

Abnormal Activity in Reward Brain Circuits in Human Narcolepsy With Cataplexy

Aurélie Ponz, PhD,^{1,2} Ramin Khatami, MD,³ Rositsa Poryazova, MD,³
Esther Werth, PhD,³ Peter Boesiger, PhD,^{4,5} Claudio L. Bassetti, MD,³
and Sophie Schwartz, PhD^{1,2}

Objective: Hypothalamic hypocretins (or orexins) regulate energy metabolism and arousal maintenance. Recent animal research suggests that hypocretins may also influence reward-related behaviors. In humans, the loss of hypocretin-containing neurons results in a major sleep-wake disorder called narcolepsy-cataplexy, which is associated with emotional disturbances. Here, we aim to test whether narcoleptic patients show an abnormal pattern of brain activity during reward processing.

Methods: We used functional magnetic resonance imaging in 12 unmedicated patients with narcolepsy-cataplexy to measure the neural responses to expectancy and experience of monetary gains and losses. We statistically compared the patients' data with those obtained in a group of 12 healthy matched controls.

Results and Interpretation: Our results reveal that activity in the dopaminergic ventral midbrain (ventral tegmental area) was not modulated in narcolepsy-cataplexy patients during high reward expectancy (unlike controls), and that ventral striatum activity was reduced during winning. By contrast, the patients showed abnormal activity increases in the amygdala and in dorsal striatum for positive outcomes. In addition, we found that activity in the nucleus accumbens and the ventral-medial prefrontal cortex correlated with disease duration, suggesting that an alternate neural circuit could be privileged over the years to control affective responses to emotional challenges and compensate for the lack of influence from ventral midbrain regions. Our study offers a detailed picture of the distributed brain network involved during distinct stages of reward processing and shows for the first time, to our knowledge, how this network is affected in hypocretin-deficient narcoleptic patients.

ANN NEUROL 2010;67:190–200

Hypocretins (HCRT¹ or orexins²) are produced by specific neurons in the hypothalamus that have extensive projections throughout the central nervous system.^{3,4} Deficient HCRT transmission was found to be associated with narcolepsy-cataplexy (NC) in humans, dogs, and knockout mice,^{5,6} suggesting a main role for HCRT in sleep/wake regulation and arousal-maintenance.^{3,7} Clinically, human NC is characterized by excessive daytime sleepiness, fragmented nighttime sleep, sleep onset rapid eye movement periods, and sudden episodes of postural muscle atonia called cataplexy.⁸ Cataplexy attacks are predominantly triggered by emotional experiences, including

the anticipation of reward when playing games.^{8–10} These behavioral observations suggest possible interactions between the hypothalamic HCRT system and reward brain circuits in humans.^{11,12}

Recent studies in rodents provided evidence for anatomical and functional links between the HCRT system and the dopamine system, the latter being critically involved in reward processes and motivated behaviors.^{13–15} First, hypothalamic HCRT neurons project densely to reward-related brain regions, including the nucleus accumbens (NAcc) and the dopaminergic ventral tegmental area (VTA).¹⁶ Second, HCRT receptors are expressed at

Published online in Wiley InterScience (www.interscience.wiley.com). DOI: 10.1002/ana.21825

Received Feb 15, 2009, and in revised form Jun 24. Accepted for publication Jul 1, 2009.

Address correspondence to Dr Schwartz, Department of Neuroscience, University Medical Center, Michel-Servet 1, 1211 Geneva 4, Switzerland.

From the ¹Department of Neuroscience, University of Geneva, Geneva, ²Geneva Neuroscience Center, University of Geneva, Geneva, ³Neurology Department, University Hospital, Zurich, ⁴Biomedical Engineering, University of Zurich, Zurich, and ⁵Swiss Federal Institute of Technology, Zurich, Switzerland.

Additional Supporting Information may be found in the online version of this article.

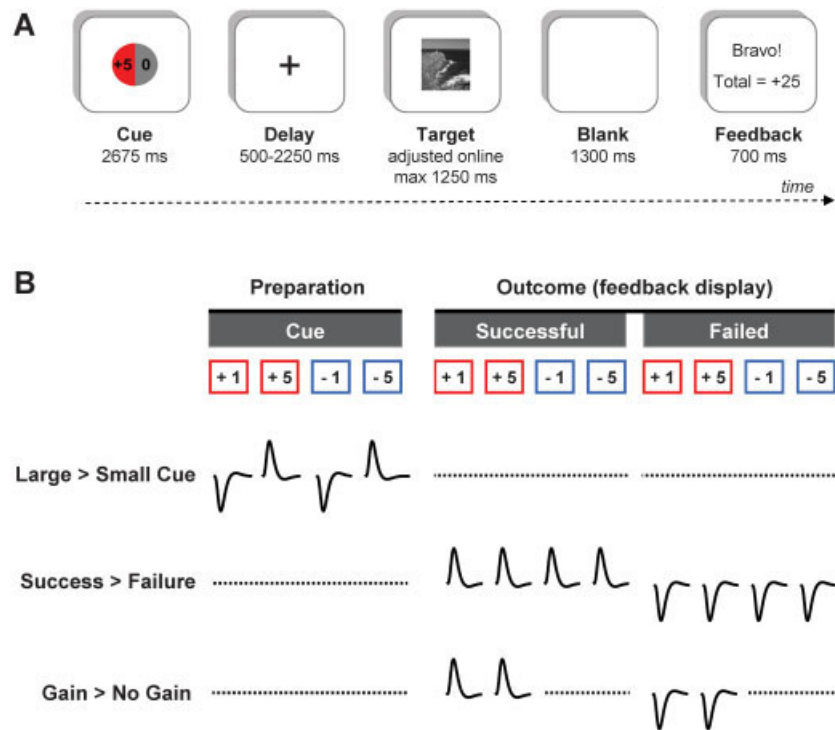


FIGURE 1: Task and main functional magnetic resonance imaging contrasts. (A) Illustration of 1 trial. Each trial started with a cue indicating how many points could be won or lost on that particular trial. The cue remained static on the screen for 1,550 milliseconds and then flickered for 1,125 milliseconds. A fixation cross was then presented during a variable interval before a target picture was briefly shown. Participants were instructed to press a button while the target picture was on the screen. To obtain a balanced proportion of won and lost trials in each condition, the duration of each target presentation was adjusted automatically by a tracking algorithm based on the subject's performance on the previous trial. If a trial was successful, then the duration of the target on the next trial was shortened (-25 milliseconds) to make the trial more difficult; if the trial was unsuccessful, then the duration of the target on the next trial was increased ($+25$ milliseconds). At the end of the trial, a feedback display indicated whether the participant successfully pressed during the target presentation, as well as the current balance in points. (B) Schematic representation of the 12 main orthogonal conditions included in our statistical model, with 4 regressors for the preparation period and 8 for the outcome period. The 3 main contrasts of interest are illustrated as events convolved with a canonical hemodynamic response function: 1) preparation to large versus small cues, 2) successful versus failed trials, 3) positive gains versus no positive gain. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

the surface of VTA dopamine neurons,^{17,18} and HCRT administration increases the firing rate of VTA neurons.¹⁹ Third, HCRT is involved in drug-seeking behaviors and associated mesolimbic dopaminergic activity.^{18,20–23} These recent findings in rodents suggest that the HCRT system regulates dopamine activity in brain reward circuits and impacts on the expression of motivated behaviors and addiction.

Although human NC is linked to hypothalamic HCRT depletion,^{5,6} it remains unknown whether NC patients show altered brain reward processing. Scarce behavioral evidence exists for reduced drug addiction in NC patients, as these patients are often treated with addictive amphetamine-like drugs but rarely become addicted to these drugs.^{8,24–26} NC patients may also present with some psychiatric disturbances.²⁷ In the present study, we tested whether human NC entails a dysfunction in brain reward circuits. We acquired whole-brain functional mag-

netic resonance imaging (fMRI) data in 14 unmedicated, nondepressed patients with clear-cut cataplexy and 14 healthy matched controls (Supplementary Table). During scanning, the subjects performed an adapted version of a monetary incentive delay task, which is known to powerfully activate the mesolimbic and midbrain reward system (Materials and Methods, Fig 1).^{28–30} As suggested by rodent studies (see above), we hypothesized that HCRT deficiency in NC patients might lead to altered reward-related responses in ventral midbrain/VTA regions during reward expectancy. We also expected that neural responses to reward experience in the mesolimbic reward system would be affected in NC patients. Finally, based on our recent study on humor processing in NC patients,¹¹ we anticipated abnormal increases in amygdala activity during positive emotions elicited by our game-like task, that is, during winning trials associated with high gains. By confirming the hypotheses above, our fMRI re-

sults provide strong evidence for abnormal brain responses to reward in NC patients, and thus suggest an implication of HCRT activity in the regulation of brain reward function in humans.

Materials and Methods

Patients and Controls

Fourteen drug-free narcoleptic patients with clear-cut cataplexy and 14 healthy controls matched for age, sex, handedness, and body mass index participated in this study (Supplementary Table and Supplementary Methods). Twelve patients (and their matched controls) were included in the final statistical analyses. HLA-DQB1*0602 was positive in all 12 patients. Hypocretin-1 levels (<120pg/ml) in the cerebrospinal fluid could be determined in 8 NC patients and was confirmed to be low or undetectable in all of them.^{6,31}

Experimental Paradigm

During fMRI scanning, the subjects performed a visuomotor game-like task, in which they could win (or lose) points if they rapidly pressed on a key while a visual target was briefly shown (Fig 1A; Supplementary Methods). Each trial started with a preparation period, during which the subjects saw a cue indicating the potential gain (+1 or +5 points) or potential loss (−1 or −5 points) associated with that particular trial. After a variable delay, the visual target that required a rapid key press was briefly presented on the screen. The trial ended with a feedback display, telling the subjects whether they had won or lost the trial. This paradigm allowed us to distinguish between a preparation period (presentation of the cue) and an outcome period (presentation of the feedback). In addition, we could compare trials with high and low incentives (large versus small cues), as well as successful versus failed trials (Fig 1B). To ensure a fixed proportion of successful trials (50–60%), a tracking algorithm adjusted the duration of each target presentation (ie, task difficulty) based on each subject's current performance.

Magnetic Resonance Imaging Methods

We acquired fMRI data on a Philips Intera 3.0 T whole-body system (Philips Medical Systems, Best, The Netherlands) across 4 scanning sessions (229 MRI volumes each) separated by brief pauses (Supplementary Methods). The fMRI data were analyzed using the standard general linear approach in SPM (www.fil.ion.ucl.ac.uk/spm). The statistical model included 12 main regressors of interest: 4 regressors corresponding to the presentation of the 4 possible cues during the preparation period, and 8 regressors corresponding to the presentation of the feedback during the outcome period (winning or losing on the 4 different cue types; Fig 1B). Individual contrast images between conditions of interest were calculated and entered into random-effects group analyses, using analyses of variance (ANOVAs) as implemented in SPM.³² Common group effects were assessed using conjunction analyses to reveal voxels that showed significant activity increase in both the patient and control populations.³³ Direct group comparisons were performed using an exclusive masking

procedure to reveal voxels that showed increased responses for a particular contrast in 1 population but not in the other. Using dedicated connectivity methods, we also assessed changes in the functional coupling between a main region of interest (ventral midbrain/VTA region) and any other brain region as a function of cue value.³⁴ Finally, we tested for correlations of brain activity with clinical characteristics in NC patients (disease duration, cataplexy, and sleep propensity scores) using regression analyses.

Results

Behavioral Results

REACTION TIMES. We analyzed the reaction times (RTs) on successful trials using an ANOVA, with Cue Value (1 point, 5 points) and Cue Valence (positive, negative) as within-subject factors, and Group (NC patients, controls) as a between-subject factor (Supplementary Methods). This statistical analysis revealed that RTs during the task did not differ between the groups (NC patients: 226.84 milliseconds, standard deviation [SD] 31.43; controls: 228.38 milliseconds, SD 23.36; $F_{1,22} = 0.02$, nonsignificant [ns]). RTs were also not influenced by the value of the cue ($F_{1,22} = 0.30$, ns), nor by the valence of the cue ($F_{1,22} = 0.47$, ns). There was no interaction between these factors. Note that RTs also influenced the duration of target presentation, because the online tracking algorithm controlled the success rate on this reaction-time task by adjusting the target duration during the task. As expected, from the RT results, we found that the average presentation times for the target did not differ between NC patients and controls ($F_{1,22} = 0.92$, ns).

SUCCESSFUL TRIALS (HITS). We analyzed the number of correct responses (hits) using an ANOVA with the same factors as for the RTs above. There was no main effect of Group on hit rates (patients: 57.29%, SD 11.8; controls: 58.33%, SD 9.96; $F_{1,22} = 0.77$, ns), with both groups achieving better scores for highest incentive trials (1 point: 54.79%, SD 9.72; 5 points: 60.83%, SD 11.21; $F_{1,22} = 6.16$, $p < 0.05$), as well as for potential losses (gains: 55.31%, SD 10.35; losses: 60.31%, SD 10.88; $F_{1,22} = 7.87$, $p < 0.01$). There was a significant interaction of Valence by Group ($F_{1,22} = 6.79$, $p < 0.01$), due to NC patients being less successful on positive than negative trials (positive: 52.5%, SD 9.66; negative: 62.08%, SD 11.97), unlike controls (positive: 58.13%, SD 10.5; negative: 58.54%, SD 9.6).

FAILED TRIALS. Failed trials in this task occurred whenever the subject pressed the response key shortly after the target picture had disappeared from the screen (<1,300 milliseconds after target presentation). Controls produced more such errors than NC patients (main effect

of Group, patients: 40.53%, SD 11.29; controls: 46.35%, SD 8.57; $F_{1,22} = 11.89$, $p < 0.01$). There was no effect of Cue Value but a main effect of Cue Valence, because failed trials were slightly more frequent with positive than negative cues (positive: 45.91%, SD 9.66; negative: 40.97%, SD 10.60; $F_{1,22} = 14.64$, $p < 0.001$). However, this valence effect was not due to a general slowing of the responses after positive cues, as there was no effect of Valence on RTs. There was no interaction between these factors.

Taken together, these behavioral results show that NC patients could achieve similar reaction times as controls during our game-like task, which is remarkable given that the patients had either no chronic treatment or had to withdraw from medication. Moreover, both populations obtained similar final outcomes, ranging from 21.5 to 24.8 points (Mann-Whitney $U = 61$, $p = 0.52$). Mostly comparable performance for both groups ensures that observed differences in brain activity between the groups are not due to major differences in performance. Importantly, the online tracking algorithm efficiently controlled the proportion of successful and failed trials, as well as their balanced distribution across conditions in each group, which guarantees adequate statistical power for valid inference from the fMRI results.

FMRI Results

STANDARD SPM ANALYSES.

Preparation period: brain response to high motivational cues. To identify brain regions more activated during reward expectancy, we compared brain activity during the presentation of large (+5/−5 points) versus small (+1/−1 point) cue values (Fig 1B; Supplementary Fig). Both NC patients and controls activated a network of brain regions involved in the expectation of a reward, including the ventral striatum (Table 1).^{29,30,35,36} Both populations also activated the amygdala, the anterior insula, and the anterior cingulate cortex, consistent with motivational processes enhancing attention and autonomic reactivity.^{37,38} As expected from augmented visual attention and motor preparation for the highest incentives in this visuomotor task, both groups showed increased activation in visual and motor regions. When directly comparing the groups, we found increased fMRI signal in controls in a region of the ventral midbrain, compatible with the VTA (−3x, −24y, −21z; Fig 2; Table 1).^{13,14,39} Activity increases were also observed in the medial prefrontal cortex and caudate nucleus, which are target regions for dopaminergic VTA projections and are involved in reward prediction.⁴⁰ No such modulation of activity by Cue Value was observed in these brain regions for NC

patients. When comparing NC patients to controls for the same fMRI contrast, we found increased activity in the insula and inferior orbitofrontal cortex for higher cue values.

Outcome period: brain response to successful trials.

To test for brain activity increases during winning on both positively and negatively cued trials, we compared any successful trials (ie, gains and no-losses) to failed trials (ie, no-gains and losses) at the onset of the feedback display (Fig 1). Both NC patients and controls showed increased activity in the dorsal striatum (caudate nucleus), as well as in a limbic region corresponding to the sublentiform extended amygdala (SLEA; Table 2). Group comparisons revealed increased activity in ventromedial prefrontal cortex (vmPFC) and NAcc during successful trials in the controls only (Fig 3A). Both these regions are known to be involved in regulating emotional processes and reward,^{14,41–43} and also receive dense dopaminergic projections from the VTA.⁴⁴ As suggested by recent animal work,^{18,20–22} HCRT depletion may affect dopaminergic modulation within brain reward networks, which would also be consistent with the observation that the midbrain/VTA was not responsive to highly motivating cues in NC patients (see above). By contrast, NC patients showed activity increases in the right dorsal striatum (putamen) and inferior lateral frontal cortex (Fig 3B).

Outcome period: brain response to successful positively cued trials.

NC patients report cataplexy when they experience strong, usually positive emotions such as when joking or winning games.^{9,10} Our game-like task also elicited positive emotions, particularly when winning on rewarded trials (ie, for actual gains on positively cued trials). Note that this condition did not trigger cataplexy in the patients during scanning. To assess changes in regional brain activity during this positive emotional condition, we compared winning on positively cued trials to failing on such trials (Fig 1). In both NC patients and controls, winning on positively cued trials activated the anterior cingulate cortex and the vmPFC, encompassing regions involved in motivation regulation (see Table 3 for a detailed list of activation clusters).^{41,45} When directly compared to NC patients, controls showed increased activity in the NAcc during actual gains and in the right lateral PFC. Conversely, NC patients showed further increases in amygdala/SLEA activity (Fig 4), which is in line with our recent fMRI study using humorous stimuli.¹¹ They also showed increased activity in the dorsal striatum (putamen; consistent with the result of the previous contrast reported above).

TABLE 1: Preparation Period: Brain Response to High Motivational Cues (Large > Small Cue Values)

Regions	L/R	MNI Coordinates			BA	Z Score
		x	y	z		
Patients and controls (conjunction)						
Occipital pole (lingual gyrus)	L	-18	-87	-18	18	4.74
Frontoinsula	L	-30	21	3	48	4.56
Putamen	L	-30	3	3	48	3.66
Occipital cortex	R	24	-99	-6	18	4.54
SMA	R	0	-12	60	6	4.28
Middle occipital cortex	L	-15	-93	18	18/17	4.09
Insula	R	33	18	-9	47	4.04
Ventral striatum (NAcc, putamen)	R	18	6	0	—	3.98
Motor cortex/precentral gyrus	L	-33	-18	66	6	3.91
Motor cortex	L	-42	-15	42	6	3.91
Amygdala	R	15	-3	-12	34	3.91
Medial PFC / ACC	R	12	51	6	10	3.85
Orbitofrontal cortex	L	-30	57	-6	11	3.71
Ventral striatum (NAcc)	L	-12	12	-15	25	3.36
Pre-SMA	R	3	27	57	6	3.27
Controls masked by patients						
SMA	R	9	-3	48	6	5.27
Cerebellum	L	-6	-66	-15	—	5.26
Supramarginal gyrus	L	57	-45	33	48	5.17
Caudate nucleus (body)	R	9	9	9	—	4.99
	L	-12	0	15	—	3.99
Medial PFC/ACC	L	-6	42	6	32	4.88
Middle occipital	R	27	-81	30	19	4.85
Ventral midbrain (including VTA)	L	-3	-24	-21	—	4.44
Patients masked by controls						
Occipital cortex	L	-33	-90	3	18	3.92
Inferior/orbito frontal cortex	R	48	36	-18	47	3.77
	L	-39	33	-9	47	3.66
Inferior frontal, insula	R	39	30	0	47	3.56

Stereotactic coordinates correspond to the standard MNI brain. Reported regions survived a threshold level of $p < 0.001$ uncorrected. Mask threshold for group comparisons was set at a conservative level of $p < 0.05$.
L = left; R = right; MNI = Montreal Neurological Institute; BA = Brodmann area; SMA = supplementary motor area; NAcc = nucleus accumbens; PFC = prefrontal cortex; ACC = anterior cingulate cortex; VTA = ventral tegmental area.

FUNCTIONAL CONNECTIVITY ANALYSIS. To further refine the functional brain network involved in the processing of high motivational cues, we conducted a functional connectivity analysis (Materials and Methods), which revealed increased functional coupling between midbrain/VTA and left NAcc (medial part, $-3x$, $12y$, $-12z$) selectively during the presentation of large cues in

controls. This result is in line with studies indicating that neuronal activity in VTA and nucleus accumbens may increase in proportion to the magnitude of anticipated reward.^{13,39,40}

REGRESSION ANALYSES. A clinically relevant question was whether any regional change in fMRI signal

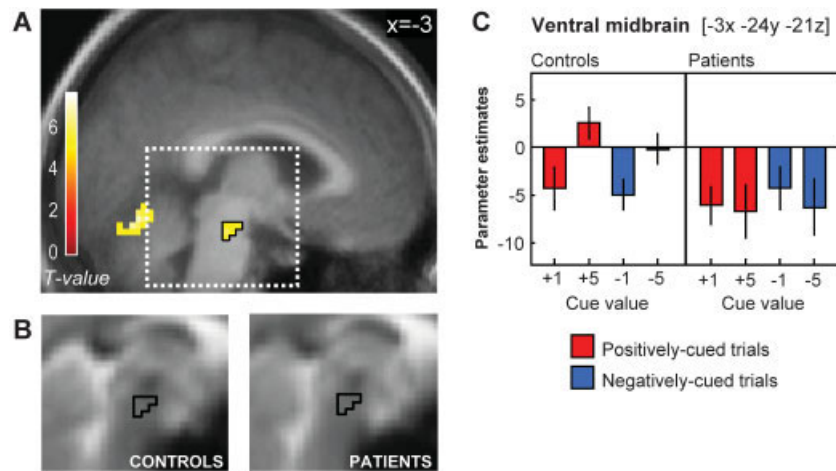


FIGURE 2: Ventral midbrain activity to large cues. (A) Increased response to large versus small cues in ventral midbrain (including ventral tegmental area) in controls masked by narcolepsy-cataplexy (NC) patients (exclusive masking procedure; see Materials and Methods). The statistical map is overlaid on a mean-normalized T1 structural scan; threshold $p < 0.001$ and mask threshold $p < 0.05$. (B) To visualize the precise localization of the activation peak and the absence of signal loss in this region, the outline of the cluster of activation is overlaid on the mean-normalized echo planar imaging (EPI) images of each population separately (controls on the left and NC patients on the right). (C) Parameter estimates extracted from the peak of this region ($-3x, -24y, -21z$) illustrate a selective activation for highest cue values in controls but not in NC patients. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

TABLE 2: Outcome Period: Brain Response to Winning (Successful > Failed Trials)

Regions	L/R	MNI Coordinates			BA	Z Scores
		x	y	z		
Patients and controls (conjunction)						
Amygdala/SLEA	R	18	3	-15	34	3.73
Caudate nucleus (head)	R	18	30	0	—	3.41
Controls masked by patients						
Ventromedial PFC	R	3	57	-6	10	4.08
Cerebellum	L	-15	-75	-27	—	3.98
Lateral PFC	L	-42	48	6	46	3.98
Precuneus	L	-30	-75	33	19	3.91
Caudate nucleus (body)	R	15	6	24	—	3.73
Ventral striatum (NAcc)	L	-21	15	-12	11	3.56
	R	9	12	-15	25	3.54
Middle occipital cortex	R	30	-78	27	19	3.51
Inferior occipital cortex	R	27	-90	-6	18	3.51
Patients masked by controls						
Putamen (dorsal)	R	24	6	9	—	3.73
Inferior lateral frontal cortex	R	45	36	9	45	3.41

Stereotactic coordinates correspond to the standard MNI brain. Reported regions survived a threshold level of $p < 0.001$ uncorrected. Mask threshold for group comparisons was set at a conservative level of $p < 0.05$. L = left; R = right; MNI = Montreal Neurological Institute; BA = Brodmann area; SLEA = sublenticular extended amygdala; PFC = prefrontal cortex; NAcc = nucleus accumbens.

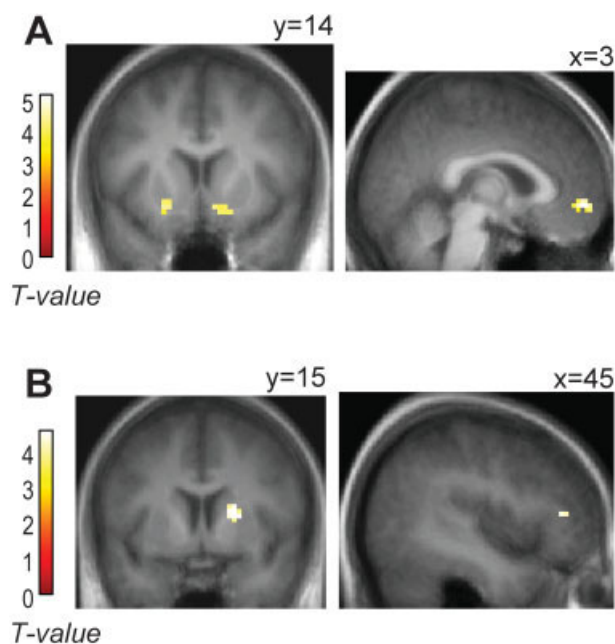


FIGURE 3: Brain responses to successful trials during the outcome period. (A) Increased response to successful trials in bilateral nucleus accumbens and in ventromedial prefrontal cortex for controls relative to narcolepsy-cataplexy (NC) patients. (B) Increased functional magnetic resonance imaging signal in the right dorsal striatum (putamen) and in the inferior lateral frontal cortex in NC patients compared with controls. Statistical maps are overlaid on mean-normalized T1 structural scans; statistical threshold $p < 0.001$. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

could relate to the patients' individual clinical characteristics. We found that response to large cues in the left NAcc ($-9x, 12y, -6z$; $R^2 = 0.82, p < 0.001$) and the vmPFC ($-3x, 42y, -12z$; $R^2 = 0.85, p < 0.001$) correlated positively with disease duration (Fig 5). Critically, although activity in these regions did not reach significance in the group of NC patients for the contrast of interest (large versus small cue values), it was significant in controls. This suggests a recovery of activity in these reward-related regions for patients who had long been suffering from narcolepsy.

Discussion

Recent animal studies have shown that the HCRT system not only contributes to arousal maintenance,^{3,7} but is also implicated in the regulation of motivated behaviors and in rewarding effects of addictive drugs.^{18,20,22,46} Because NC patients have low or undetectable levels of HCRT, we propose that narcolepsy with cataplexy may provide a valid model of the pathology to test the role of HCRT in the human reward system. The present fMRI study is the first, to our knowledge, to assess reward-related brain responses in NC patients.

Abnormal Reward-Related Brain Responses in NC

A major result of our study is the lack of VTA activation during the presentation of high-incentive cues in NC patients. This observation corroborates our initial hypothesis, based on the animal literature, that HCRT deficiency in NC patients would affect activity in ventral midbrain/VTA regions.^{13,14} In addition, functional connectivity analyses in the control group confirmed increased coupling of activity between the VTA and the NAcc during the presentation of highly motivating cues, thus substantiating enhanced VTA-NAcc interplay during large-incentive conditions. We also found that brain responses to reward experience (ie, during successful trials) in NC patients were affected in regions receiving dense HCRT projections and/or those modulated by dopaminergic inputs from the VTA, such as the NAcc and prefrontal cortex.⁴⁴ Taken together, these results suggest that the HCRT system, which is deficient in narcolepsy with cataplexy,^{5,6,31} may contribute to the regulation of brain reward functions, either via direct HCRT projections or via a modulation of dopaminergic VTA projections.^{16,20–22} It has been proposed that the HCRT system might regulate reward processing by increasing dopamine output via the potentiation of glutamatergic/opioid synaptic transmission in the VTA, which would enhance reinforcing effects of rewards.^{47–49} Accordingly, a dysfunction across reward-related regions, as revealed by our fMRI study during game playing, may limit the reinforcing effects of reward in HCRT-deficient NC patients, and could explain why psychostimulant abuse is extremely rare these patients.^{8,24–26}

Atypical Ventral-Dorsal Striatum Dissociation in NC

During successful trials, NC patients showed enhanced activity in the dorsal striatum (putamen), which is involved in stimulus-action reward associations and which mediates affective properties of outcomes in rewarding conditions.^{36,50–52} Activity increase in this region is consistent with NC patients being particularly responsive to emotionally positive contexts^{9,11} and echoes the recent observation by Chabas et al,⁵³ who reported a hyperperfusion in this same region during a cataplectic episode. Increased dorsal striatum activity contrasts with the lack of response in ventral striatum (in particular the NAcc) during winning in NC patients. However, fMRI signal in the NAcc and the vmPFC during high-incentive cues correlated with the duration of narcolepsy disease in the patients (Fig 5), suggesting that some adaptive mechanisms in patients more experienced with the disease could restore neural activity in these regions (in the absence of associated VTA activation). This result is consistent with

TABLE 3: Outcome Period: Brain Response to Actual Gains on Positively Cued Trials (Gain > No Gain)

Regions	L/R	MNI Coordinates			BA	Z Scores
		x	y	z		
Patients and controls (conjunction)						
ACC, ventromedial PFC	R	6	36	12	24	3.68
	L	-9	51	9	10/32	3.3
Controls masked by patients						
Lateral PFC	R	51	33	24	45	3.93
Middle occipital cortex	L	-30	-75	33	19	3.9
Ventral striatum (NAcc)	R	9	12	-15	25	3.57
Patients masked by controls						
Amygdala/SLEA	L	-24	-3	-12	34	4.09
Putamen (dorsal)	R	21	9	9	48	3.95
Amygdala	L	-30	-12	-27	36	3.91
Precuneus, posterior cingulate	L	-9	-54	21	23/30	3.87
Hippocampus	L	-24	-21	-15	30	3.84
	R	27	-21	-15	20	3.59
Cerebellum	R	21	-45	-51	—	3.76
Thalamus	R	15	-9	3	—	3.72
Lateral PFC	L	-39	39	3	45/47	3.68
	R	48	39	9	45	3.35
Amygdala	R	15	-3	-18	28	3.59
Putamen (ventral)	R	21	12	-9	48	3.47

Stereotactic coordinates correspond to the standard MNI brain. Reported regions survived a threshold level of $p < 0.001$ uncorrected. Mask threshold for group comparisons was set at a conservative level of $p < 0.05$. L = left; R = right; MNI = Montreal Neurological Institute; BA = Brodmann area; ACC = anterior cingulate cortex; PFC = prefrontal cortex; NAcc = nucleus accumbens; SLEA = sublenticular extended amygdala.

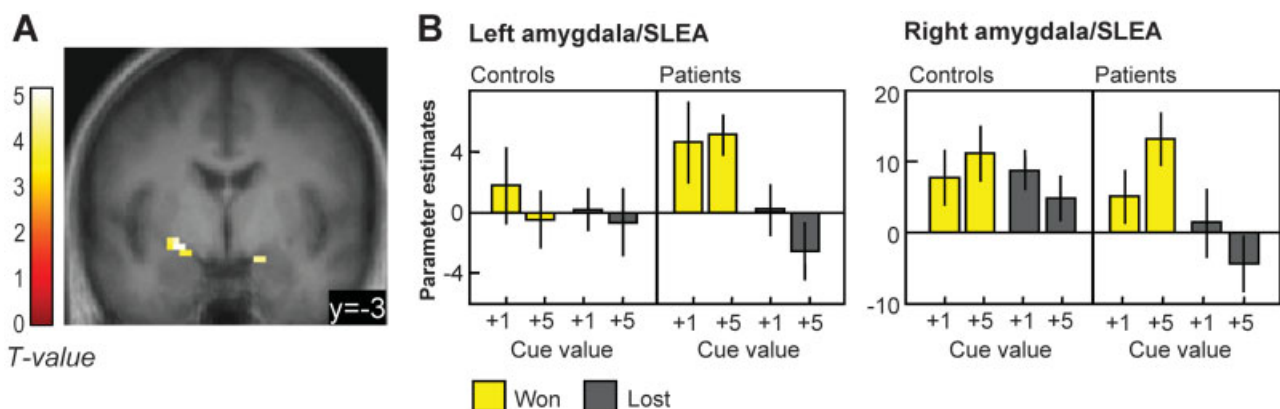


FIGURE 4: Brain responses to successful positively cued trials. (A) Increased bilateral amygdala/sublenticular extended amygdala (SLEA) response for actual gains in narcolepsy-cataplexy (NC) patients compared with controls. (B) Parameter estimates show a selective increase of functional magnetic resonance imaging signal in left and right amygdala/SLEA in response to successful positively cued trials compared to failed positively cued trials in the patients but not in the controls. Statistical maps are overlaid on mean-normalized structural scan; threshold $p < 0.001$. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

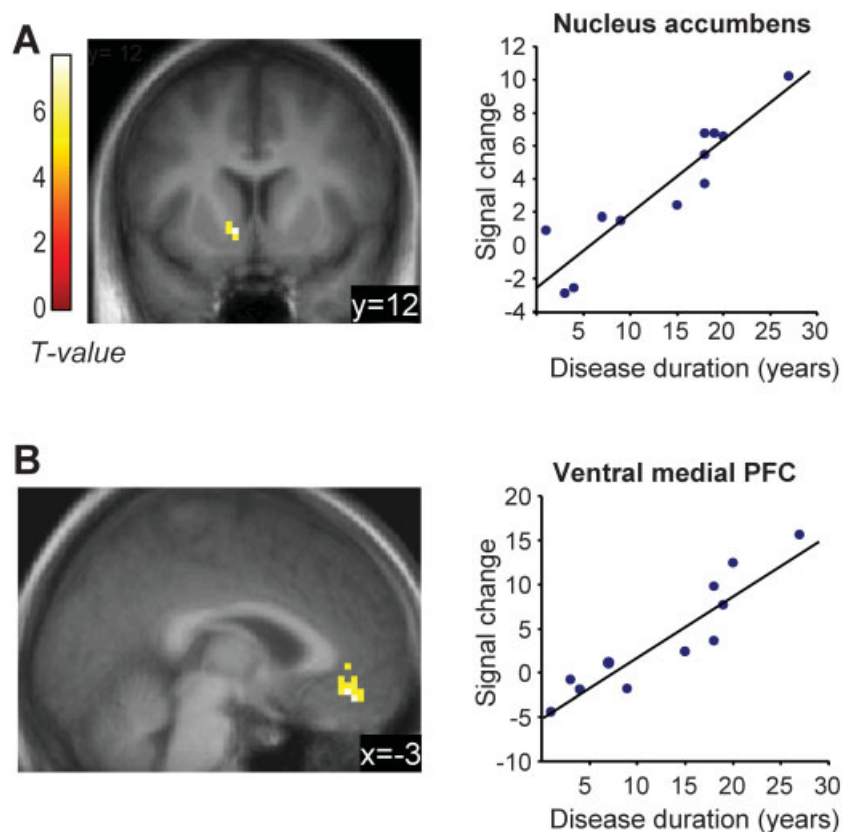


FIGURE 5: Linear regression between brain responses to high motivational cues and disease duration in narcolepsy-cataplexy (NC) patients. Positive correlation between the duration of the disease in NC patients and functional magnetic resonance imaging response to large (vs small) cues in (A) the nucleus accumbens ($r^2 = 0.85$, $p < 0.001$) and (B) the ventromedial prefrontal cortex (vmPFC) ($r^2 = 0.82$, $p < 0.001$). Plots show signal change during large versus small cues as a function of disease duration in the left nucleus accumbens (A) and vmPFC (B). Statistical maps are overlaid on mean-normalized T1 structural scans; threshold $p < 0.001$. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

the patients' reports of increased control over cataplexy attacks with time.⁵⁴ Thus, HCRT depletion may lead to an atypical ventral-dorsal striatum dissociation during reward processing, which might also reflect putative compensatory mechanisms in NC patients.

Because withdrawal from amphetamine-like drugs may modulate striatal activity,^{55,56} we would like to stress that only 6 out of the 12 NC patients withdrew from a chronic treatment (modafinil, $n=3$; modafinil and fluoxetine, $n=1$; ephedrine, $n=1$; clomipramine, $n=1$; Supplementary Table). Note that modafinil has been found to modulate many neurotransmitter systems, including glutamate, gamma aminobutyric acid, histamine, dopamine, and hypocretin activity.⁵⁷ Abnormalities in striatal activity found at the group level during reward processing in the patient population are therefore unlikely to be due to withdrawal from amphetamine-like drugs.

Putative Role for the HCRT System in Affective Responses to Reward

In the present study, we used a task in which subjects could win or lose points specifically because NC patients report

that they may experience cataplexy when confronted with highly positive or motivating situations, such as winning games. When comparing winning to losing on positively cued trials, NC patients showed increased activity in the dorsal striatum (see previous section), as well as in bilateral amygdala. Enhanced amygdala activity in the patients during positive emotional signals replicates our previous fMRI findings using humorous pictures as positive emotional stimuli.¹¹ We also recently reported that NC patients failed to exhibit amygdala-dependent startle potentiation during the presentation of unpleasant stimuli.⁵⁸ These observations document abnormal emotional processing for both positive and aversive signals in NC. The HCRT system may modulate amygdala-mediated emotional responses either via direct HCRT projections to the amygdala,^{4,59} or via projections to the VTA,¹⁶ which can in turn increase prefrontal dopamine efflux and attenuate amygdala response.^{44,45} In this context, increased vmPFC activation during high-motivating cues in the patients with a longer disease history could reflect a more efficient control over affective responses to emotional signals.^{42,60}

Conclusions

By using a stringent and unbiased whole-brain fMRI approach and a game-like task, our study offers a detailed picture of distributed brain regions involved during distinct stages of reward processing in both healthy controls and unmedicated NC patients. Our fMRI data on NC patients provide a first indication, to our knowledge, that HCRT activity may influence brain reward function in humans, and significantly extend existing results from animal cellular neurophysiology. Our findings may also have important clinical implications, because they suggest that the HCRT system could become a target for future treatments of addiction, which might benefit from the development of new pharmacological tools that modulate HCRT receptor activity in humans.⁶¹

This work was supported by grants from the Swiss National Science Foundation (#3200B0-104100 and #320030-118272 to C.B., E.W., R.K., S.S.; #310000-114008 to S.S.). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

We thank the patients and the healthy volunteers for participating in this study. We also thank Conny Schmidt for optimization of imaging sequences.

References

- de Lecea L, Kilduff TS, Peyron C, et al. The hypocretins: hypothalamus-specific peptides with neuroexcitatory activity. *Proc Natl Acad Sci U S A* 1998;95:322-327.
- Sakurai T, Amemiya A, Ishii M, et al. Orexins and orexin receptors: a family of hypothalamic neuropeptides and G protein-coupled receptors that regulate feeding behavior. *Cell* 1998;92:573-585.
- Saper CB, Scammell TE, Lu J. Hypothalamic regulation of sleep and circadian rhythms. *Nature* 2005;437:1257-1263.
- Peyron C, Tighe DK, van den Pol AN, et al. Neurons containing hypocretin (orexin) project to multiple neuronal systems. *J Neurosci* 1998;18:9996-10015.
- Baumann CR, Bassetti CL. Hypocretins (orexins) and sleep-wake disorders. *Lancet Neurol* 2005;4:673-682.
- Mignot E, Lammers GJ, Ripley B, et al. The role of cerebrospinal fluid hypocretin measurement in the diagnosis of narcolepsy and other hypersomnias. *Arch Neurol* 2002;59:1553-1562.
- Adamantidis AR, Zhang F, Aravanis AM, et al. Neural substrates of awakening probed with optogenetic control of hypocretin neurons. *Nature* 2007;450:420-424.
- Bassetti C, Aldrich MS. Narcolepsy. *Neurol Clin* 1996;14:545-571.
- Sturzenegger C, Bassetti CL. The clinical spectrum of narcolepsy with cataplexy: a reappraisal. *J Sleep Res* 2004;13:395-406.
- Anic-Labat S, Guilleminault C, Kraemer HC, et al. Validation of a cataplexy questionnaire in 983 sleep-disorders patients. *Sleep* 1999;22:77-87.
- Schwartz S, Ponz A, Poryazova R, et al. Abnormal activity in hypothalamus and amygdala during humour processing in human narcolepsy with cataplexy. *Brain* 2008;131:514-522.
- Reiss AL, Hoelt F, Tenforde AS, et al. Anomalous hypothalamic responses to humor in cataplexy. *PLoS One* 2008;3:e2225.
- Schultz W. Multiple reward signals in the brain. *Nat Rev Neurosci* 2000;1:199-207.
- O'Doherty JP, Buchanan TW, Seymour B, Dolan RJ. Predictive neural coding of reward preference involves dissociable responses in human ventral midbrain and ventral striatum. *Neuron* 2006;49:157-166.
- Pessiglione M, Seymour B, Flandin G, et al. Dopamine-dependent prediction errors underpin reward-seeking behaviour in humans. *Nature* 2006;442:1042-1045.
- Fadel J, Deutch AY. Anatomical substrates of orexin-dopamine interactions: lateral hypothalamic projections to the ventral tegmental area. *Neuroscience* 2002;111:379-387.
- Marcus JN, Aschkenasi CJ, Lee CE, et al. Differential expression of orexin receptors 1 and 2 in the rat brain. *J Comp Neurol* 2001;435:6-25.
- Narita M, Nagumo Y, Hashimoto S, et al. Direct involvement of orexinergic systems in the activation of the mesolimbic dopamine pathway and related behaviors induced by morphine. *J Neurosci* 2006;26:398-405.
- Korotkova TM, Sergeeva OA, Eriksson KS, et al. Excitation of ventral tegmental area dopaminergic and nondopaminergic neurons by orexins/hypocretins. *J Neurosci* 2003;23:7-11.
- Harris GC, Wimmer M, Aston-Jones G. A role for lateral hypothalamic orexin neurons in reward seeking. *Nature* 2005;437:556-559.
- Boutrel B, Kenny PJ, Specio SE, et al. Role for hypocretin in mediating stress-induced reinstatement of cocaine-seeking behavior. *Proc Natl Acad Sci U S A* 2005;102:19168-19173.
- Borgland SL, Taha SA, Sarti F, et al. Orexin A in the VTA is critical for the induction of synaptic plasticity and behavioral sensitization to cocaine. *Neuron* 2006;49:589-601.
- Georgescu D, Zachariou V, Barrot M, et al. Involvement of the lateral hypothalamic peptide orexin in morphine dependence and withdrawal. *J Neurosci* 2003;23:3106-3111.
- Nishino S, Mignot E. Pharmacological aspects of human and canine narcolepsy. *Prog Neurobiol* 1997;52:27-78.
- Guilleminault C, Carskadon M, Dement WC. On the treatment of rapid eye movement narcolepsy. *Arch Neurol* 1974;30:90-93.
- Parkes JD, Baraitser M, Marsden CD, Asselman P. Natural history, symptoms and treatment of the narcoleptic syndrome. *Acta Neurol Scand* 1975;52:337-353.
- Bassetti C. The spectrum of narcolepsy. In: Bassetti C, Billiard M, Mignot E, eds. *Narcolepsy and Hypersomnia*. New York, NY: Informa Healthcare; 2007:97-108.
- Knutson B, Adams CM, Fong GW, Hommer D. Anticipation of increasing monetary reward selectively recruits nucleus accumbens. *J Neurosci* 2001;21:RC159.
- Knutson B, Westdorp A, Kaiser E, Hommer D. fMRI visualization of brain activity during a monetary incentive delay task. *Neuroimage* 2000;12:20-27.
- Knutson B, Taylor J, Kaufman M, et al. Distributed neural representation of expected value. *J Neurosci* 2005;25:4806-4812.
- Baumann CR, Khatami R, Werth E, Bassetti CL. Hypocretin (orexin) deficiency predicts severe objective excessive daytime sleepiness in narcolepsy with cataplexy. *J Neurol Neurosurg Psychiatry* 2006;77:402-404.
- Penny W, Holmes A, Friston KJ. Random effects analysis. In: Frackowiak RS, Friston KJ, Frith CD, et al, eds. *Human Brain Function*. 2nd ed. Oxford, UK: Academic Press; 2003:635-654.

33. Friston KJ, Penny WD, Glaser DE. Conjunction revisited. *Neuroimage* 2005;25:661–667.
34. Friston KJ, Buechel C, Fink GR, et al. Psychophysiological and modulatory interactions in neuroimaging. *Neuroimage* 1997;6: 218–229.
35. Schultz W, Apicella P, Scarnati E, Ljungberg T. Neuronal activity in monkey ventral striatum related to the expectation of reward. *J Neurosci* 1992;12:4595–4610.
36. Haruno M, Kawato M. Different neural correlates of reward expectation and reward expectation error in the putamen and caudate nucleus during stimulus-action-reward association learning. *J Neurophysiol* 2006;95:948–959.
37. Vuilleumier P. How brains beware: neural mechanisms of emotional attention. *Trends Cogn Sci* 2005;9:585–594.
38. Critchley HD, Melmed RN, Featherstone E, et al. Volitional control of autonomic arousal: a functional magnetic resonance study. *Neuroimage* 2002;16:909–919.
39. D'Ardenne K, McClure SM, Nystrom LE, Cohen JD. BOLD responses reflecting dopaminergic signals in the human ventral tegmental area. *Science* 2008;319:1264–1267.
40. Knutson B, Cooper JC. Functional magnetic resonance imaging of reward prediction. *Curr Opin Neurol* 2005;18:411–417.
41. Wager TD, Davidson ML, Hughes BL, et al. Prefrontal-subcortical pathways mediating successful emotion regulation. *Neuron* 2008; 59:1037–1050.
42. Knutson B, Fong GW, Bennett SM, et al. A region of mesial prefrontal cortex tracks monetarily rewarding outcomes: characterization with rapid event-related fMRI. *Neuroimage* 2003;18:263–272.
43. Critchley HD, Mathias CJ, Dolan RJ. Neural activity in the human brain relating to uncertainty and arousal during anticipation. *Neuron* 2001;29:537–545.
44. Vittoz NM, Berridge CW. Hypocretin/orexin selectively increases dopamine efflux within the prefrontal cortex: involvement of the ventral tegmental area. *Neuropsychopharmacology* 2006;31: 384–395.
45. Phelps EA, Delgado MR, Nearing KI, LeDoux JE. Extinction learning in humans: role of the amygdala and vmPFC. *Neuron* 2004;43:897–905.
46. Boutrel B, de Lecea L. Addiction and arousal: the hypocretin connection. *Physiol Behav* 2008;93:947–951.
47. Ungless MA, Whistler JL, Malenka RC, Bonci A. Single cocaine exposure in vivo induces long-term potentiation in dopamine neurons. *Nature* 2001;411:583–587.
48. Borgland SL, Malenka RC, Bonci A. Acute and chronic cocaine-induced potentiation of synaptic strength in the ventral tegmental area: electrophysiological and behavioral correlates in individual rats. *J Neurosci* 2004;24:7482–7490.
49. Zheng H, Patterson LM, Berthoud HR. Orexin signaling in the ventral tegmental area is required for high-fat appetite induced by opioid stimulation of the nucleus accumbens. *J Neurosci* 2007;27:11075–11082.
50. Delgado MR. Reward-related responses in the human striatum. *Ann N Y Acad Sci* 2007;1104:70–88.
51. Schonberg T, Daw ND, Joel D, O'Doherty JP. Reinforcement learning signals in the human striatum distinguish learners from nonlearners during reward-based decision making. *J Neurosci* 2007;27:12860–12867.
52. O'Doherty J, Dayan P, Schultz J, et al. Dissociable roles of ventral and dorsal striatum in instrumental conditioning. *Science* 2004;304:452–454.
53. Chabas D, Habert MO, Maksud P, et al. Functional imaging of cataplexy during status cataplecticus. *Sleep* 2007;30:153–156.
54. Passouant P, Billard M. The evolution of narcolepsy with age. In: Guilleminault C, Dement WC, Passouant P, eds. *Narcolepsy*. New York: Spectrum Publications, Inc, 1976:179–196.
55. Tong J, Ross BM, Schmunk GA, et al. Decreased striatal dopamine D1 receptor-stimulated adenylyl cyclase activity in human methamphetamine users. *Am J Psychiatry* 2003;160:896–903.
56. London ED, Simon SL, Berman SM, et al. Mood disturbances and regional cerebral metabolic abnormalities in recently abstinent methamphetamine abusers. *Arch Gen Psychiatry* 2004;61: 73–84.
57. Ballon JS, Feifel D. A systematic review of modafinil: potential clinical uses and mechanisms of action. *J Clin Psychiatry* 2006; 67:554–566.
58. Khatami R, Birkmann S, Bassetti CL. Amygdala dysfunction in narcolepsy-cataplexy. *J Sleep Res* 2007;16:226–229.
59. Date Y, Ueta Y, Yamashita H, et al. Orexins, orexigenic hypothalamic peptides, interact with autonomic, neuroendocrine and neuroregulatory systems. *Proc Natl Acad Sci U S A* 1999;96: 748–753.
60. Critchley HD, Elliott R, Mathias CJ, Dolan RJ. Neural activity relating to generation and representation of galvanic skin conductance responses: a functional magnetic resonance imaging study. *J Neurosci* 2000;20:3033–3040.
61. Brisbare-Roch C, Dingemans J, Koberstein R, et al. Promotion of sleep by targeting the orexin system in rats, dogs and humans. *Nat Med* 2007;13:150–155.