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An Investigation to Dissociate the Analgesic and Anesthetic Properties of Ketamine Using Functional Magnetic Resonance Imaging

Richard Rogers, M.B.B.S., F.R.C.A., Richard G. Wise, Ph.D.,† Deborah J. Painter, M.B.Ch.B., F.R.C.A.,‡ Sarah E. Longe, M.Sc.,§ Irene Tracey, D.Phil.‖*

Background: **Anatomic sites within the brain, which activate in response to noxious stimuli, can be identified with the use of functional magnetic resonance imaging. The aim of this study was to determine whether the analgesic effects of ketamine could be imaged.**

Methods: **Ketamine was administered to eight healthy volunteers with use of a target-controlled infusion to three predicted plasma concentrations: 0 (saline), 50 (subanalgesic), and 200 ng/ml (analgesic, subanesthetic). Volunteers received noxious thermal stimuli and auditory stimuli and performed a motor task within a 3-T human brain imaging magnet. Activation of brain regions in response to noxious and auditory stimuli and during the motor task was compared with behavioral measures.**

Results: **The analgesic subanesthetic dose of ketamine significantly reduced the pain scores, and this matched a decrease in activity within brain regions that activate in response to noxious stimuli, in particular, the insular cortex and thalamus. A different pattern of activation was observed in response to an auditory task. In comparison, smaller behavioral and imaging changes were found for the motor paradigm. The lower dose of ketamine gave similar but smaller nonsignificant effects.**

Conclusion: **The analgesic effect can be measured within a more global effect of ketamine as shown by auditory and motor tasks, and the analgesia produced by ketamine occurs with a smaller degree of cortical processing in pain-related regions.**

KETAMINE, a dissociative anesthetic, is as a noncompetitive antagonist to the phencyclidine site of the *N*-methyl-D-aspartate (NMDA) receptor for the excitatory neurotransmitter glutamate. The clinical analgesic effect of ketamine is almost entirely NMDA mediated, although ketamine has been shown to bind to other receptors, such as non-NMDA glutamate, nicotonic and muscarinic cholinergic, monoaminergic, and opioid receptors, as well as interacting with sodium and calcium channels.[1] The word *dissociative* has two contexts, first in the clinical sense of a dissociation of the patient from his or her environment, which is unlike other anesthetic states. Second, there is an electrophysiologic dissociation between the thalamus and the limbic system during ketamine anesthesia.[2] Ketamine is an unusual intravenous induction agent because it is analgesic at subanesthetic doses. In these subanesthetic doses, it is also known to cause cognitive impairment, depersonalization, schizophrenic-like psychoses, and memory impairment.[3–5]

The central nervous system has a crucial role in the perception of pain. Pain is a complex experience that is more than the transduction of noxious stimuli, having sensory/discriminative and emotional/affective dimensions.[6] *Pain* is rarely measured objectively but is defined as a subjective experience,[7] and *analgesia* is defined as the reduction of this subjective phenomenon, pain. Brain imaging has the potential to remove this subjectivity and objectively measure nociceptive processing in the brain. Positron emission tomography and functional magnetic resonance imaging (fMRI) have been used to image a "pain matrix," a network of sites within the brain that are active in response to noxious stimulation.[8,9] These studies have shown that the most consistently activated sites are thalamus, insula/SII cortex, anterior cingulate cortex, and primary sensory cortex. The measured brain response to noxious stimulation can be affected by many factors, including the nature of the noxious stimulus and various psychologic parameters, such as attention, as well as pharmacologic agents, and this has been well reviewed by Peyron *et al.*[10] Crucially, fMRI produces objective measures of brain activity, which, if correlated with subjective experience, have the potential to identify the neural correlates of analgesia.[11]

In a previous study, we investigated the analgesic properties of remifentanil, a μ-opioid agonist, with the use of fMRI.[12] To explore the general applicability of fMRI to quantify pharmacologic analgesia, we chose ketamine as an analgesic drug that produced analgesia by a mechanism dissimilar to that of opioid analgesia.

* Consultant Anaesthetist, Nuffield Department of Anesthetics, University of Oxford, John Radcliffe Hospital. † Postdoctoral Research Fellow, Centre for Functional Magnetic Resonance Imaging of the Brain, Department of Clinical Neurology, University of Oxford, John Radcliffe Hospital; and Department Human Anatomy and Genetics, University of Oxford. ‡ Anaesthetic Research Fellow, Nuffield Department of Anesthetics, University of Oxford, John Radcliffe Hospital; and Department of Human Anatomy and Genetics, University of Oxford. § Research Assistant, Centre for Functional Magnetic Resonance Imaging of the Brain, Department of Clinical Neurology, University of Oxford, John Radcliffe Hospital. ‖ University Lecturer, Head Pain Group, Centre for Functional Magnetic Resonance Imaging of the Brain, Department of Clinical Neurology, University of Oxford, John Radcliffe Hospital; and Department Human Anatomy and Genetics, University of Oxford.

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Address correspondence to Dr. Rogers: Nuffield Department of Anesthetics, John Radcliffe Hospital, Oxford, Oxfordshire, OX3 9DU, United Kingdom. Address electronic mail to: rogers@fmrib.ox.ac.uk.

The aim of this study was to objectively measure the analgesic effect of ketamine with use of fMRI. We hypothesized that a reduction in the brain processing of the response to noxious stimuli would match the reduction in behavioral pain scores. In addition, we were interested in how the activity of specific brain regions involved in processing noxious stimuli would be reduced and how this compared with the brain activity arising from non–pain-related tasks.

Materials and Methods

Subjects

Ethical permission for this study was obtained from the local ethics committee (Central Oxfordshire Research Ethics Committee, Oxford, Oxfordshire, United Kingdom). Volunteers gave written informed consent.

Eight male, right-handed volunteers with a mean age of 28 yr (range, 19–37 yr) who had no history of psychiatric illness were recruited. They were excluded if they were taking medication or drugs acting on the central nervous system for therapeutic or recreational use. A medical history and, where appropriate, a physical examination were performed to ensure the patients were fit and well (American Society of Anesthesiologists physical status I) and that there was no contraindication to the administration of ketamine or to magnetic resonance imaging.

On their first visit, outside the scanner, volunteers were given a ketamine infusion that was slowly increased to the maximum dose (predicted plasma concentration of 200 ng/ml) to test for tolerance and analgesia and to familiarize the volunteers with the method of noxious heat delivery and pain scoring. The second visit was a scanning session during which three predicted plasma concentrations of ketamine (0, 50, and 200 ng/ml) were administered to the subjects. Imaging response was measured to pain and auditory stimuli. Four of the original subjects returned for a third visit to determine the effects of ketamine at these doses on the motor system. During this session, imaging and behavioral data were collected relating to a motor task.

Drug Administration

Volunteers fasted before all visits (6 h for solids, 2 h for clear fluids) and were supervised for approximately 2 h after the ketamine infusion was stopped. Ketamine was administered *via* an indwelling intravenous cannula (23-gauge Wallace Y-Can; SIMS Portex Ltd., London, United Kingdom) inserted into the right antecubital vein. Racemic ketamine hydrochloride as a 1-mg/ml solution was administered by means of a computer-controlled continuous infusion[13] (target-controlled infusion, Graseby 3500 pump; SIMS Graseby Ltd., Watford, Hertfordshire, United Kingdom, programmed and supplied by Anesthesia Technology Ltd., Leeds, United Kingdom). The infusion was governed by a three-compartment model, using the pharmacokinetic parameters calculated by Domino *et al.*,[14] to maintain a stable plasma concentration of ketamine. Predicted plasma concentrations are reported.

Ketamine produces analgesia at plasma concentrations of 100–200 ng/ml.[15–17] Standard intravenous anesthetic induction doses (2–2.5 mg/kg) produce a peak plasma concentration of 9,000–25,000 ng/ml, with awakening at approximately 1,000 ng/ml.[14] Psychological effects are dose-dependent and are rare at subanesthetic doses[18,19]; doses up to a plasma concentration of 200 ng/ml have been well tolerated in volunteers.[20] For this study, a maximum predicted plasma concentration of 200 ng/ml ketamine was chosen to provide analgesia and avoid anesthesia. A potential confounding factor in this study was nonanalgesic drug effects on the blood oxygen level–dependent (BOLD) signal. For these reasons, multiple doses were investigated to attempt to differentiate the effects of ketamine on different types of cortical processing. Three different plasma concentrations of ketamine were studied. A control saline infusion was the 0 ketamine concentration. The second was a subanalgesic dose (predicted plasma concentration of 50 ng/ml). A third analgesic subanesthetic dose (200 ng/ml) was chosen. A predicted plasma concentration of 50 ng/ml is approximately equivalent to a steady state infusion (after 20 min) of 0.18 $mg \cdot kg^{-1} \cdot h^{-1}$, and 200 ng/ml is approximately equivalent to a steady state infusion of 0.71 $mg \cdot kg^{-1} \cdot h^{-1}$.

Physiologic Data

Heart rate, noninvasive blood pressure, and oxygen saturation were measured continuously with use of a 9500 Multigas Monitor (MR Equipment Corporation, United Kingdom supplier Wardray Premise Ltd., Thames Ditton, Surrey, United Kingdom).

Experimental Stimulus Paradigm

The design of the first experiment is shown in figure 1. Ketamine was administered in a fixed stepwise increase in dose in a single scan session. The target-controlled infusion was allowed to run for 10 min after each change in concentration to allow the drug to reach steady state before the stimulus paradigm and imaging were started. Noxious and auditory stimuli were presented to the subjects in a pseudo-random sequence for 14 min while echoplanar imaging was performed. This was repeated at each of the three concentrations of ketamine. The sequence of stimuli was randomized, with a mean stimulus interval of 41 s (range, 27–66 s). The noxious stimuli were separated by a minimum delay of 59 s. Alternate noxious and alternate auditory stimuli were offset in time by a whole number of interscan (repetition time) periods plus half repetition time (1.5 s). This improves the effective temporal sampling of the BOLD

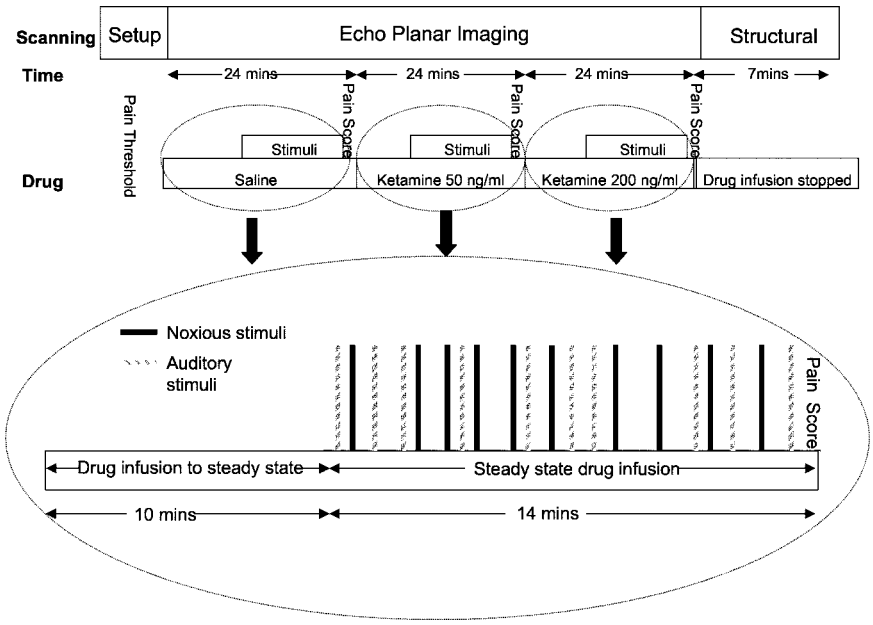

Fig. 1. (*Top*) Experimental design. After initial pain thresholding and scan setup, echo planar imaging data were collected, and three identical blocks of stimuli were delivered to the volunteer under different drug conditions. (*Bottom*) Detail of one of these blocks. After 10 min to allow drug stabilization, 10 auditory and 10 noxious stimuli were delivered in a pseudo-random sequence. After the last stimulus, the pain scores were collected. After the third block of stimuli, the drug infusion was stopped, and a high-resolution structural scan was collected. For the motor experiment, the motor task replaced the periods of presentation of auditory and noxious stimuli.

response to the stimulus and thus reduces the associated systematic errors of invariant slice acquisition timing with respect to the stimulus onset.

At the end of each block of pain and auditory stimuli for a given ketamine concentration, the volunteers were asked to rate the average pain for those 10 noxious stimuli on two subjective pain scales. Pain (heat) intensity and averseness were scored with use of two 11-point (0–10) combined numeric and verbal descriptor scales. In the heat/intensity scale, 2 represents warm, no pain; 5 represents mild pain; and 10 represents unbearable pain. In the averseness scale, 2 represents discomforting; 5 represents distressing; and 10 represents excruciating. These scales were projected onto a screen by a digital projector (In Focus LP1000; In Focus Inc., Wilsonville, OR). After each ketamine infusion, the subjects were asked if they had noted any change in the noise of the scanner.

Thermal Noxious Stimuli. The thermal noxious stimuli of 3 seconds' duration were administered to the dorsum of the volunteer's left hand by a thermal resistor. This device was built to work within the scanner and consisted of a 1.5 × 2-cm copper sheet, which delivered a fast ramping stimulus (30°–60°C in 0.8 s) and simultaneously measured the skin temperature under the device. The position of the device was kept constant during each session, delivering the stimulus to the same area of skin. This device was controlled with use of in-house software. The software controlled the maximum temperature and duration of the thermal stimulus and received triggers from the scanner to enable synchronization with data collection.

Before scanning and while volunteers lay in the scanner, the volunteers were "thresholded" for pain (mean temperature, 56.2°C; range, 55–57.5°C). The temperature of the thermal noxious stimulus was adjusted in an iterative fashion until the volunteers consistently scored the pain as 8 out of 10 (strong pain) on the thermal pain intensity scale in response to three noxious stimuli of the same temperature delivered 1 min apart.

Auditory Stimuli. A multifrequency, amplitude-modulated tone of 3 seconds' duration was presented to the volunteers. The auditory stimuli were presented binaurally with magnetic resonance–compatible electrostatic headphones (MRC Institute of Hearing Research, Nottingham, United Kingdom).

Motor Task. On a separate occasion, four of the original volunteers returned to perform a motor paradigm within the scanner. In these additional imaging sessions, the motor paradigm took the equivalent place of the noxious/auditory paradigm in figure 1. The motor task was repeated at each ketamine concentration. Volunteers practiced the task immediately before scanning to reduce the practice effect on reaction times during scanning. The volunteers were presented with a symbolic representation of a right hand displayed on the projector screen. In the active condition, this indicated which digit should be used to press one of five buttons on a button box. In the control condition, this indicated a digit, but no button was to be pressed. The trials were presented in blocks of 30. Four blocks of requests requiring button presses were alternated with four blocks of nonbutton requests; each block lasted 45 s. Within each block, a new digit trial was presented every 1.5 s. A total of 240 digit images were displayed, of which 120 required a button press and 120 did not. An equal number of requests were made for each digit, and the order of presentation of digits was randomized.

The button-press data were used to calculate reaction times and to detect errors. An error was considered to

have occurred if a button was pressed when the request was not to press, or when no button was pressed when a button press was requested or the wrong digit was pressed. Each button press was counted as correct or incorrect (maximum of 1 error per button press). Erroneous button presses were eliminated from calculating mean reaction times. Mean reaction times were calculated from the delay between the start of the request and a correct button press.

Functional Magnetic Resonance Imaging Protocol

Volunteers were scanned in a 3-T human magnetic resonance imaging system, with a 1-m bore magnet (Oxford Magnet Technology Ltd., Oxford, United Kingdom) with use of a birdcage radio frequency coil and a reduced bore head gradient coil (Magnex SGRAD MKIII; Magnex Scientific Ltd., Oxford, United Kingdom). The magnet was driven by a Varian Unity Inova console (Varian Medical Systems, Palo Alto, CA) using Siemens' gradients (Siemens Medical Ltd., Bracknell, Berkshire, United Kingdom).

A functional magnetic resonance whole brain gradient-echo echoplanar imaging sequence was used with the following parameters: repetition time, 3,000 ms; echo time, 30 ms; flip angle, 90°; field of view, 256 × 256 mm; matrix, 64 × 64. Twenty-one contiguous slices were taken through the whole brain, with a slice thickness of 6 mm.

A three-dimensional turbo FLASH T1-weighted (axial) high-resolution anatomic scan was obtained at the end of the first experiment for each volunteer for coregistering the individual volunteer scans to a common standard template.[21] Imaging parameters were as follows: repetition time, 20 ms; echo time, 4.6 ms; flip angle, 12°; field of view, 256 × 256 mm; matrix, 256 × 256. Sixty-four contiguous slices of 3 mm were taken.

Statistical Analysis

Behavioral Data. Subjective pain scores, under the different drug conditions, for intensity and averseness were compared with use of a single-tailed paired Student *t* test based on the *a priori* hypothesis. Reaction times to the motor task were analyzed with a two-tailed Student *t* test. A value of $P \leq 0.05$ was taken as statistically significant.

Imaging Data.

First-level Analysis. The imaging data were analyzed to identify regions exhibiting significant changes in BOLD signal,[22] using a multistage process by the analysis package FSL (Functional Magnetic Resonance Imaging of the Brain Software Library#; version 3.0, release date October 2002).[23] The following prestatistical processing was applied from within the FSL package: motion correction using McFLIRT (Motion Correction Functional Magnetic Resonance Imaging of the Brain Linear Image Registration Tool),[24,25] spatial smoothing using a Gaussian kernel of full-width half maximum of 5.0 mm, mean-based intensity normalization of all volumes by the same factor, and nonlinear high-pass temporal filtering (set at 100 s). The data were modeled with use of FILM (Functional Magnetic Resonance Imaging of the Brain Improved Linear Model).[26] This uses a general linear modeling approach[27] to fit a model describing the experimental design to the imaging data so that the areas of the brain activated in response to the stimulus can be identified. FILM uses a robust and accurate nonparametric estimate of time series autocorrelation to prewhiten the time series of each voxel[26]; this improves estimation efficiency. Model fitting generates a parameter estimate image for each stimulus type. For each voxel, the higher the parameter estimate is, the more strongly the data correspond to the stimulus waveform, implying activity in response to that stimulus. The parameter estimate for each stimulus type is proportional to the fMRI signal change associated with that stimulus.

Second-level Analysis. A second level of analysis of image data was then performed with use of FEAT (fMRI Expert Analysis Tool, a software subset of FSL). Registration of functional image data sets to high-resolution structural and Talairach standard images was performed with use of FLIRT.[24] A fixed-effect group analysis was performed to combine the first-level analysis for individual subjects. This generated, for each ketamine dose, a map of brain activity associated with each stimulus type that was representative of the group of volunteers. The thresholding parameters used for the group fixed-effect analysis were a group Z score of greater than 2.3 at each voxel and then a further significance threshold of $P < 0.01$ applied to each cluster of voxels exceeding the Z threshold, hence addressing the problem of multiple comparisons.[28–30]

A further quantitative analysis of the dose-dependent change in brain activity within brain regions of interest was performed for the group. The regions of interest were defined in two ways. The first method was to create a "global functional mask" from the saline (zero ketamine) fixed-effect group activation maps for each stimulus type: pain, auditory, and motor. The global functional masks were used to represent the total significant brain activity for a paradigm, which was compared with the behavioral results.

In the second approach, pain activity was further investigated by defining the following anatomic regions of interest known to be involved in pain processing: thalamus, insular cortex, anterior cingulate cortex, and primary somatosensory cortex. These were manually defined from the Talairach atlas[21] with use of Talairach coordinates on the standardized brain image. The limits of these anatomic masks are tabulated in table 1. The

FSL. Available at: www.fmrib.ox.ac.uk. Accessed October 1, 2002.

Table 1. Limits of Anatomic Masks

	Talairach Coordinates		
	x	y	z
Thalamus, left	−2, −20	−28, 0	0, +16
Thalamus, right	+2, +20	−28, 0	0, +16
Insula, left	−26, −68	−36, +34	−4, +16
Insula, right	+26, +68	−36, +34	−4, +16
Anterior cingulate	−15, +15	+2, +58	−8, +46
Primary sensory, left	−4, −68	−4, −46	+24, +76
Primary sensory, right	+4, +68	−4, −46	+24, +76

The anatomic masks used were irregular, three-dimensional objects that lie within the quoted outer limits. Coordinates are given in millimeters.

resulting masks were applied to standardized individual subjects' data in Talairach space. The mean of the parameter estimates within the defined regions of interest (functional or anatomic) for the three drug conditions for each subject was then calculated. A group mean and SE, across the volunteers, of the pain, auditory, and motor parameter estimates, representing stimulus-induced signal changes, were thus calculated.

Statistical Analysis of Imaging Results. The regional mean parameter estimates were compared by performing a one-factor, within-subjects analysis of variance to check for heterogenicity of covariance, using the general linear model in SPSS version 9.0 (SPSS Inc., Chicago, IL). If there was homogenicity of covariance, paired Student *t* tests were performed. Single-tailed *P* values for the *t* statistic are quoted for those sites where there was an *a priori* hypothesis (analgesia reduces activity in sites processing noxious stimuli). The auditory data and motor data were included in this study for comparison with the pain results, so two-tailed *P* valves are given because these were *post hoc* calculations.

Results

Neurobehavioral Effects

The lower dose of ketamine (predicted plasma concentration of 50 ng/ml) produced barely perceptible subjective effects. At a predicted plasma concentration of 200 ng/ml, all volunteers noted mild changes, such as feeling sleepy, drunk, light-headed, or dizzy; a feeling of distortion of the arms or face; numbness; and heaviness. Within the scanner, the effects were heightened. There were no reported visual hallucinations. All subjects were able to maintain verbal contact throughout the study and were able to describe their experiences clearly afterward. Memory was not tested, but subjects apparently had complete recall of these events.

Thermal Noxious Stimuli

The behavioral pain data are displayed in figure 2, showing the mean and SE of pain intensity and averseness at each dose of ketamine. The high-dose ketamine infusion (200 ng/ml predicted plasma concentration) produced a significant decrease in both pain intensity and averseness compared with saline. Pain intensity decreased from a score of 7.8 ± 0.13 (mean ± SEM) during the saline infusion to 5.0 ± 0.7 ($P = 0.004$) at 200 ng/ml ketamine. That is a decrease from strong pain to mild pain. Pain averseness decreased from 4.9 ± 0.8 (distressing) during saline to 1.9 ± 0.6 (discomforting) ($P = 0.004$) at 200 ng/ml ketamine. The pain scores at the threshold dose of 50 ng/ml ketamine were not statistically different from those associated with the saline dose. The pain intensity score decreased from 7.8 ± 0.1 during the saline infusion to 7.4 ± 0.2, and the averseness score decreased from 4.9 ± 0.8 during the saline infusion to 4.6 ± 0.8.

The pain imaging data are shown in the color-rendered image (fig. 3, A–C), which displays the response of the brain to the thermal noxious stimuli at the three drug doses. Characteristic sites are activated, such as the thalamus, insular cortex, anterior cingulate cortex, and primary sensory cortex. These images show strong insula activation, with relatively little activity in the primary sensory cortex. The Talairach coordinates for the voxels

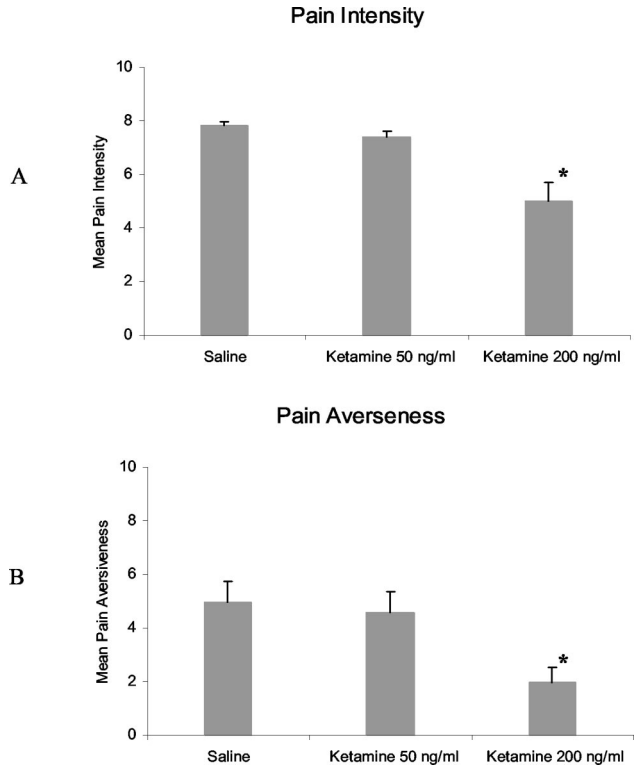

Fig. 2. Mean pain scores for the three drug conditions for pain intensity (*A*) and pain averseness (*B*). Mean pain scores (n = 8) are plotted with *error bars* showing the SEM. At 50 ng/ml ketamine, there is a small, nonsignificant decrease in pain intensity and pain averseness. The decrease at 200 ng/ml ketamine is statistically significant (intensity, $P = 0.004$; averseness, $P = 0.004$ compared with saline). * Statistically significant decrease in pain score in comparison with saline.

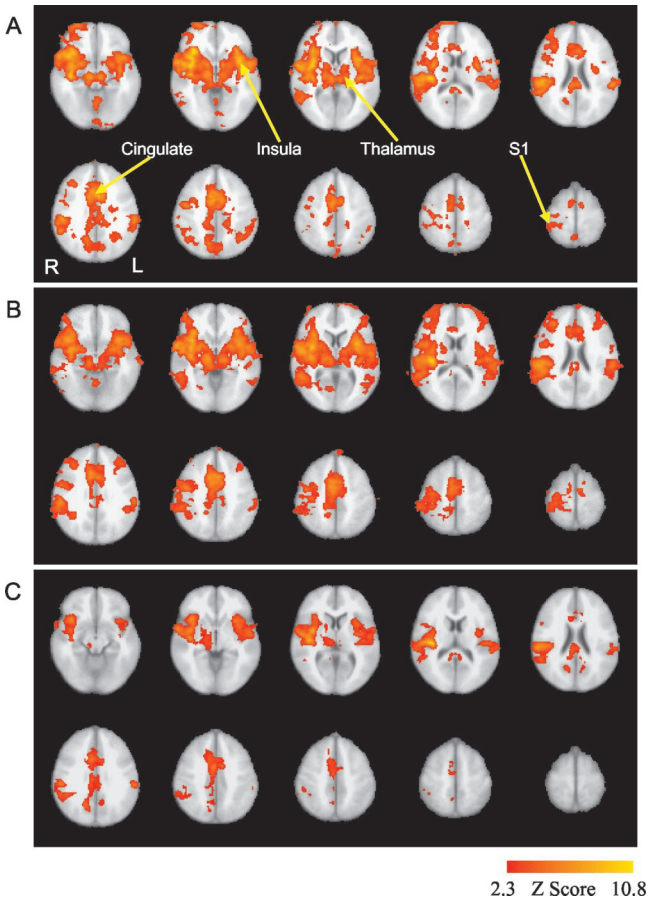

Fig. 3. Fixed-effect group activation in response to nociceptive stimulation during saline infusion (*A*), ketamine predicted plasma concentration of 50 ng/ml (*B*), and ketamine predicted plasma concentration of 200 ng/ml (*C*). The images consist of a color-rendered statistical map of activation (Z scores) registered onto high-resolution structural scans that have been transformed to standardized brain geometry (Talairach space). For clarity, 10 representative images are shown. The standard threshold for voxel activation was taken as Z > 2.3 ($P < 0.05$), and clusters were defined with a significance threshold of $P = 0.01$. Color is used to display the Z scores for voxel activation, from *red* (lowest Z) to *yellow* (highest Z). Anatomic left (*L*) and right (*R*) are marked and with labels indicating relevant anatomic sites.

with the maximum Z score within each anatomic region are given in table 2.

The magnitude of the global activation to the thermal stimulus is shown in figure 4, which illustrates the group average parameter estimates in response to pain, in the functionally defined pain region of interest. The BOLD response to pain shows a nonsignificant decrease at low-dose ketamine compared with saline (saline *vs.* 50 ng/ml ketamine, $P = 0.16$) but a significant reduction at the higher (200 ng/ml) dose of ketamine (saline *versus* 200 ng/ml ketamine, $P = 0.02$).

Figure 5 shows the BOLD signal change in response to painful stimulation within anatomic regions of interest. The limits of the anatomic masks used to define these areas are defined in table 1. At the maximum ketamine dose (200 ng/ml), there was a significant reduction in the magnitude of the BOLD signal response to painful stimulation compared with saline in the thalamus (right, $P = 0.03$; left, $P = 0.045$) and insular cortex ($P = 0.03$, right and left). However, a non–statistically significant reduction was seen in the anterior cingulate cortex and the primary sensory cortex. The pattern of changing activity with increasing ketamine concentration of the primary sensory cortex was different from the other anatomic sites. It showed lower signal changes in response to painful stimuli, with a less clear match of changing fMRI response to the behavioral data with increasing drug concentration.

To provide an alternative method of comparison of the effects of ketamine, for each anatomic region at each drug dose, the voxel with the maximum Z value was located. Table 2 provides the Talairach coordinates and Z values of these voxels. A blank result indicates that no voxel within that region achieved the significance threshold of Z = 2.3.

Auditory Stimuli

No subjects reported a perceptual change in the noise of the scanner at 50 ng/ml, but all subjects described the scanner as sounding different at 200 ng/ml. However, we were unable to quantify this perceptual change.

The effects of ketamine on the imaging results can be seen in figure 4, which shows the group average parameter estimates in response to the auditory stimuli in the functionally defined auditory regions. This shows a nonsignificant increase at 50 ng/ml ketamine ($P = 0.057$) followed by a significant decrease at 200 ng/ml ketamine (saline *versus* 200 ng/ml ketamine, $P = 0.001$). Table 2 provides the coordinates and Z value of the most active voxel for each drug dose in response to the auditory stimulus.

Motor Task

The behavioral results for the effects of ketamine on the motor task can be analyzed in two ways: comparing the number of button-press errors, or comparing the reaction time for the task. The number of button-press errors in performing the motor experiment was low (between 1 and 5) at each dose. The total number of errors made by all the volunteers at each dose was the same—13, *i.e.*, on average, 3.25 errors out of a potential maximum of 240 errors/session. The mean reaction time to press the button during the saline infusion was 465 ± 2.9 ms (mean ± SEM), which increased to 483 ± 6.3 ms and 503 ± 4.8 ms at the 50- and 200-ng/ml predicted plasma concentrations, respectively. The increases in reaction times represent 4% and 8% increases above the saline values for the 50- and 200-ng/ml predicted plasma concentrations, respectively. These increases are statistically significant: $P = 0.04$ for saline *versus* 50 ng/ml ketamine and $P < 0.001$ for saline *versus* 200 ng/ml ketamine (two-tailed *t* test).

Table 2. Regional Maximum Z Scores

Anatomic Region	Saline				50 ng/ml Ketamine				200 ng/ml Ketamine			
	Talairach Coordinates			Maximum Z Score	Talairach Coordinates			Maximum Z Score	Talairach Coordinates			Maximum Z Score
	x	y	z		x	y	z		x	y	z	
Pain paradigm												
Insula, right	34	26	−2	10.1	38	−20	14	10.8	38	−20	14	8.8
Insula, left	−36	10	−4	8.1	−34	8	2	9.0	−54	−6	−2	5.4
Anterior cingulate	−2	14	32	7.6	10	14	34	8.7	4	2	40	5.1
Thalamus, right	6	−20	4	6.0	14	−18	−2	7.6	12	−12	−2	4.2
Thalamus, left	−6	−18	2	5.8	−14	−18	−4	6.6	−6	−22	10	2.7
Primary sensory, right	36	−20	60	4.7	56	−26	24	7.8	56	−24	26	6.3
Primary sensory, left	−62	−26	28	5.6	−58	−24	26	6.6	−60	−26	28	5.4
Prefrontal, right	36	40	14	4.5	42	56	18	4.5	—	—	—	< 2.3
Auditory paradigm												
Superior temporal gyrus, left	−52	−20	0	10.3	−54	−20	2	12.4	−54	−22	2	9.9
Superior temporal gyrus, right	58	−40	6	8.4	60	−14	−2	10.5	62	−14	0	8.4
Motor paradigm												
Cerebellum, right	22	−56	−34	4.7	22	−56	−32	4.6	—	—	—	< 2.3
Postcentral gyrus, left	−44	−32	60	4.6	−44	−32	60	4.9	−36	−18	64	4.5

Maximum Z scores for each paradigm at different ketamine doses in different anatomic regions. Talairach coordinates are given in millimeters.

The imaging results for the motor task are displayed in figure 4, which shows the group average parameter estimate in response to the motor stimuli in the functionally defined motor regions. The imaging response to the motor task shows non–statistically significant decreases at 50 and 200 ng/ml. Saline *versus* 200 ng/ml ketamine yielded a *P* value of 0.5, and saline *versus* 50 ng/ml ketamine yielded a *P* value of 0.7 (two-tailed *t* test). As with the other paradigms, table 2 provides data on the most activated voxel in response to the motor task. A blank result indicates that no voxel exceeded the threshold Z value of 2.3 within that anatomic region.

Discussion

We have compared the subjective reduction in pain perception (analgesia) produced by different doses of ketamine with objective changes in activity of key brain areas during noxious thermal stimulation. To specifically isolate the analgesic effects of ketamine from generalized anesthetic effects, we compare this with the effect of ketamine on non-noxious stimuli: responses to an auditory stimulus and a motor task. Therefore, our aim was to measure the specific effects of ketamine on cerebral processing of noxious stimuli.

At a predicted plasma concentration of 200 ng/ml, ketamine had a significant analgesic effect, reducing the behavioral pain score from strong pain (heat pain score 7.8) to mild pain (heat pain score 5), and the measured fMRI signal (parameter estimate) was statistically significantly reduced from 97.8 to 36.9 (61% reduction). The imaging results are therefore consistent with the behavioral analgesic effects of ketamine.

Investigating drugs with use of fMRI is becoming more common; however, confounds exist that should be taken into account in any experimental design. The signal measured in fMRI (BOLD) is altered by local neuronal firing, which requires oxygen and alters local blood flow, thus changing the local concentration of deoxyhemoglobin.[22] An analgesic, as part of its primary effect, may reduce regional neuronal activity, thus reducing the BOLD signal related to the noxious stimulation. However, the drug, by binding to receptors, may also increase local metabolic consumption, thus increasing the BOLD signal. In addition, the drug may independently alter cerebral blood flow and cerebral blood volume, thus altering the BOLD signal. This is why it is crucial to have a simultaneous recording of the fMRI signal with a behavioral measure of the paradigm. A concurrence of behavioral and imaging results is a strong argument suggesting that the imaging is measuring the specific behavioral function in the brain. Alternative imaging techniques can help to unravel some of these confounds. Using optical imaging in monkeys, isoflurane reduces the magnitude of the global signal (analogous to the BOLD signal), and this reduction occurs in both the oxyhemoglobin and deoxyhemoglobin signal with no significant change in time course.[31] With the current fMRI experiment, we cannot distinguish these confounds, such as the difference between drug-induced changes in neuronal firing and drug-induced changes in cerebral blood flow. In the presence of a reduced behavioral pain score, a decreased activation to a noxious stimulus in a region of the brain that normally processes pain is most likely due to the drug binding to receptors, in these regions or functionally connected regions, reducing the capacity of the neurons to fire.

In addition, the effects of ketamine are not confined to analgesia, because it clearly has profound cerebral anesthetic properties. To investigate the specificity of ket-

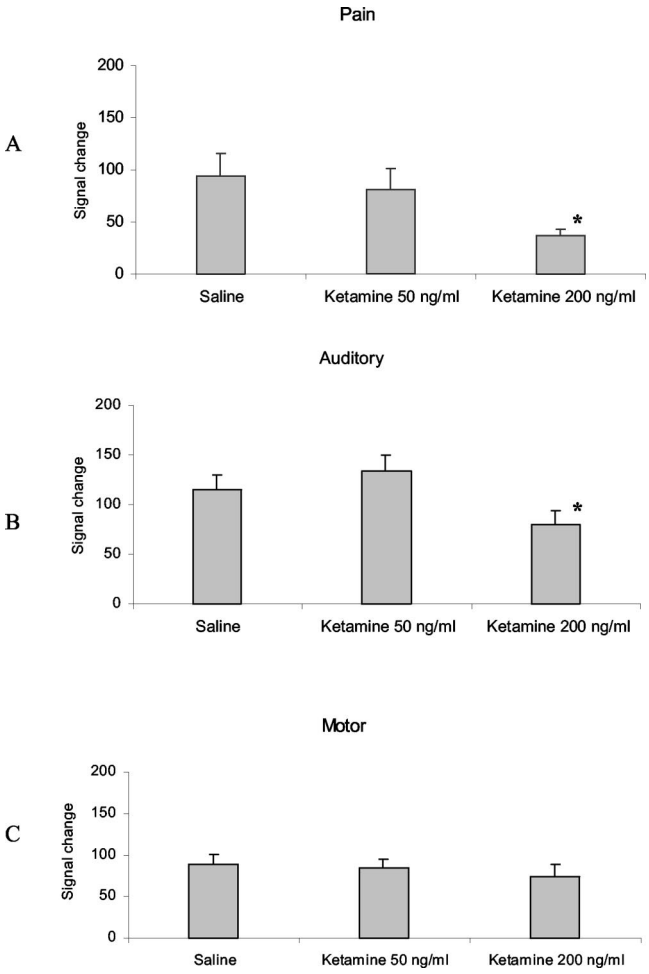

Fig. 4. Comparison of activation during pain (*A*), auditory stimuli (*B*), and motor tasks (*C*). These graphs show the level of activation within areas of the brain that respond to pain (*A*), auditory stimuli (*B*), or motor tasks (*C*) under the three drug conditions. Pain, auditory, and motor regions of interest were defined from the group activity map during the saline infusion. A mean parameter estimate within each mask for each subject at each dose was calculated. The displayed value is the average for the eight volunteers, with *error bars* showing the SEM. * Statistically significant change from the saline infusion.

amine on the processing of nociceptive stimuli, we investigated additional tasks. The imaging result on the processing of an auditory stimulus at different ketamine doses was complex: an increase at 50 ng/ml followed by a decrease in signal at 200 ng/ml. These results show that ketamine has effects beyond the processing of noxious stimuli. These results were difficult to interpret because we had no objective quantifiable behavioral measure of audition to compare with the imaging data. However, it is interesting to note the different pattern of response to the auditory stimuli compared with the other two paradigms at the different drug doses.

For this reason, we performed an additional experiment with a motor task, which allowed the comparison of imaging with behavioral results at different doses of ketamine. The reduced statistical power of using four subjects is offset by the larger number of data points collected for each subject, which produces similar SEs for the different paradigms. Ketamine had no effect on the number of button-press errors. The reaction times were statistically significantly increased, being increased by 4% and 8% at 50 and 200 ng/ml ketamine, respectively. This suggests that the ketamine at these doses causes only a minor degree of cognitive impairment. These behavioral results are comparable with the modest 6% and 17% reduction in the fMRI signal in motor areas of the brain, which is a non–statistically significant effect.

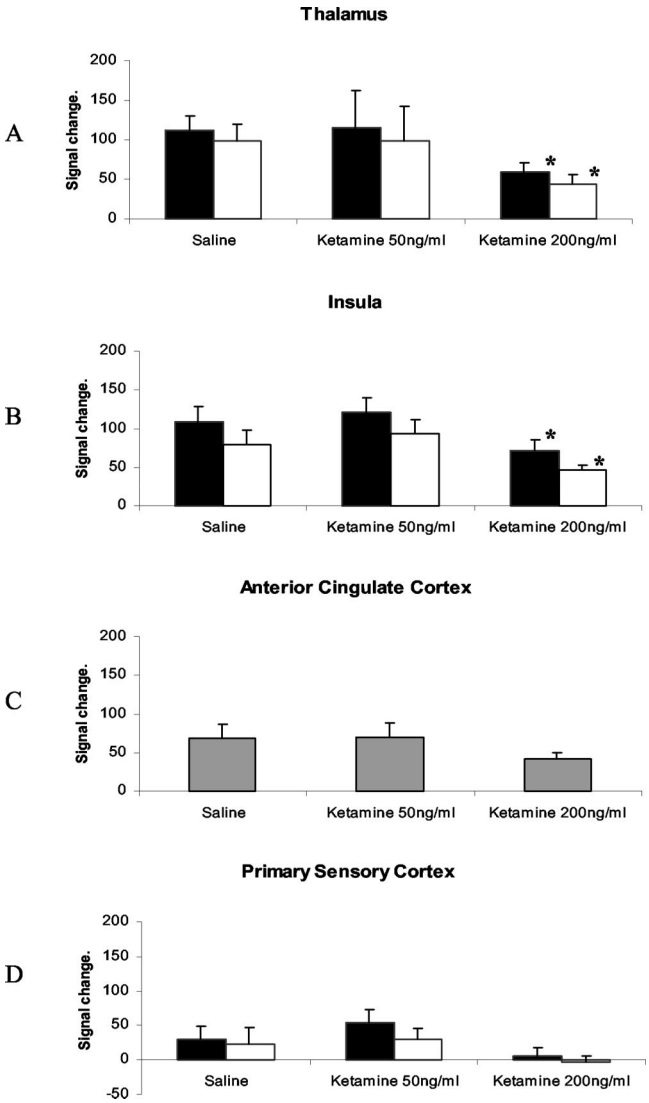

Fig. 5. Functional magnetic resonance signal change associated with noxious stimuli in anatomically defined regions of interest: thalamus (*A*), insular cortex (*B*), anterior cingulate cortex (*C*), and primary sensory cortex (*D*). The *vertical axis* represents a parameter estimate proportional to the signal change above baseline (in arbitrary units). The *horizontal axis* displays the predicted plasma concentration of ketamine. *Black bars* = anatomic right; *white bars* = anatomic left; *gray bars* = midline structures. * Statistically significant reduction in signal from saline.

Considering the results from these three paradigms, we can conclude that the effects we measured in the BOLD signal are unlikely to be solely due to a global effect on cerebral hemodynamics or cognition. This is because we have found differences in the imaging results between the different paradigms, differences in magnitude and direction of change. These differences show close similarities to their respective behavioral results. This concurrency between the different behavioral and imaging results for the different paradigms indicates specific rather than global effects of ketamine. In addition, early work by Breier *et al.*,[32] using fluorodeoxyglucose positron emission tomography and a task to control for mental activity, showed no significant difference in global metabolic activity between ketamine and placebo infusion. A recent study by Abel *et al.*,[33] using ketamine and fMRI with a cognitive task, found specific focal task-dependent effects of ketamine rather than global task-independent effects, which support the notion that ketamine does have specific effects that can be determined with use of fMRI. Therefore, these two studies support our findings that analgesic effects of ketamine can be distinguished from anesthetic effects with fMRI.

The order of administration of the three drug dosages could not be randomized without making the scanning times unacceptably long or introducing extra sessions. This raises the possibility of either a cumulative drug effect or alternatively adaptation or sensitization to the noxious stimulus. However, the drug dose range is sufficiently large (fourfold) to minimize a cumulative drug effect, and we have not seen adaptation or sensitization with this stimulus type in previous studies.

As expected, the BOLD activation to noxious stimuli was widespread and bilaterally activated with a small, consistent increase in the contralateral side.[34] At the higher dose of ketamine (200 ng/ml), there was a variability of response in different anatomic regions of interest. The thalamus and insula showed the greatest reduction in activation, and these two regions of the brain are known to be the regions most strongly associated with pain intensity.[10] The anterior cingulate has a complex role in pain processing, with subdivisions having not only different roles in pain processing but also demonstrating reciprocal inhibition.[35] Surprisingly, the reduction in activity in the anterior cingulate cortex did not reach statistical significance, although it did show a trend toward decreased activation.

The activation of the primary sensory cortex did not show a clear relation to the behavioral pain scores. Pain studies have shown variable activation of the primary sensory cortex, and this seems to be dependent on the size, position, and nature of the nociceptive stimuli.[35] We used a device that was built for the purpose of delivering the noxious stimulus. It is small, remains in a fixed position, and has the advantage of a very fast ramp time, 30–60°C in 0.8 s. This provides a stimulus with a relatively small sensory and a relatively large nociceptive component, which may explain the poor activation of the sensory cortex.

Interestingly, examination of the imaging data shows an area of prefrontal cortex (table 2) that is activated in response to noxious stimuli during the saline infusion and at 50 ng/ml ketamine but is no longer active at 200 ng/ml ketamine. These areas include regions that we have previously identified as having a role in processing the anticipation/anxiety component of pain.[36] We postulate that the decreased activation in this region at the higher dose of ketamine is due to a reduction in level of anticipation and anxiety associated with the painful stimulus.

Ketamine has analgesic effects at multiple sites within the nervous system, not only cortical, but also at the spinal cord. Separate imaging of the spinal cord and brain would be needed to discriminate the relative contribution of the effects of ketamine at these different central locations. However, it is clear that large and measurable decreases in activation to noxious stimuli occur within the brain that are variable across different pain processing areas, which cannot be explained purely by a altered input from the spinal level.

Ketamine is an anesthetic drug, and so it would be expected to have global cognitive effects. Behavioral pain scores are reduced under conditions of reduced vigilance to the noxious stimulus, and this has been imaged.[37,38] It is interesting to speculate as to whether part of the analgesic action of ketamine is by reducing vigilance to the task. However, distraction causes a much smaller reduction in the behavioral pain score and thus is unlikely to be more than a component of the analgesic effect of ketamine. In addition, we found no significant effects on the error rate measured during the motor task, which supports the notion that global cognitive effects were not a confound in this study.

The great power of this approach to the study of analgesia is the ability to simultaneously record behavioral subjective pain scores and the activity in specific regions of the brain while altering drug dose. From this, not only can analgesic effects be objectively measured, but also information can be gained about the mechanisms of analgesia. Analgesic effects can be separated from anesthetic effects. Further experience with different drugs will provide insight into the differing types of mechanisms by which analgesia can be produced in humans.

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