chronic neuropathic pain conditions usually develops over a period of days to weeks and lasts for years. While some of the underlying mechanisms may be very similar, when one considers the entire CNS, the pathophysiology of chronic pain probably involves more complex, progressive changes as the pain condition persists and adaptive processes in neural circuits are engaged to maintain homeostasis. That such modulatory changes of physiology do occur has been amply demonstrated for adult human motor cortex (Karni *et al.*, 1995; Sadato *et al.*, 1995) and somatosensory cortex in both normal subjects and patients with pathological pain (phantom limb pain) (Elbert *et al.*, 1995; Flor *et al.*, 1995). Plasticity in nociceptive circuits has also been demonstrated in dorsal spinal cord using experimental persistent pain models (Hunt *et al.*, 1987; Iadarola *et al.*, 1988; Hylden *et al.*, 1989; Herdegen *et al.*, 1991; reviewed in Coderre *et al.*, 1993) and with capsaicin

(Pettit and Schwark, 1996). Thus, fully developed pathological chronic pain and allodynia, as opposed to the acute experimental capsaicin model, may present with a characteristic set of abnormal physiological conditions in affected patients (Di Piero *et al.*, 1991; Hsieh *et al.*, 1995; Iadarola *et al.*, 1995) that may be distinct from the present model. However, at the very least, the capsaicin allodynia provides a highly informative opportunity to understand the early phases of these conditions.

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