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High opiate receptor binding potential in the human lateral pain system

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To determine how opiate receptor distribution is co-localized with the distribution of nociceptive areas in the human brain, eleven male healthy volunteers underwent one PET scan with the subtype-nonselective opioidergic radioligand [^{18}F]fluoroethyl-diprenorphine under resting conditions. The binding potential (BP), a parameter for the regional cerebral opioid receptor availability, was computed using the occipital cortex as reference region. The following regions of interest (ROIs) were defined on individual MR images: thalamus, sensory motor strip (SI/MI area), frontal operculum, parietal operculum, anterior insular cortex, posterior insular cortex, anterior cingulate cortex (ACC; peri- and subgenual part of "classical ACC" only), midcingulate cortex (MCC, posterior part of "classical ACC"), putamen, caudate nucleus and the amygdala.

BP for [^{18}F]fluoroethyl-diprenorphine was lowest in the sensory motor strip (0.30). Highest BP was found in thalamus (1.36), basal ganglia (putamen 1.22, caudate 1.16) and amygdala (1.21). In the cingulate cortex, ACC (1.11) had higher BP than MCC (0.86). In the operculo-insular region, we found high BPs in all ROIs: anterior insula (1.16), posterior insula (1.05), frontal operculum (0.99) and parietal operculum (0.77). Factor analysis of interindividual variability of opiate receptor BP revealed four factors (95% explained variance): (1) operculo-insular areas, ACC, MCC and putamen, (2) amygdala and thalamus, (3) caudate and thalamus, (4) SI/MI and MCC.

Nociceptive areas of the lateral pain system (frontoparietal operculum and insula) have opiate receptor BPs significantly higher than SI/MI, comparable to anterior and midcingulate areas of the medial pain system. These findings suggest that the cortical anti-nociceptive effects of opiates are not only mediated by ACC and MCC, but also by the operculo-insular cortex, if it can be assumed that opioid binding mediates anti-nociception in those structures.

Keywords: Opioid; PET; Human; Imaging; Insula; Operculum

Introduction

Endogenous opiates play a role in a multitude of bodily functions, including learning, memory, reward, eating, drinking, sexual activity, pregnancy, mood, locomotion, cardiovascular, gastrointestinal, renal and hepatic function, respiration, thermoregulation and immunological responses (Vaccarino and Kastin, 2001). The most common clinical use of opiates is, however, for their analgesic effects, which are mediated by inhibitory, mostly μ-opiate receptor (OR)-mediated effects in the peripheral nerve, the spinal cord, brainstem, thalamus and cortex. The highest opiate receptor density is in lamina II of the dorsal horn, i.e. in a nociceptive nucleus of the spinal cord. Opiate receptors are also present in other parts of the nociceptive system, including the periaqueductal gray, thalamus, anterior cingulate cortex and insula

Abbreviations: PET, positron emission tomography; MRI, magnetic resonance imaging; OR, opiate receptor; BP, binding potential, a measure of OR availability; ACC, anterior cingulate cortex: peri-and subgenual part of "classical ACC" only; MCC, mid-cingulate cortex: posterior part of "classical ACC"; OIC, operculo-insular cortex; SI/MI, primary sensory motor area; SII, secondary somatosensory cortex; ROI, region of interest; [^{18}F]FEDPN, 6-O-(2-[^{18}F]Fluoroethyl)-6-O-desmethyldiprenorphine; TDD, 3-O-trityl-6-O-desmethyl-diprenorphine; HPLC, High-performance liquid chromatography.

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(Yaksh et al., 1988; Jones et al., 1991; Casey et al., 2000; Bencherif et al., 2002); these opiate receptors can be mapped by positron emission tomography (PET). Early PET studies (Jones et al., 1991; Vogt et al., 1995b) reported that the primary somatosensory cortex (SI), a part of the lateral pain system, has one of the lowest opiate receptors densities and that opiate receptors are primarily associated with the medial pain system (medial thalamus, anterior cingulate cortex) that is thought to mediate the affective–motivational component in pain perception (Melzack and Casey, 1968; Rainville et al., 1997; Treede et al., 1999; Price, 2000).

Concepts of the role of the anterior cingulate cortex and other cortical structures in pain perception have evolved over the past decade. What was formerly summarily termed anterior cingulate cortex has been functionally divided into a more posterior motor part called midcingulate cortex (MCC) and the ACC proper in the perigenual region (Vogt et al., 1995a), with higher opiate receptor density than MCC both in rats and in humans (Vogt et al., 1995b, 2001). As part of the lateral system, the region around the Sylvian fissure contains several nociceptive regions, including SII in the parietal operculum, medial parts of the frontal operculum and anterior as well as dorsal aspects of the insula (Treede et al., 2000; Craig, 2002; Frot and Mauguière, 2003). While the insula was traditionally considered to be part of the medial pain system due to its output into the limbic system, it receives direct nociceptive input from lateral thalamic nuclei (Apkarian and Shi, 1994) and may thus also be seen as part of the lateral pain system. The operculo-insular cortex (OIC) has one of the shortest latencies of activation by painful stimuli (Frot and Mauguière, 2003; Schlereth et al., 2003).

Little is known about opiate receptor distribution in the OIC. The insula has a high density (Atweh and Kuhar, 1977; Pfeiffer et al., 1982; Jones et al., 1991), but the frontoparietal operculum has not been studied as a separate area in its own right. We now set out to use PET imaging with the subtype-unselective opioid receptor ligand [^{18}F]Fluoroethyl-diprenorphine ([^{18}F]FEDPN, Wester et al., 2000) coregistered with structural MRI to perform a region of interest analysis of opiate receptor availability in nociceptive cortex regions. We compared these regions with regions known for high (thalamus, basal ganglia) or low (SI/MI, occipital cortex) opiate receptor density.

Methods

This study was carried out in accordance with the Helsinki Declaration and was approved by the local ethics committee, the German Federal Health Administration (BfArM) and the German radiation protection authorities (BfS). Informed written consent was obtained from each subject.

Subjects

Eleven healthy male volunteers (age 23–50, mean: 34) were included in this study and were paid for participation. The subjects had neither current nor previous history of relevant physical illness, no current or past psychiatric disorders, no family history of major psychiatric disorder in first-degree relatives and they were not regularly taking medication. All subjects received a physical and mental state examination, blood analysis and cerebral magnetic resonance imaging.

Radiochemistry

The fully automated production of [^{18}F]FEDPN was performed using a modified TracerLab FX_{F-N} synthesis module from GE Medical Systems following a modified procedure based on the method described by Wester et al. (2000) applying the secondary labeling precursor 2-[^{18}F]fluoroethyltosylate to 3-*O*-trityl-6-*O*-desmethyl-diprenorphine (TDD). The 2-[^{18}F]fluoroethyltosylate was synthesized as described elsewhere (Piel et al., 2003) and obtained in a diethyl ether solution which was evaporated in a stream of nitrogen. To the dried 2-[^{18}F]fluoroethyltosylate, a solution of 2 mg TDD and 5 mg sodium hydride in 300 μL of DMF was added, and the resulting mixture was stirred for 8 min at 100°C. The reaction mixture was cooled to room temperature, 300 μL HCl (2 N) was added slowly and stirred for 5 min at 40°C. After the mixture was cooled to room temperature, it was diluted with 4 ml aqueous ammonia (25%), stirred for 2 min and purified using semi-preparative HPLC (Luna 10 μ C18(2), 250 × 21.2 mm inner diameter, acetonitrile/0.1 N ammonium formate 55:45, 15 ml/min, t_r: 23.2 min). The HPLC fraction containing the product was diluted with 40 ml 0.1 N ammonium formate, loaded on a Strata-X cartridge, washed with 10 ml water, eluted with 2 ml ethanol and diluted with 18 ml physiological saline solution to yield 1.3–2.0 GBq (radiochemical yield 19 ± 4%) of [^{18}F]FEDPN. HPLC analysis (Luna 5 μ, C18(2), 250 × 4.6 mm inner diameter, methanol/0.1 N ammonium formate 70:30, 1 ml/min, t_r: 12.2 min) showed that the radiochemical purity was >99%, while the specific activity (determined via UV–calibration curve) was >1700 GBq/mmol.

PET imaging

Images were acquired on a Siemens ECAT EXACT whole-body PET scanner. The camera has a field-of-view of 16.2 cm in 47 planes with a plane spacing of 3.375 mm, an axial resolution of 6.0 mm FWHM in 3D mode (Wienhard et al., 1992) and an in-plane resolution of 6.0 mm (resolution in center with scanner in 3D mode). Data acquisition comprised a series of 30 time frames, of scan duration increasing progressively from 20 s to 10 min, resulting in a total scanning time of 124 min. A 15-min transmission scan using a ^{68}Ge source was carried out prior to each study for subsequent attenuation correction. A mean of 150 MBq (±50 MBq) [^{18}F]FEDPN was injected intravenously as a bolus into a cubital vein over approximately 30 s. The specific activity at time of injection was >0.5 GBq/μmol (mean ± SD).

Images were reconstructed with filtered back projection using a Ramp filter and a Hanning filter. A frame-by-frame motion correction was applied by matching cortical isodensity contour points. A mean occipital time–activity curve was generated from three regions of interest drawn on three subsequent transaxial slices.

We decided to use non-invasive quantification methods in order to avoid a nociceptive stimulus during arterial blood sampling, which might change the opioid receptor status. Binding potentials (BP) of volumes of interest were calculated using the non-invasive Logan Plot (Logan et al., 1996) with reference region input according to the following equation:

$$\frac{\int_0^t c_t\,\mathrm{d}t}{c_t} \cong \frac{V_d}{V'_d}\frac{\int_0^t c_r\,dt}{c_t} + c$$

where c_t is the tissue radioligand activity in the receptor-containing region of interest, c_r is the tissue radioligand activity of [^{18}F]FEDPN in the reference tissue (occipital cortex), V_d is the volume of distribution of the receptor-rich region and V'_d is the volume of distribution of the reference region. The ratio V_d/V'_d (VDR) was estimated with a non-linear least square minimization procedure, and BP was calculated from VDR. The occipital cortex was chosen as a reference region since it is generally considered to have very low opiate receptor density (Sadzot et al., 1991).

MR image acquisition

For determination of regions of interest and as basis for the overlay with BP images, a high resolution anatomical MR dataset was acquired from each subject. The MR image acquisition was performed in a 1.5-T scanner (Siemens Magnetom Vision) using a T1-weighted MPRAGE (Magnetization Prepared Rapid Acquisition Gradient Echo) sequence in sagittal direction (180 slices of 1 mm thickness each, 256 × 256 matrix, repetition time 9.7 ms, echo time 4 ms).

ROI analysis

The following regions of interest (ROIs), relevant for the nociceptive system, were defined on individual MR images: thalamus, frontal operculum, parietal operculum, anterior insular cortex, posterior insular cortex, anterior cingulate cortex (ACC), midcingulate cortex (MCC) and the amygdala. In addition to these, we chose reference regions known for high (caudate nucleus, putamen) and low opiate receptor BPs (SI/MI) as positive and negative controls.

The ROI determination was performed by drawing the outline of the structure on the MR image slice through the center of each anatomical region chosen (see Fig. 1) and then calculating the 2D area. Fig. 1A illustrates the location of the insula, at the level of the anterior commissure (equivalent to Talairach y coordinate = 0; this is also the approximate y coordinate of the lower edge of the central sulcus). The insular ROIs were subdivided into an anterior and a posterior region, measured on average 10.1 ± 2.1 mm anterior and 6.2 ± 2.1 mm posterior of the anterior commissure. The opercular ROIs were located at the inner face of the operculum (Fig. 1B), directly across the insula, down to the "lip" facing the temporal lobe, and were as well divided into an anterior and posterior part. This region has usually not been defined as an ROI in neuroimaging studies, but there is electrophysiological evidence that it may contain a primary nociceptive cortex area (Frot and Mauguière, 2003; Vogel et al., 2003). According to recent concepts (Vogt et al., 1995a), we have subdivided the anterior cingulate gyrus into midcingulate cortex (MCC) and perigenual/subgenual anterior cingulate cortex (ACC, Fig. 1C). As a representative region for SI/MI, we placed an ROI into the hand area of the primary sensory–motor cortex, recognized as an omega-shaped "knob" (Yousry et al., 1997). All regions were assessed bilaterally. Subsequently, the ROIs were superimposed onto the realigned BP images, and the mean BP within that volume was calculated.

Data analysis

Data are presented as mean ± standard deviation, when biological variability is to be illustrated (ROI sizes and locations), and as mean ± standard error of the mean, when significant differences between mean values are being illustrated (as in Fig. 3). Differences in BP in the various ROIs in both hemispheres were assessed by 2-way ANOVA followed by post hoc tests (Student's t tests) for comparison of BPs between the different regions. To assess inter-correlations between areas as a function of interindividual variability (cf. Zubieta et al., 2001), we performed a factor analysis applying a principle component analysis of the correlation matrix followed by varimax rotation. The varimax rotation comprises orthogonal (independent) factors. We retained factors with eigenvalues ≥ 1 from the scree plot.

Results

High [^{18}F]FEDPN binding was seen in the thalamus, the basal ganglia (caudate, putamen) and further lateral, the insula (Fig. 2). The regions with the lowest opiate receptor availability were the occipital cortex (transverse slice), which was used as reference region, and the sensory–motor strip around the central sulcus (sagittal slice). The sagittal slice demonstrates that, within the insula, opiate receptors are mostly located in its anterior and dorsal parts. The dorsal insula is immediately adjacent to the frontal and parietal operculum, separated by the circular sulcus of the insula. This raises the question to what extent the ORs may extend from the insula into the operculum (see coronal slices).

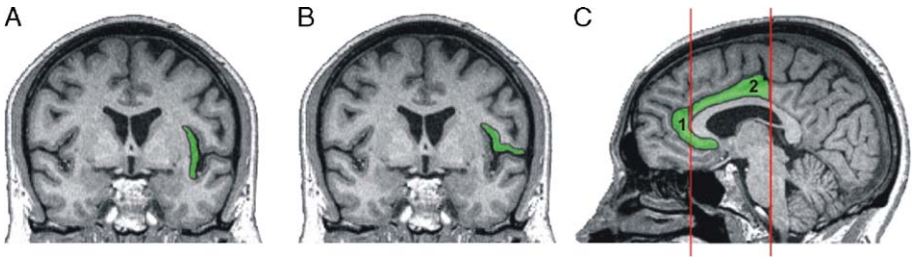

Fig. 1. Example for placement of regions of interest (ROIs) in individual MRI scans (marked with green lines). (A) ROIs for the insula were separated into an anterior and a posterior part at the vertical plane through the anterior commissure ($y = 0$). The location of the frontal slices (coronal) was on average 10.1 ± 2.1 mm anterior, the location of the posterior slices 6.2 ± 2.1 mm behind the anterior commissure (in coronal plane). Insular and opercular ROIs were measured on the same coronal slices (anterior and posterior) as demonstrated in examples A and B. (B) ROIs in the opercular cortex were placed in its inner vertical part, facing the insula, down to the "lip" facing the temporal lobe. Frontal (anterior) and parietal (posterior) operculum are separated by the central sulcus, which at this level is also located approximately at $y = 0$. (C) Two ROIs were chosen within the cingulate cortex. The anterior cingulate (ACC, green area 1) included the rostral perigenual part up to the tangent at the anterior part of the corpus callosum (left vertical line), the midcingulate (MCC, green area 2) included the adjacent part ending at the coronal plane through the posterior commissure (right vertical line).

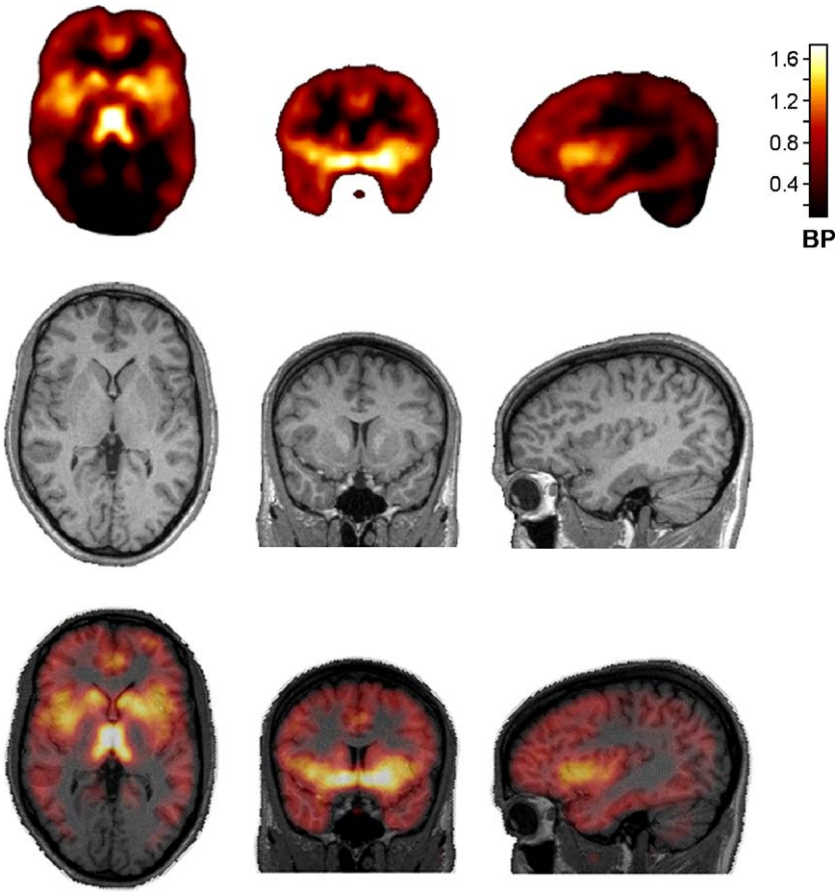

Fig. 2. The upper row shows the [^{18}F]FEDPN binding potential image of one subject in transverse, coronal and sagittal slices. The middle row shows the corresponding individual MRI slices and the lower row the BP maps superimposed onto the MRI (PET/MRI overlay). BP was high in the thalamus, the basal ganglia, the anterior cingulate and the opercular and insular cortex. High opioid binding in the operculum is unlikely to be caused by smoothing artifacts from the insula as the contours of the thalamus (with the highest BP) are quite sharp.

Mean BPs in the predefined regions of interest are shown in Fig. 3. Opiate receptor availability was low in the sensory motor strip, however, significantly different from zero ($P < 0.001$). Highest availability was found in thalamus, basal ganglia (putamen and caudate nucleus) and in the amygdala. In the cingulate cortex, ACC had higher BP than MCC ($P < 0.001$). In the operculo-insular region, [^{18}F]FEDPN BPs were of similar magnitude as in the cingulate cortex. Statistical analysis revealed a significant difference between ROIs ($F_{11,110}$: 51.1, $P < 0.001$), no differences between left and right hemispheres ($F_{1,10}$: 0.23, $P = 0.64$) and no interaction. Thus, for subsequent analyses, both sides (hemispheres) were pooled.

Post hoc comparisons (t tests) showed that SI/MI had lower BP than all other ROIs ($P < 0.001$). Since one focus of our study was opiate receptor binding in the operculo-insular region, we investigated more closely BPs in this area: the insula (anterior and posterior part combined) had a significantly higher BP than the operculum (frontal and parietal part combined) in post hoc t test ($P < 0.001$). The anterior part (anterior insula and frontal operculum combined) had a higher opiate receptor binding than the posterior part (posterior insula and parietal operculum combined) of the OIC ($P < 0.001$). This was also true for testing just within the insula (anterior vs. posterior, $P < 0.001$) or within the operculum (frontal vs. parietal, $P < 0.001$). Both anterior and posterior insula had a significantly higher BP than the posterior opercular cortex (SII; $P < 0.001$). The BP in ACC was significantly higher than in MCC ($P < 0.001$).

Table 1 shows the average size and location of the different ROIs. Except for a slightly larger ROI in the right than the left thalamus, there was no significant difference in size between ROIs in both hemispheres. To give an impression where the ROIs were located in the brains, the relative positions of the slice where the BP was measured is given as distance (in mm) from the individual anterior commissure (AC). Measured on transversal slices, the ROIs for thalamus and SI/MI were located above and ROIs for basal ganglia were located below AC. On coronal slices, ROIs for the anterior OIC were located anterior to and ROIs for posterior OIC were located posterior to AC, on average 16 mm apart; ROIs for the amygdala were near AC level. On sagittal slices, ROIs for ACC/MCC were 4 mm lateral of the midline (medial fissure).

A factor analysis of interindividual differences in regional patterns of [^{18}F]FEDPN BP yielded four factors (one major and three minor) explaining 88% of total variance. Since the factor structure was almost identical in left and right hemispheres, we repeated the analysis with both hemispheres combined (four factors explained 95% of the variance, Table 2). Factor 1, explaining 45% of the variance, had high loadings on insula and operculum, cingulate (ACC/MCC) and putamen. Factor 2 (18%) loaded primarily on amygdala and thalamus, factor 3 (17%) loaded on

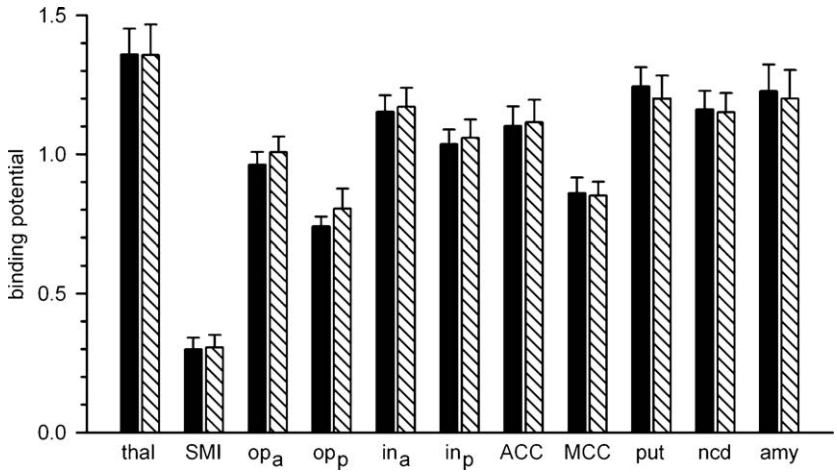

Fig. 3. Mean [^{18}F]FEDPN binding potentials for all regions of interest in both hemispheres. Open bars: left hemisphere, hatched bars: right hemisphere. Thal: thalamus, SMI: primary sensory–motor cortex ('hand knob'), OPa: anterior (frontal) operculum, OPp: posterior (parietal) operculum, INa: anterior insula, INp: posterior insula, ACC: anterior cingulate cortex, MCC: midcingulate cortex, put: putamen, ncd: nucleus caudatus, amy: amygdala. Highest BPs were found in the thalamus, the lowest were found in the somatosensory (hand) area. Insula, operculum and cingulate cortex had similarly high BPs. Means ± standard errors from 11 subjects.

caudate nucleus and thalamus and, finally, factor 4 (15%) on SI/MI and MCC.

Discussion

To date, the lateral pain system, including the primary and secondary somatosensory cortex (SI, SII), has been considered to be an area of low opioid receptor density, whereas structures of the medial system, like medial thalamus and cingulate cortex, are known to have a much higher opioid receptor density. In this PET study in a sample of healthy subjects, we calculated the opioid receptor BP in 11 bilateral regions of interest (ROIs), each of which was assigned on the individual MR scan of each subject. No normalization procedures were used (except spatial filtering of the BP maps), enabling us to assign a priori the binding regions to clear anatomical structures. The main findings of the present investigation were (1) high opiate receptor availability in frontal operculum and SII, (2) higher binding in ACC than in MCC and (3) a factor analysis of interindividual variability revealing a grouping of regions into meaningful entities.

Regions associated with the lateral and medial pain systems

The primary somatosensory cortex (SI) is usually considered to be the prototypical cortical area of the lateral pain system. It exhibits a fairly low opioid receptor density, as has been shown in previous PET studies (Vogt et al., 1995b; Jones et al., 1991). Although sparsely distributed, our data show that opioid receptors are not completely absent: BP in the hand area of SI/MI was significantly different from zero. This is unlikely to be due to spillover from adjacent regions, as shown by the sharp boundaries of the thalamus. Moreover, specific opiate receptor binding in SI has been shown in rat, dog and horse (Ding et al., 1996; Hellyer et al., 2003).

In contrast, the cingulate cortex, which is recognized as a major part of the medial pain system, has a high opioid receptor density (Jones et al., 1991; Vogt et al., 1995b; Tölle et al., 1999). In the rat brain, perigenual cingulate areas 24 and 32 have the highest opiate

Table 1
Size and location of ROIs

ROI	ROI size (cm^2 ± SD)		Slice orientation	Slice location relative to AC (mm ± SD)	
	Left	Right		Left	Right
Thalamus	3.15 ± 0.82	3.61 ± 0.86	Tra (*z*)	6.2 ± 2.7	6.2 ± 2.7
SI/MI	1.96 ± 0.51	1.88 ± 0.62	Tra (*z*)	49.5 ± 2.8	49.4 ± 2.8
Operc ant	1.77 ± 0.22	1.65 ± 0.43	Cor (*y*)	10.1 ± 2.1	10.1 ± 2.1
Operc post	1.75 ± 0.24	1.80 ± 0.32	Cor (*y*)	−6.2 ± 2.1	−6.2 ± 2.1
Insula ant	1.96 ± 0.35	1.97 ± 0.29	Cor (*y*)	10.1 ± 2.1	10.1 ± 2.1
Insula post	1.70 ± 0.29	2.00 ± 0.26	Cor (*y*)	−6.2 ± 2.1	−6.2 ± 2.1
ACC	3.59 ± 1.81	3.76 ± 1.03	Sag (*x*)	−3.7 ± 0.8	3.8 ± 1.7
MCC	8.35 ± 1.91	9.12 ± 1.26	Sag (*x*)	−3.7 ± 0.8	3.8 ± 1.7
Putamen	3.09 ± 0.47	3.01 ± 0.40	Tra (*z*)	−0.5 ± 3.1	−0.5 ± 3.1
Ncd	1.45 ± 0.26	1.43 ± 0.25	Tra (*z*)	−0.4 ± 3.2	−0.4 ± 3.2
Amygdala	0.73 ± 0.13	0.75 ± 0.11	Cor (*y*)	−0.5 ± 2.5	−0.5 ± 2.5

ROI sizes in the left and right hemispheres and ROI locations relative to the anterior commissure (AC). Thus, slice locations can roughly be compared to Talairach space. Sagittal slices (Sag) = *x*: (+) right, (−) left hemisphere; coronal slices (Cor) = *y*: anterior (+) or posterior (−); transverse slices (Tra) = *z*: (+) above or (−) below AC. SI/MI: primary sensory–motor cortex (hand area); operc ant/post: anterior/posterior opercular cortex; ACC: anterior cingulate; MCC: midcingulate cortex; ncd: caudate nucleus

Table 2
Factor analysis of binding potentials

ROI	Factor 1	Factor 2	Factor 3	Factor 4
Thalamus	0.31	0.57[a]	0.61[a]	0.39
SI/MI	0.23	0.36	0.29	0.85[b]
$Operc_{ant}$	0.83[b]	0.40	0.28	0.09
$Operc_{post}$	0.84[b]	0.10	0.40	0.26
$Insula_{ant}$	0.80[b]	0.38	0.12	0.39
$Insula_{post}$	0.80[b]	0.31	0.32	0.37
ACC	0.92[b]	0.26	0.24	0.14
MCC	0.62[a]	0.44	0.22	0.52[a]
Putamen	0.80[b]	0.02	0.52	0.17
Ncd	0.44	0.07	0.83[b]	0.22
Amygdala	0.25	0.91[b]	0.05	0.29
Explained variance	45%	18%	17%	15%

SI/MI: primary sensory–motor cortex (hand area); operc: opercular cortex; ant/post: anterior/posterior; ACC: anterior cingulate; MCC: midcingulate cortex; ncd: caudate nucleus.
[a] Evenly distributed across 2 factors.
[b] Assigned to a single factor.

receptor binding and receive the highest input of axons bearing (presynaptic) opiate receptors as compared to midcingulate or posterior cingulate areas (Vogt et al., 2001). Our data confirm the presence of a gradient in BP, with highest availability in anterior (peri- and subgenual part) as compared to midcingulate cortex with a marked difference of approximately 25%, even though the average size of the ROIs in the midcingulate was even larger than in the anterior part.

Strangely, the opercular cortex, a part of the lateral pain system comprising the frontal operculum and the parietal part (SII), showed opioid receptor BP of similar magnitude as in cingulate cortex. The anterior part (frontal operculum) exhibited even higher BP than the midcingulate cortex, making it a very important candidate for (cortical) opiate modulation. When tested with laser evoked potentials, this region exhibits one of the shortest latencies to activation by noxious heat stimuli (Frot and Mauguière, 2003; Vogel et al., 2003), consistent with its direct thalamo-cortical nociceptive input (Apkarian and Shi, 1994) that largely bypasses SI. It has been suggested that the frontoparietal operculum, including SII, may be more important for nociception and pain sensation than SI (Treede et al., 2000). The opiate receptors in this region have not been studied explicitly, but data reported in two recent studies are consistent with our novel finding. Patients with central poststroke pain exhibited reduced opiate receptor BP in several brain regions (Jones et al., 2004; Willoch et al., 2004), including the parietal operculum (labeled as SII or inferior parietal cortex) and frontal operculum (labeled as insula, but located lateral of the circular sulcus according to its Talairach coordinates 40, −9, −4; Talairach and Tournoux, 1998).

Although very close to one another and grouped within the same factor in our factor analysis, the insular cortex had even higher opiate receptor BP than the opercular cortex. These findings had been reported before as evidence for opiate receptor modulation within the medial pain system (Jones et al., 1991). If the controversial concept that the dorsal insula receives nociceptive input from the thalamic nucleus VMpo is correct, this cortex region would actually belong to the lateral system (Craig, 1995).

In intracranial recordings of the insula, Frot et al. (2005) only recently found increasing amplitudes of laser evoked potentials with increasing stimulus energies in the painful range, whereas recordings of SII showed intensity coding below pain threshold with a plateau at higher intensities, suggesting an important role of the dorsal insula in pain intensity coding. These functional findings are well paralleled by our BP quantification, with a significantly higher BP in the insula than in SII (posterior opercular cortex). Furthermore, direct stimulation of the insula in humans has been reported to be painful (Ostrowsky et al., 2002), demonstrating the functional significance for opiate receptors in this region for pain modulation. The insula as a whole, however, integrates several aspects of interoception (Craig, 2002), and OR receptors in the insular cortex are probably involved in autonomic and other functions as well (Vaccarino and Kastin, 2001).

Opiate effects on sensory and affective pain components

The high opiate receptor binding in perigenual ACC and the low binding in sensorimotor cortex have led to the suggestion that opiate effects at the cortical level may be limited to the medial pain system that is involved with processing of information with affective content (Jones et al., 1991; Vogt et al., 1995b). Although we could confirm the high opiate receptor BP in the cingulate cortex and the difference between ACC and MCC, the high opiate receptor binding in all parts of the operculo-insular cortex suggests that nociceptive areas that process sensory information can also have high opiate receptor binding. Recent imaging studies have suggested a more prominent role of the operculo-insular cortex in the lateral pain system than for SI (Bushnell et al., 1999; Peyron et al., 2000; Hofbauer et al., 2001, Apkarian et al., 2005). In fact, there is behavioral evidence that opioids can influence both affective and sensory pain components in a dose-dependent manner (Price et al., 1985). Whereas opiate effects on the sensory pain component have previously been assigned to spinal sites only (lamina II), our findings suggest that sensory pain components may be influenced at cortical level as well.

Factor analysis

The factor analysis revealed four groups of ROIs as possibly distinct entities. ROIs had been chosen to include nociceptive cortex areas and a few positive and negative control regions, whereas other parts of the brain with moderate to high opiate receptor BP such as the frontal cortex or the cerebellum were not analyzed. Thus, OR functions not related to nociception may not be fully represented in the observed factor structure. Taking that into account, the factor to be consistently co-activated significantly loaded on cingulate cortex, insula, opercular cortex and putamen, suggesting that these structures constitute a functional unit in pain perception. Cingulate cortex, opercular and insular cortex have been shown to be activated in both electrophysiological and imaging studies, whenever a painful stimulus was applied (Casey, 1999; Peyron et al., 2000; Apkarian et al., 2005). The putamen, which, at first glance, does not seem to belong into this group, has also been shown to be activated in human as well as animal pain studies (Chudler and Dong, 1995) with a somatotopical representation for painful stimuli (Bingel et al., 2004).

Notably, the caudate nucleus did not load on the same factor as the putamen, although both nuclei belong to the basal ganglia.

These findings suggest that nociceptive functions in the basal ganglia may be restricted to the putamen. Thus, the four regions could represent a functional unit within the pain system. The second factor, the amygdala, can be seen as processing area for the emotional pain component. The third factor comprised of the caudate nucleus and the thalamus, a combination that may be related to cognitive functions (Mori, 2002). SI/MI together with a weaker MCC loading formed the last factor, explaining 15% of variance and could be interpreted as part of the attentional–orientational network initiating the reaction after a painful stimulus (MCC: motor area).

The medial pain system, especially the cingulate cortex, has been target for neurosurgery in the past to alleviate pain in patients with intractable pain syndromes (Hurt and Ballantine, 1974). If the pain system comprises of relevant pathways or subsystems apart from the medial pathway, as suggested by our factor analysis of BPs, this could be an important reason why cingulotomy has sometimes little or only short-lasting effects.

Technical considerations

In group analyses, brain areas adjacent to each other, such as insula and opercular cortex, run the risk of being merged in the course of spatial normalization. Moreover, automatically assigned coordinates sometimes do not reflect correct Brodmann areas or even correct lobes. This is particularly true for the operculo-insular region, where coordinates labeled as insula may be located in the operculum and vice versa. By reporting locations with respect to the individual sulcus separating insula and operculum without spatial normalization, we could basically rule out this source of error.

BPs were calculated by means of the non-invasive Logan model with the occipital cortex as a reference region. The use of non-invasive models seems an appropriate approach for the quantification of ^{11}C- or ^{18}F-labeled diprenorphine (Willoch et al., 2004; Spilker et al., 2004), although the occipital cortex might not fulfil the criteria of an ideal reference region since it may contain a small amount of opiate receptors (Asselin et al., 2003). Spilker et al. (2004) also used this area as the reference region and found consistent BP values between invasive and non-invasive quantification method, with the non-invasive BPs showing less interindividual variability.

Conclusions

The present study shows that the operculo-insular cortex, a part of the lateral pain system, has high opiate receptor density of similar magnitude as the anterior and midcingulate cortex of the medial pain system. These data suggest that in addition to the affective–motivational pain component also the sensory-discriminative pain component may be influenced by opiate receptor agonists at a cortical level. Furthermore, our factor analysis data suggest that the operculo-insular cortex forms a functional unit together with the anterior cingulate cortex and the putamen.

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