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Sleep Enhances Plasticity in the Developing Visual Cortex

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Summary

During a critical period of brain development, occluding the vision of one eye causes a rapid remodeling of the visual cortex and its inputs. Sleep has been linked to other processes thought to depend on synaptic remodeling, but a role for sleep in this form of cortical plasticity has not been demonstrated. We found that sleep enhanced the effects of a preceding period of monocular deprivation on visual cortical responses, but wakefulness in complete darkness did not do so. The enhancement of plasticity by sleep was at least as great as that produced by an equal amount of additional deprivation. These findings demonstrate that sleep and sleep loss modify experience-dependent cortical plasticity in vivo. They suggest that sleep in early life may play a crucial role in brain development.

Introduction

Scientists have long suspected that neuronal connections are remodeled during sleep. Studies in humans (Ekstrand et al., 1977; Rottenberg, 1992; Karni et al., 1994; Plihal and Born, 1999; Ficca et al., 2000; Gais et al., 2000; Stickgold et al., 2000a, 2000b) and animals (Fishbein and Gutwein, 1977; McGrath and Cohen, 1978; Block and Hennevin, 1979; Smith, 1985, 1995) have shown that sleep and sleep loss influence learning and memory—two processes thought to depend on synaptic plasticity. Recent findings in rodents (Pavlides and Winson, 1989; Wilson and McNaughton, 1994; Skaggs and McNaughton, 1996; Qin et al., 1997; Kudrimoti et al., 1999; Poe et al., 2000; Louie and Wilson, 2001), birds (Dave et al., 1998; Dave and Margoliash, 2000), and humans (Macquet et al., 2000) suggest that neuronal activity initiated during wake is reactivated and possibly consolidated during subsequent sleep. Sleep and sleep loss also modify the expression of several genes (Cirelli and Tononi, 1999; Ribeiro et al., 1999) and gene products (Neuner-Jehle et al., 1995, 1996) that may be important for synaptic plasticity. Certain forms of long-term potentiation can also be elicited in sleep (Hennevin et al., 1993; Bramham et al., 1994), suggesting that synaptic connections are strengthened during sleep. In addition, sleep amounts are very high and undergo dramatic modifications during developmental periods of heightened synaptogenesis and synaptic plasticity (Roffwarg et al., 1966; Jouvet-Mounier et al., 1970; Davis et al., 1999). These findings together provide strong, suggestive evidence that synaptic circuits are modified during sleep. There are, however, no studies that provide direct evidence that sleep and sleep loss modify experience-dependent changes in synaptic circuitry.

We examined the effects of sleep and sleep loss on a well-described form of synaptic remodeling in vivo. During a critical period of development in the cat, occluding vision in one eye initiates a rapid remodeling of synaptic weights and morphology in thalamocortical circuits. (Hubel and Wiesel, 1970; Olson and Freeman, 1980; Antonini and Stryker, 1993; Crair et al., 1997). Short periods of monocular deprivation (MD) reduce the magnitude and specificity of cortical neuronal responses to the deprived eye (Freeman, 1979; Olson and Freeman, 1980; Crair et al., 1997). These changes in synaptic weights are followed by anatomical rearrangements in thalamic afferents that reduce cortical territories innervated by the deprived eye (Antonini and Stryker, 1993). The amount and stimulus properties of waking visual experience required for this form of synaptic remodeling, known as ocular dominance plasticity, have been well described (Freeman, 1979; Singer, 1979; Freeman and Olson, 1982). Whether or not sleep contributes to this form of synaptic remodeling is unknown.

Results

We used the following experimental design to test the role of sleep in ocular dominance plasticity. Cats at the peak of the critical period were divided into four experimental groups. In all four groups, we first collected a baseline 6 hr sample of sleep and wake using standard polysomnographic recordings of the electroencephalogram (EEG) and electromyogram (EMG). Following this baseline period, cats in all four groups had one eye sutured shut and were kept awake in a lighted environment for an additional 6 hr. This monocular deprivation period provided a standard stimulus for the induction of plasticity in all groups. The four groups differed in their experience thereafter, as illustrated in Figure 1. Cats in the first group (MD6; $n = 5$) were immediately anesthetized for physiological measurement of ocular dominance in primary visual cortex, using optical imaging of intrinsic cortical signals and extracellular unit recording. Cats in a second group (MDS; $n = 6$) were allowed to sleep for an additional 6 hr in complete darkness before making optical and unit recordings. Cats in the third group (MDSD; $n = 7$) were treated identically to those in the MDS group except that they were kept awake rather than allowed to sleep during the 6 hr in complete darkness before the recordings. Cats in the fourth group (MD12; $n = 4$) were also kept awake for an additional 6 hr but remained in a lighted environment, effectively giving them an additional 6 hr of monocular deprivation before the recordings.

These experiments allowed us to determine, first,

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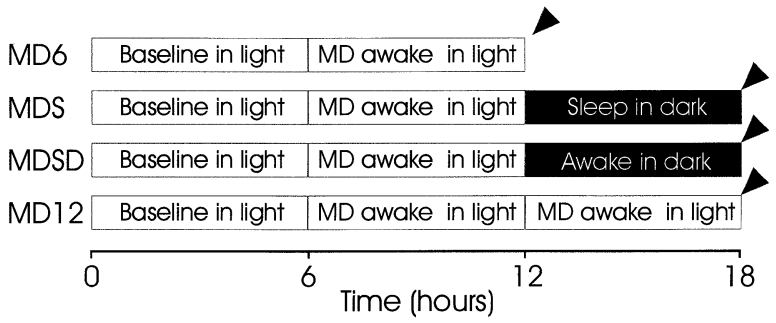

Figure 1. Sleep and Lighting Conditions for the Four Experimental Groups

Group names at left are defined in the text. During the baseline period, animals were kept in the light and were allowed to sleep ad lib. The baseline period was immediately followed by a 6 hr period of monocular deprivation in the light, during which time the animals were kept awake. The four groups differed in their experience thereafter as indicated. Vigilance states were monitored continuously until the time of physiological recordings of ocular dominance. The time at which animals were anesthetized for physiological recordings is indicated by the arrowhead for each group.

whether the effects of MD were enhanced by sleep immediately thereafter (MD6 compared to MDS); second, whether the enhancement of plasticity observed in group MDS was due to sleep or merely to the passage of time following the inducing stimulus (MDS compared to MDSD); and third, whether the procedure used to sleep deprive the cats itself directly impeded ocular dominance plasticity (MD12 compared to MDSD).

Vigilance States

Figure 2 illustrates the measurement of vigilance states, including wakefulness, rapid eye movement (REM) sleep, and non-REM sleep, in the different experimental groups.

Baseline Period

To ensure that all groups of cats were at the same stage of development and were similar in their baseline sleep, we assessed sleep patterns during a 6 hr period at the beginning of each experiment. While sleep amounts change rapidly during development, Figure 3 confirms, as expected for cats of similar ages, that there were no differences between vigilance state amounts among the four groups. Non-REM sleep ($df = 3$, $F = 2.04$, $p > 0.15$), REM sleep ($df = 3$, $F = 1.90$, $p > 0.17$), and wake ($df = 3$, $F = 1.33$, $p > 0.30$) amounts were similar in all groups of cats. We also analyzed non-REM/REM sleep EEG slow-wave activity (SWA; 0.5–4.0 Hz) power ratios, since these EEG power ratios are sensitive to maturational effects on neonatal EEGs (Frank and Heller, 1997). There were no significant differences in non-REM/REM sleep EEG SWA ratios ($df = 3$, $F = 2.04$, $p > 0.15$) among the four groups (MD6 mean $\pm$ SEM, 5.7 $\pm$ 0.5; MDS, 5.7 $\pm$ 0.6; MDSD, 6.3 $\pm$ 0.3; MD12, 8.2 $\pm$ 1.4). These results indicate that cats in all four groups were at similar stages of sleep–wake development at the start of each experiment.

MD Period

To ensure that all groups of cats received equivalent amounts of monocular visual experience, we kept all cats awake in a lighted environment for 6 hr after surgically closing the lids of one eye. Mild forms of sleep deprivation (see Experimental Procedures) were necessary because undisturbed cats will spend most of their time asleep (Figures 2 and 3). To verify that all groups of cats were similarly alert during the MD period, we measured the amounts of sleep, wake, and EEG SWA during the MD period. Measuring EEG SWA during the MD period provided an additional estimate of alertness in our animals because increases in EEG SWA are associated with sleepiness and a corresponding loss of attentiveness, which may be important for ocular dominance plasticity (Singer, 1982). There were no significant differences in the amount of wakefulness among the groups ($df = 3$, $F = 0.73$, $p > 0.548$), and there were only small differences in EEG SWA levels during the MD period. Although there was slightly more EEG SWA in the MDS, MD6, and MD12 groups compared to the MDSD group ($df = 3$, $F = 7.13$, $p < 0.002$; Tukey $p < 0.05$), the amounts of EEG SWA in all groups were a small percent of those found in baseline non-REM sleep (MD6 mean $\pm$ SEM, 21.1% $\pm$ 3.1%; MDS, 24.6% $\pm$ 3.9%; MDSD, 14.6% $\pm$ 1.3%; MD12, 23.7% $\pm$ 4.5%). These results indicate that the cats in the four groups were all alert during the MD period.

Ad Lib Sleep Period

Since sleep amounts and architecture are often modified following sleep deprivation (Dijk and Edgar, 1999) or tasks requiring synaptic remodeling (Smith and Lapp, 1986; Ambrosini et al., 1992; Guiditta et al., 1995), we examined changes in sleep amounts, architecture (latency to REM sleep, duration of individual REM and non-REM sleep episodes), and depth (sleep continuity and non-REM EEG SWA) that occurred in the ad lib sleep period in group MDS relative to the baseline period. Sleep was only slightly altered relative to baseline during the ad lib sleep period (Table 1). The most consistent modifications were increases in REM sleep amounts and the duration of REM sleep episodes in the ad lib period relative to baseline values. There were no significant modifications of non-REM sleep other than a small increase in non-REM sleep amounts in the 3rd and 4th hr of the ad lib sleep period.

Sleep Deprivation

Both REM and non-REM sleep were greatly reduced in the MDSD and MD12 groups compared to their respective baseline values (Figure 3). REM sleep was reduced 93%–98% (from baseline) in MDSD and MD12 cats. The reductions in non-REM sleep were more variable (40%–80% reduction).

The Effects of Sleep and Sleep Loss on Ocular Dominance Plasticity

Microelectrode Recordings

We made microelectrode recordings in primary visual cortex to measure neuronal responses to stimulation of one or the other eye. Unit responses were ranked on

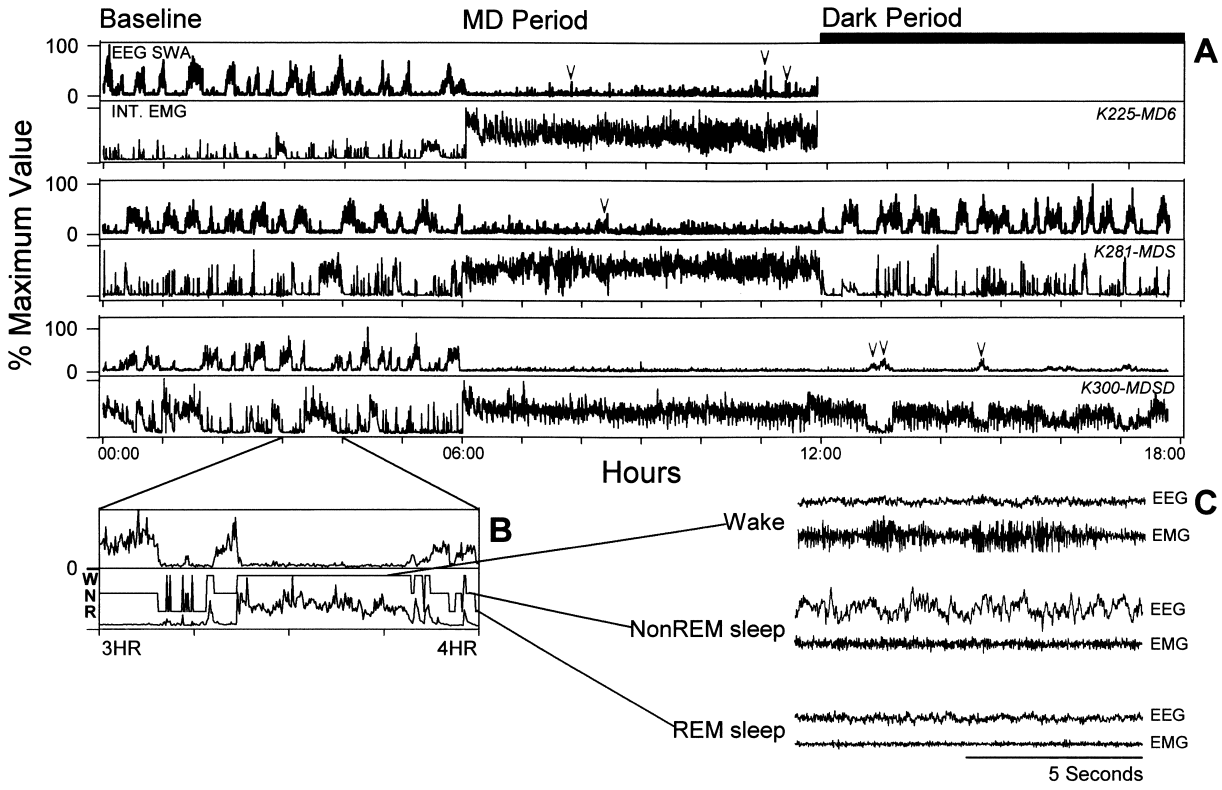

Figure 2. Representative Sleep and Wake Recordings from Three Experimental Groups

(A) Time course of EEG and EMG signals from three representative experiments. EEG SWA and integrated EMG signals expressed as a percentage of their respective maximum values are shown for each experiment. (Top) MD6 (cats assayed for changes in ocular dominance immediately after 6 hr of MD). (Middle) MDS (6 hr of MD followed by 6 hr of ad lib sleep in the dark). (Bottom) MDSD (6 hr of MD followed by 6 hr of SD in the dark). Arrows mark regions of small SWA increases during wake.

(B) Expansion of 1 hr of the MDSD experiment, with a hypnogram superimposed on the EMG trace in the bottom panel. Wake (W) is characterized by intermediate to high EMG signals coupled with low EEG SWA values. Non-REM sleep (N) is characterized by low to intermediate EMG signals coupled to intermediate to high EEG SWA values. Very low EMG signals and very low EEG SWA values characterize REM sleep (R).

(C) Polygraphic EEG and EMG signals from selected 10 s epochs of wake, non-REM, and REM sleep in expanded hypnogram.

the conventional seven-point ocular dominance scale based on the relative responses to stimulation through the two eyes (Hubel and Wiesel, 1970), and ocular dominance histograms were calculated for each hemisphere. The overall ocular dominance of each hemisphere was summarized in a scalar measure called the contralateral bias index (CBI), for which a value of 1 indicates complete dominance of the eye contralateral to the hemisphere under study, 0 indicates complete dominance of the ipsilateral eye, and 0.5 indicates equal representation of the two eyes. The degree to which neurons were activated by both eyes as opposed to one or the other eye is summarized in the monocularity index (MI), for which a value of 1 would indicate that no neurons are activated by both eyes, and 0 would indicate that all neurons are activated equally by both eyes. The overall effect of MD is summarized by a shift index (SI), ranging from 0 in normal animals to ± 1, indicating complete shifts toward one eye or the other; this index is calculated as the difference between the CBIs in the two hemispheres (Reiter et al., 1986; Issa et al., 1999; Trachtenberg et al., 2000).

The effects of MD on the visual cortex were strongly influenced by subsequent sleep. As indicated by the

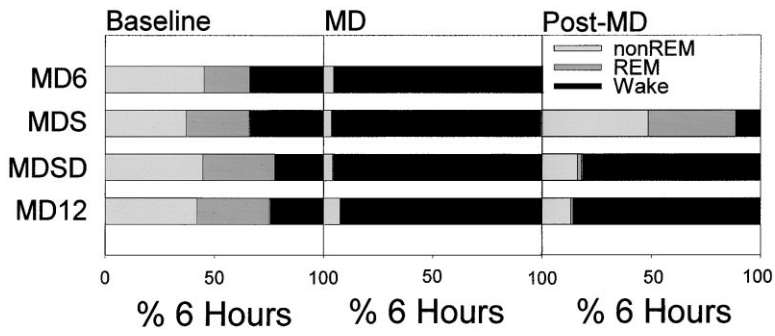

Figure 3. Mean Amounts of REM Sleep, Non-REM Sleep, and Wake in Groups MD6, MDS, MDSD, and MD12 Expressed as a Percentage of Each 6 Hr Period

Sleep was increased in the MDS group and reduced in the MDSD and MD12 groups during the post-MD period relative to baseline (Student's t test, $p < 0.05$). There were no significant differences between groups in sleep–wake amounts during baseline or monocular deprivation (MD) periods (ANOVA, $p > 0.05$).

Table 1. Sleep Amounts and Sleep Architecture in the Ad Lib Sleep Period after a 6 Hr MD

	Baseline	Post-MD Hours 1–2	Post-MD Hours 3–4	Post-MD Hours 5–6	Post-MD Overall	Pearson Overall
Non-REM % TRT	45.6 ± 2.4	45.3 ± 2.3	52.8 ± 1.6[a]	47.9 ± 0.9	48.6 ± 1.2	NS
REM % TRT	33.6 ± 2.8	43.3 ± 2.5[a]	36.1 ± 1.6	40.2 ± 1.3	39.9 ± 1.2[a]	unit SI: $r = -0.77$, $p < 0.05$
Non-REM SWA %	—	110.7 ± 7.6	119.0 ± 8.7	110.0 ± 8.2	113.2 ± 4.6	
Non-REM bout	3.7 ± 0.2	3.7 ± 0.3	3.9 ± 0.4	3.6 ± 0.3	3.7 ± 0.2	
REM bout	3.7 ± 0.2	5.5 ± 0.6[a]	4.0 ± 0.5	4.0 ± 0.4	4.5 ± 0.3[a]	NS
Sleep continuity	7.41 ± 0.8	9.5 ± 2.0	9.6 ± 2.3	8.1 ± 0.6	9.0 ± 1.0	
REM latency	7.3 ± 1.1	5.6 ± 0.6	6.0 ± 0.7	6.2 ± 0.9	5.9 ± 0.4	

Mean (± SEM) results from the entire baseline and ad lib sleep (in 2 hr bins and overall) periods for the group MDS are shown. The amounts of REM and non-REM sleep are expressed as a percentage of total recording time (TRT = 6 hr). The mean (± SEM) duration of REM and non-REM bouts, latency to REM sleep, and episodes of uninterrupted sleep (sleep continuity) are given in minutes (see Experimental Procedures for details). Changes in non-REM EEG SWA are expressed as a percentage of baseline values (Student's t test, $p < 0.05$). NS indicates nonsignificant correlations between measures of ocular dominance and sleep variables that differed from baseline in the MDS group.
[a] Indicates significant difference from 6 hr mean baseline values (Student's t test, $p < 0.05$).

ocular dominance histograms (Figure 4A) and the CBI and SI scores (Table 2; Figure 5B), 6 hr of MD (MD6 group) caused a small but consistent loss of response to the deprived eye. As indicated by increases in unit MI scores (Figure 5A), 6 hr of MD also reduced binocular responses in visual cortex. These changes in ocular dominance were much greater in cats allowed to sleep ad lib in the dark for 6 hr after the MD period (Figures 4B and 5; Table 2). The loss of response to the deprived eye was nearly doubled as measured by the SI (MDS versus MD6, Mann–Whitney, $p < 0.02$), and binocularity was further reduced as measured by the MI (MDS versus MD6, Mann–Whitney, $p < 0.004$). The increase produced by sleep in the effects of the initial 6 hr of MD were similar to and slightly greater than the increase produced by an additional 6 hr of MD (MD12 group; Figures 4D and 5; Table 2). Sleep deprivation in the dark, however, completely prevented the enhancement of cortical plasticity observed in the sleeping cats. Unit recordings in group MDSD showed that neuronal responses in both hemispheres were only slightly shifted away from the deprived eye (Figure 4C). The unit SI (MDS versus MDSD, Mann–Whitney U, $p < 0.01$), MI (MDS versus MDSD, Mann–Whitney U, $p < 0.05$), and CBI values showed that cortical plasticity was significantly reduced in the MDSD group compared to the MDS group (Figures 5A and 5B; Table 2). Indeed, the mean ocular dominance shift in the animals deprived of sleep in the dark (MDSD) was even less that that immediately following MD (MD6 group), although this difference did not reach statistical significance.

The differences among experimental groups were not due to differences in the cortical laminae sampled in the different experiments (Trachtenberg et al., 2000). While there were not enough data to compare the effects of MD in individual laminae across groups, electrode path reconstructions showed that a similar proportion of units was sampled from layer IV in all groups (MD6: granular, 24.5%, extragranular, 75.5%; MDS: granular, 22.6%, extragranular, 74.4%; MDSD: granular, 35%, extragranular, 65%; MD12: granular, 29%, extragranular, 71%).

These differences were also not due to our assessment of ocular dominance with knowledge of the experimental condition. In the three experimental groups that were studied after a period of sleep or sleep deprivation, ocular dominance was reanalyzed blind to the experimental condition from computer records of the visual responses. CBIs obtained from this blind analysis were

Table 2. Contralateral Bias Indices from Unit and Optical Recordings from Primary Visual Cortex in Cats from the Four Groups

	Unit		Optical	
	IPSI	CONTRA	IPSI	CONTRA
MD6	*n = 172*	*n = 177*	*n = 5*	*n =5*
	0.67 ± 0.03	0.42 ± 0.05	0.64 ± 0.05	0.48 ± 0.02
MDS	*n = 238*	*n = 201*	*n = 6*	*n = 6*
	0.75 ± 0.02	0.32 ± 0.02	0.76 ± 0.04	0.44 ± 0.05
	a: ($p < 0.003$, $p < 0.04$)	*a: ($p < 0.05$, $p < 0.006$)*	*b: ($p < 0.008$)*	*b: ($p < 0.04$)*
MDSD	*n = 211*	*n = 204*	*n = 6*	*n = 6*
	0.61 ± 0.06	0.44 ± 0.03	0.59 ± 0.02	0.50 ± 0.03
MD12	*n = 161*	*n = 140*	*n =4*	*n =4*
	0.78 ± 0.03	0.38 ± 0.08	0.69 ± 0.03	0.38 ± 0.02
	a: ($p < 0.013$, $p < 0.035$)		*b: ($p < 0.03$)*	*a: ($p < 0.02$, $p < 0.02$)*

Data represent mean (± SEM) contralateral bias index (CBI) values in cats provided 6 hr of MD and then immediately tested (MD6), 6 hr of MD + 6 hr of sleep (MDS), 6 hr of MD + 6 hr of sleep deprivation (MDSD), and 12 hr of MD (MD12). Values in italics represent number of unit recordings made in the hemisphere ipsilateral (IPSI) and contralateral (CONTRA) to the deprived eye and number of hemispheres optically imaged. "*a*" indicates significant differences between a specific group and both the MD6 and MDSD groups; "*b*" indicates significant difference between a specific group and only the MDSD group. Values in parentheses are the Mann–Whitney U significance probabilities for each comparison.

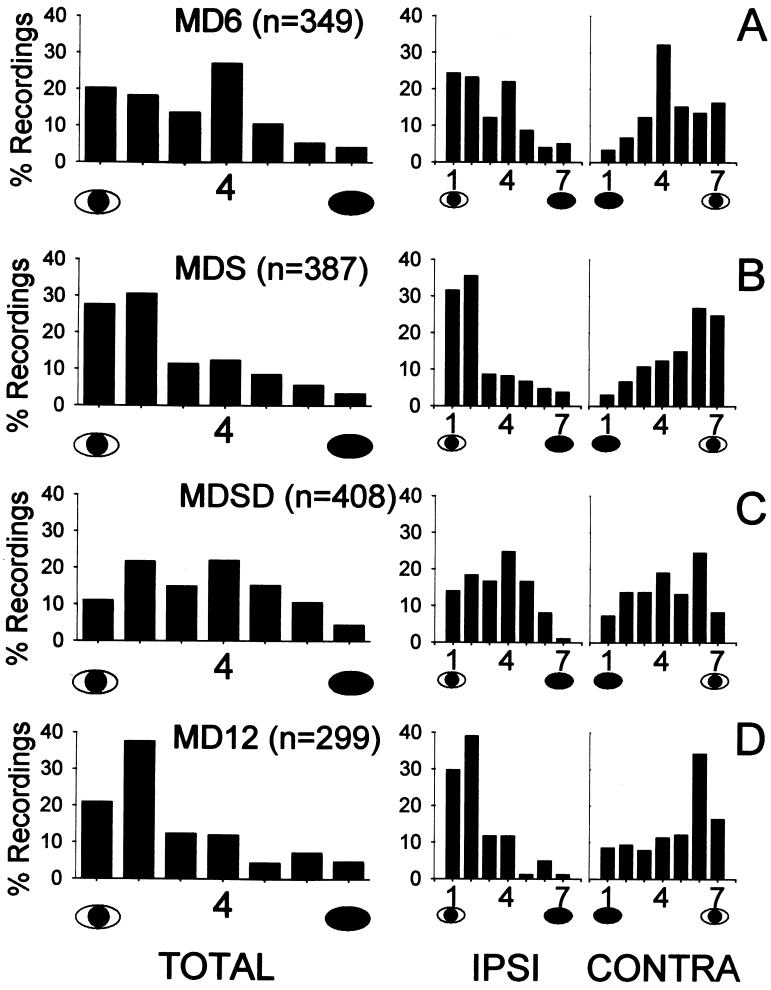

Figure 4. Effects of Sleep and Sleep Deprivation on Ocular Dominance Plasticity

Ocular dominance histograms compiled from pooled unit recordings in both hemispheres (TOTAL) and in hemispheres ipsilateral (IPSI) and contralateral (CONTRA) to the deprived eye for group MD6 (A), group MDS (B), group MDSD (C), and group MD12 (D). Total histograms are ranked such that 1 represents cells driven exclusively by the nondeprived eye, 7 represents cells driven exclusively by the deprived eye, and 4 represents cells that are driven equally by the two eyes. Histograms for each hemisphere are ranked according to the traditional seven-point scale of Hubel and Wiesel (1970).

highly correlated with those from the initial, witting analysis (Pearson $r = 0.94$, $p < 0.01$) and produced similar mean SIs in the different groups (MDS: 0.39 blind, 0.32 witting; MDSD: 0.12 blind, 0.11 witting; MD12: 0.32 blind, 0.30 witting).

Optical Imaging of Intrinsic Cortical Signals

Optical imaging of intrinsic cortical signals provides an additional quantitative and objective measure of the responses to the two eyes over a large region of visual cortex (Crair et al., 1997; Issa et al., 1999). Ocular dominance at each pixel was computed from a comparison of responses to the two eyes at the optimal stimulus orientation, analogous to ocular dominance as studied with unit recordings (Issa et al., 1999). Optical CBIs and SIs were computed in a similar manner as unit CBIs and SIs (Issa et al., 1999).

Optical imaging data confirmed the findings obtained with microelectrode recordings in primary visual cortex. Figure 6 shows that stimuli presented to the nondeprived eye produced bright, well-oriented polar maps, indicating strong and selective responses, in animals from all experimental groups. These resembled optical maps obtained from normally sighted cats. In contrast, responses to the deprived eye were weaker and less selective, as indicated by the dimmer polar maps, revealing an effect of MD in all groups. Figure 7 shows maps of the ratio of the responses to the two eyes and ocular dominance histograms compiled from microelectrode recordings for each hemisphere in a representative animal from each group. The effect of MD is also evident in the reduced size of cortical territories dominated by the deprived eye.

As with the unit recordings, ocular dominance plasticity assessed from optical recordings was greatest in the MDS group and least in the MDSD group. Optical CBIs (Table 2) and SIs (Figure 5C) thus showed an enhancement of ocular dominance plasticity in cats allowed to sleep (MDS group) compared either to cats tested immediately after the MD period (MD6) or to cats kept awake in the dark for an additional 6 hr (MDSD). The magnitude of enhancement of plasticity by sleep was similar to that of an additional 6 hr of MD (MD12 group).

Contributions of REM and Non-REM Sleep to Ocular Dominance Plasticity

We performed a series of post-hoc analyses to determine what aspects of sleep were most important for the enhancement of plasticity observed in the sleeping cats.

Changes in ocular dominance in the MDSD and MDS groups were positively correlated with the amount of non-REM sleep in the dark period. Figure 8 shows a scatter plot of the effects of MD as a function of the amount of non-REM sleep. Both unit (Pearson: $r = 0.95$,

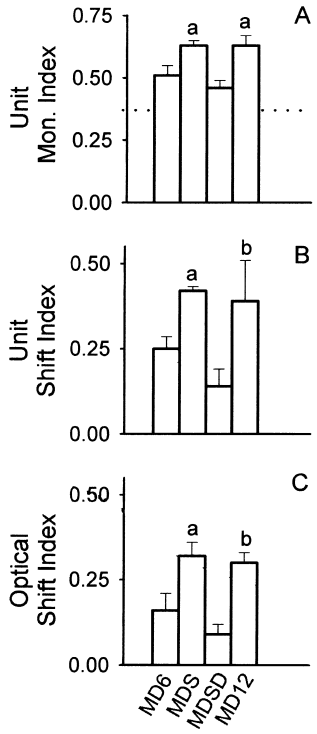

Figure 5. Scalar Measurements of Ocular Dominance in Groups MD6, MDS, MDSD, and MD12

The mean (± SEM) monocularity index (A) and shift index (B) compiled from microelectrode data and the shift index compiled from optical data (C) are shown for all four groups. "a" indicates significant differences between a specific group and both the MD6 and MDSD groups; "b" indicates significant difference between a specific group and the MDSD group only (Mann–Whitney U, $p < 0.05$). Dashed horizontal line in (A) indicates value of monocularity index in normal animals. The shift index for a normal animal is 0.

$p < 0.01$) and optical (Pearson: $r = 0.89$, $p < 0.05$) SIs were strongly dependent on the amount of non-REM sleep. Non-REM sleep amounts were also positively correlated with MI scores ($r = 0.81$, $p < 0.05$). A very large portion of the variance in ocular dominance scores is thus accounted for by the variance in the amount of non-REM sleep (79% of optical SI scores, 90% of unit SI scores, and 66% of unit MI scores). These results suggest that the change in ocular dominance triggered by unequal visual experience from the two eyes during wakefulness may be specifically enhanced during subsequent non-REM sleep.

To investigate the contribution of REM sleep to ocular dominance plasticity, we first attempted a similar correlation analysis between REM sleep and ocular dominance plasticity, but this proved unfruitful because REM sleep amounts, on average, tended to cluster at 0% in the MDSD group or near 40%–45% of total recording time in the MDS group, with no data points between these extremes. We then performed a series of experiments identical to those in the MDSD group, except that we attempted to selectively deprive cats of REM sleep during the dark period. While REM sleep deprivation has been successfully performed in older cats (Oksenberg et al., 1996), in five attempts in additional 4- to 4.5-week-old cats, we were unable to eliminate REM sleep without also altering non-REM sleep. Instead, in agreement with previous findings in animals with polyphasic sleep cycles (Benington et al., 1994), REM sleep–deprived cats attempted to enter REM sleep with increasing frequency, resulting in increasingly frequent interruptions of non-REM sleep. This produced a progressive reduction in non-REM sleep amounts and/or continuity (measured by the average change in non-REM sleep episode duration) and non-REM sleep depth (as measured by EEG SWA; data not shown). For this reason, we were not able to obtain data on selective REM sleep deprivation.

In the MDS group, there was a significant increase in REM sleep during the ad lib sleep period compared to baseline. In this group, REM sleep amounts were negatively correlated with ocular dominance plasticity as measured by unit SI scores (Pearson $r = -0.77$, $p < 0.05$). This negative correlation (and the corresponding r^2 value: 0.59) suggests a possible inhibitory effect of REM sleep on ocular dominance plasticity. Such an effect, however, is quite small, since unit SI scores varied over a narrow range among cats in the MDS group (range: 0.38–0.47), and correlations between REM sleep and either optical SI scores or unit MI scores were not significant.

Discussion

We examined the effects of sleep and sleep loss on the cortical plasticity evoked by MD during the cat critical period for visual development. We found that the effects of MD on visual cortical responses were greatly enhanced by a 6 hr sleep period in the dark, as measured by both microelectrode recording and optical imaging. The enhancement of cortical plasticity appeared to occur specifically as a consequence of non-REM sleep. Sleep deprivation during this time completely blocked the enhancement of cortical plasticity observed after sleep. These results provide evidence that sleep and sleep loss modify experience-dependent cortical plasticity in vivo. These findings and their implications for our understanding of cortical plasticity and the function of sleep in developing animals are discussed below.

Sleep, Consolidation, and Ocular Dominance Plasticity

Consolidation, defined as a time-dependent process that converts labile memory traces into more permanent and/or enhanced forms, has been reported in many studies of memory and learning (McGaugh, 2000) and may occur during sleep (Smith, 1985, 1995; Karni et al., 1994; Buzsaki, 1998; Gais et al., 2000; Stickgold et al., 2000a, 2000b). Consolidation periods in ocular dominance plasticity, in contrast, are less consistently reported, and a role for sleep in these processes has not previously been demonstrated. For example, several studies have shown that interleaving monocular experience with periods of darkness (Pettigrew and Garey, 1974; Peck and Blakemore, 1975; Olson and Freeman, 1980; Ramachandran and Ary, 1982; Malach et al., 1984; Yinon and Goshen, 1984) or opportunities to sleep (Mioche and Singer, 1989) stabilizes and/or enhances the effects of MD. Anesthetics also prevent shifts in ocular dominance if administered shortly after monocular experience, suggesting that changes in ocular domi-

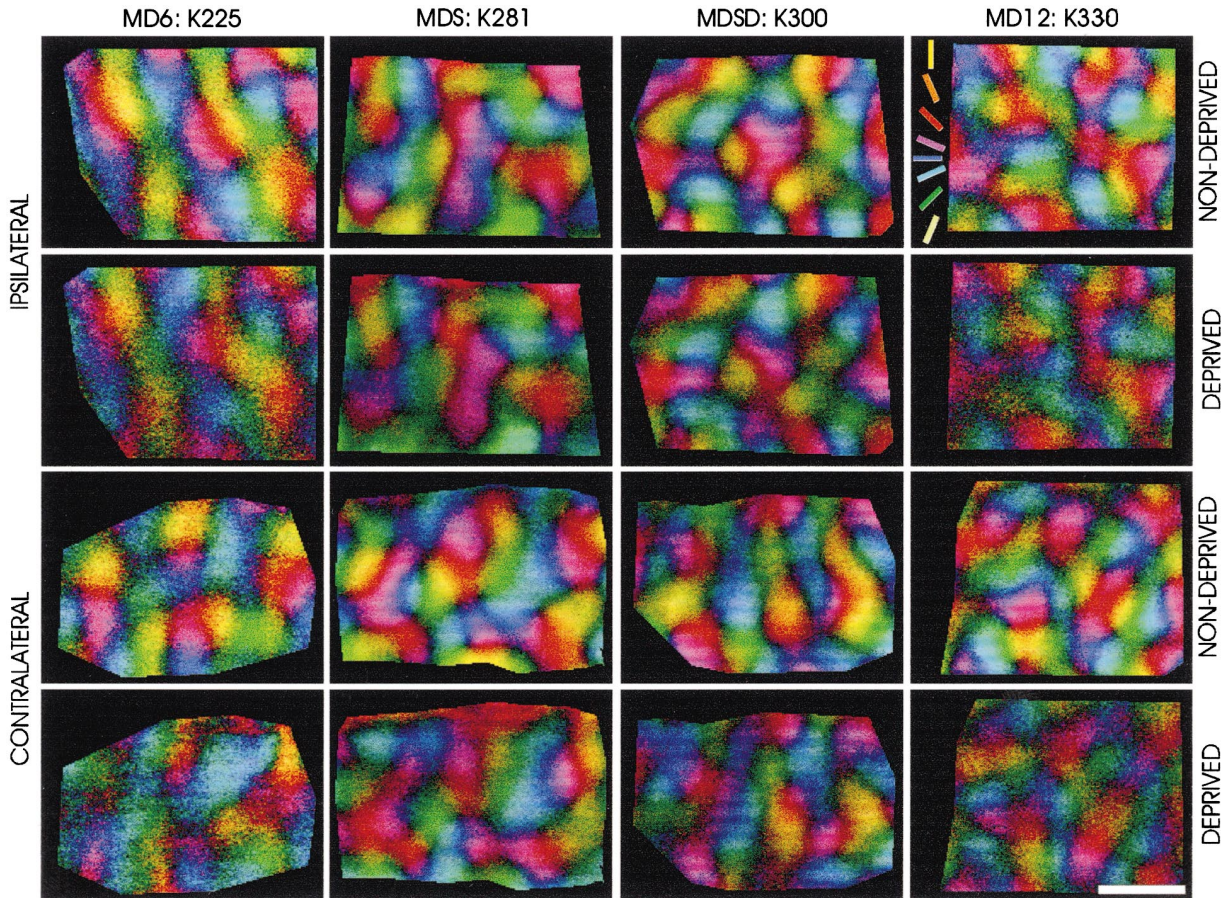

Figure 6. Polar Optical Maps from a Representative Animal from Each Experimental Group

Images in the top two rows are of the hemisphere ipsilateral to the deprived eye; images in the bottom two rows are of the contralateral hemisphere. The top image of each pair was produced in response to stimulation through the nondeprived eye, and the bottom image was produced in response to the deprived eye. The hue of a pixel corresponds to the stimulus orientation that best activated it. The brightness of the pixel represents how well tuned to orientation and how strongly activated the pixel was by a stimulus at its preferred orientation. The two images of each pair were scaled identically, but the different pairs of images were scaled independently. Therefore, image intensity can be compared between eyes but cannot be compared across hemispheres or animals. Cortical activity driven through the nondeprived eye is strong and well oriented, similar to the activity pattern observed in normal animals. Responses to the deprived eye are weaker and less well tuned compared to responses to the nondeprived eye. White bar represents 1 mm.

nance are initially labile and consolidate over time (Rauschecker and Hahn, 1987). In addition, the recovery of neuronal selectivity produced by visual experience in dark-reared animals is enhanced following opportunities to sleep (Imbert and Buisseret, 1975). Freeman and Olson, on the other hand, reported that placing cats in the dark after a period of MD reduced the effect of MD on cortical responses (Freeman and Olson, 1979, 1982). In none of these studies, however, were vigilance states determined, nor was the amount of visual experience quantitatively assessed. Consequently, while these studies provide some evidence for consolidation periods in ocular dominance plasticity, they tell us little about the role of sleep in this process. Our findings demonstrate that there is a consolidation period in ocular dominance plasticity, and they suggest that this consolidation process occurs during sleep.

Possible Mechanisms

This study has established an effect of sleep and sleep deprivation on a form of developmental cortical plasticity. What mechanisms might account for this effect? One trivial explanation might be that plasticity is lost in the MDSD group as a result of the stress produced by sleep deprivation. A second trivial explanation is that sleep deprivation does not reduce plasticity; instead, the smaller MD effect in the MDSD group might reflect continued plasticity driven by similar levels of activity in the two eyes during the dark period, which restores binocular responses. Most interestingly, the results may reflect an enhancement of plasticity by sleep-specific processes that reinforce the unequal visual experience of the prior waking period.

Sleep Deprivation and Stress

Because of the well-known relationship between the endocrine system and the sleep–wake cycle, even the small amounts of sleep deprivation used in this study might increase the release of stress hormones, such as corticosterone and norepinephrine (Tobler et al., 1983; Cauter and Spiegel, 1999). Differences in stress hormone release, however, are unlikely to contribute to our results. For example, while corticosterone can inhibit

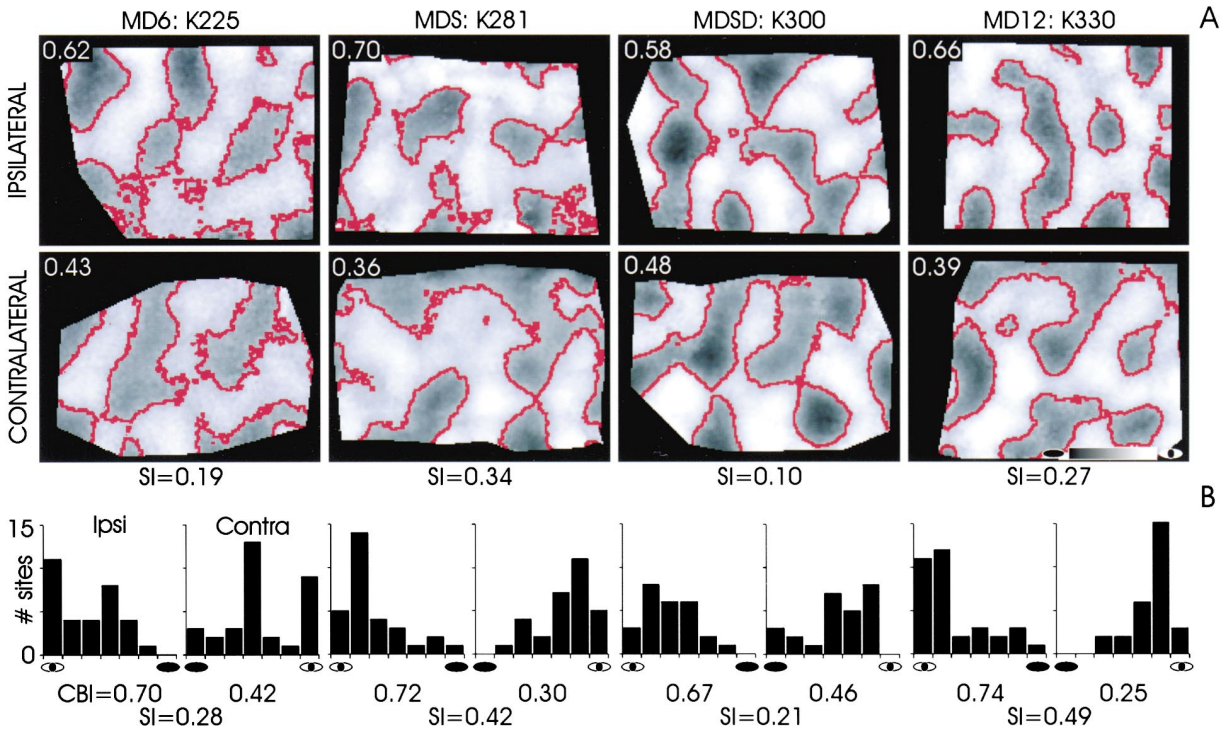

Figure 7. Ocular Dominance Data from a Representative Animal from Each Experimental Group

(A) Ocular dominance maps of primary visual cortex in cats monocularly deprived with or without a subsequent sleep period. Ocular dominance ratio maps constructed using the optimal stimulus orientation at each pixel are shown for both hemispheres of one animal from each of the four experimental conditions (MD6, MDS, MDSD, and MD12). The top row shows ocular dominance ratio maps of the hemisphere ipsilateral to the deprived eye; the bottom row shows ocular dominance ratio maps of the hemisphere contralateral to the deprived eye. In these maps, dark pixels are dominated by the deprived eye, while bright pixels are dominated by the nondeprived (open) eye. Red contour lines show the boundaries between regions of deprived and nondeprived eye preference. In the MDS condition, the deprived eye regions in both hemispheres are smaller than in the MDSD group. The CBI calculated for each hemisphere is shown in the top left corner of each frame; the SI for each animal is shown below the images. The grayscale bar represents 1 mm.

(B) Ocular dominance histograms from randomly placed microelectrode penetrations in the two hemispheres of the individual animals illustrated above. Compare with pooled data from all animals shown in Figure 4.

ocular dominance plasticity, prolonged (weeks) administration of very high concentrations of corticosterone are required to reduce plasticity to levels comparable to what we observe in sleep-deprived cats (Daw et al., 1990). Given the relatively weak effects of corticosterone on ocular dominance plasticity, it is unlikely that the transient release of corticosterone induced by 6–12 hr of SD had a major effect on our results. Norepinephrine concentrations are also linked to the sleep–wake cycle (Jones, 1994) and are elevated during natural wake as well as during sleep deprivation (Irwin et al., 1999). However, as is true for corticosterone, elevated norepinephrine levels are unlikely to be a factor in our result. Norepinephrine has permissive effects on ocular dominance plasticity (Kasamatsu et al., 1979), and transient increases in stress-induced neuromodulators, such as norepinephrine, facilitate rather than inhibit processes dependent on synaptic remodeling (McEwen and Sapolsky, 1995; De Kloet et al., 1999). Furthermore, if 6–12 hr of SD inhibited ocular dominance plasticity via stress hormone release, then ocular dominance plasticity should have also been inhibited in the MD12 group. This was not the case. Ocular dominance plasticity was increased, not inhibited, in the MD12 group. Thus, stress is unlikely to be a mediator of the effect that we have found.

Does Dark Experience Restore Binocularity?

An alternative explanation for the reversal of the effects of MD in the sleep-deprived cats is that equal levels of spontaneous activity in the two eyes during darkness reverse preceding monocular experience. This seems an unlikely explanation for the following reasons. First, although some studies have shown that periods of darkness can reverse the effects of MD (Freeman and Olson, 1979, 1982), this effect is not always found (Pettigrew and Garey, 1974; Peck and Blakemore, 1975; Olson and Freeman, 1980; Ramachandran and Ary, 1982; Malach et al., 1984; Yinon and Goshen, 1984), and it is not clear if the animals were awake or asleep during the dark period (see above discussion). Second, even if this effect can account for the small and statistically insignificant reduction in ocular dominance shift found in the MDSD group compared to the MD6 group, it cannot account for the enhanced ocular dominance shift found in the sleeping cats (MDS), who were also in the dark and thus would also be expected to have spontaneous activity in the two eyes. For these reasons, then, "dark experience" cannot by itself account for our results, and the vigilance state must instead play a central role.

Sleep and Cortical Plasticity

An attractive explanation for our results is that sleep-specific neuronal processes consolidate changes in oc-

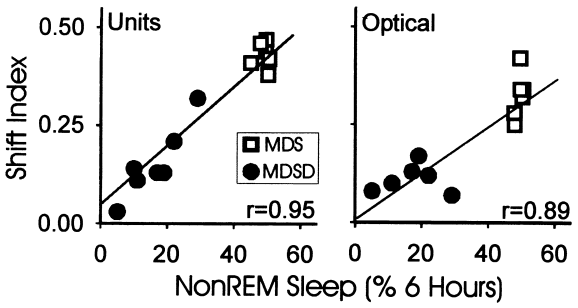

Figure 8. Linear Regression and Correlation Coefficients in Groups MDS and MDSD between Non-REM Sleep Amounts in the Dark Period and Changes in Ocular Dominance

Open squares represent the MDS group; filled circles represent the MDSD group. (Left) Correlation between unit shift index and non-REM sleep. (Right) Correlation between optical shift index and non-REM sleep (one data point from the unit experiments was excluded from the optical results in the MDSD group since optical maps in this animal could not be used). The solid line represents the linear regression between non-REM sleep amounts and ocular dominance scores for the entire distribution.

ular dominance evoked by MD. According to this hypothesis, the large shift in ocular dominance found in the sleeping group (MDS) is due to sleep-dependent consolidation of the labile changes in ocular dominance that occurred during the previous MD period. In the sleep-deprived group (MDSD), this sleep-dependent consolidation is disrupted, and changes in ocular dominance do not progress.

This consolidation hypothesis is supported by several findings that suggest both REM and non-REM sleep contribute to synaptic remodeling. REM sleep, for example, appears to be necessary for the consolidation of certain memory tasks in animals (Fishbein and Gutwein, 1977; McGrath and Cohen, 1978; Block and Hennevin, 1979; Smith, 1985, 1995) and humans (Karni et al., 1994). Non-REM sleep has also been linked with learning in animals (Ambrosini et al., 1992; Guiditta et al., 1995) and, more recently, the consolidation of procedural memory for visual discrimination tasks in humans (Gais et al., 2000; Stickgold et al., 2000a, 2000b; but see Karni et al., 1994). These latter findings are particularly interesting because procedural memory in humans, like ocular dominance plasticity in cats, is primarily mediated by changes in subcortical and cortical circuitry (Pascual-Leone et al., 1994). Sleep might therefore be expected to influence ocular dominance plasticity.

Our finding of a positive correlation between changes in ocular dominance and non-REM sleep time indicates that experience-dependent changes in cortical circuits are specifically strengthened during non-REM sleep. Two possible mechanisms may account for these results. The first is that patterned neuronal activity from the two eyes initiated during MD is replayed during non-REM sleep, and this reiteration strengthens synaptic changes evoked by MD. This hypothesis is supported by the findings that complex patterns of waking neuronal activity are reactivated during non-REM sleep (Wilson and McNaughton, 1994; Skaggs and McNaughton, 1996; Qin et al., 1997; Kudrimoti et al., 1999) and that brain activity during non-REM sleep promotes plasticity in corticothalamic networks (Steriade, 1999). Alternatively or in addition, it is possible that neurohumeral substances, such as neurotrophins, necessary for cortical plasticity are released during non-REM sleep. The transcription of several genes known to be important for synaptic plasticity increases during wake (Cirelli and Tononi, 1999), and the translation of some of these genes may in turn occur during sleep (Neuner-Jehle et al., 1995, 1996). Whether substances important for ocular dominance plasticity are specifically released during non-REM sleep is unknown, but non-REM sleep is associated with increased release of somatic growth factors (Cauter and Spiegel, 1999) and enhanced protein synthesis in the brain (Ramm and Smith, 1990; Nakanishi et al., 1997), which suggests that such substances might be released during this vigilance state.

Sleep and the Developing Brain

For many years, scientists have speculated that sleep might be important for the developing brain (Roffwarg et al., 1966; Marks et al., 1995; Mirmiran, 1995). While both REM and non-REM sleep could conceivably contribute to brain maturation, REM sleep has historically been viewed as a state that facilitates neuronal development (Roffwarg et al., 1966; Mirmiran, 1995; Davis et al., 1999). We now provide evidence that demonstrates a possible role for non-REM sleep in brain development.

The correlation between non-REM sleep time and cortical plasticity suggests that, during critical periods of development, experience-dependent changes in cortical circuitry are consolidated during non-REM sleep. This finding is supported by previous studies that have shown that prolonged REM sleep deprivation or the elimination of REM sleep pontine-geniculate-occipital waves in the cat does not block the anatomical effects of MD in the lateral geniculate nucleus (Oksenberg et al., 1996; Shaffery et al., 1999). Since non-REM sleep amounts were not reduced in these latter studies, these results are consistent with our observation that, while REM sleep may slightly inhibit experience-dependent changes in cortical circuitry, the consolidation of these changes requires non-REM sleep.

A role for non-REM sleep in developmental cortical plasticity is further suggested by maturational changes in non-REM sleep that coincide with periods of heightened cortical plasticity. In the cat, there is a steep decline in REM sleep and a sharp increase in non-REM sleep amounts near the beginning of the critical period for visual development (Jouvet-Mounier et al., 1970). In rats, the beginning of the critical period for visual development coincides with the development of non-REM sleep homeostasis. Prior to the 4th postnatal week, non-REM sleep EEG SWA does not intensify following sleep deprivation, indicating that the regulatory relationship between wake and non-REM sleep matures in parallel with periods of heightened cortical plasticity (Frank et al., 1998). Taken together, these findings suggest that non-REM sleep may also be important for the developing brain.

Conclusions

In conclusion, we find that sleep enhances cortical synaptic remodeling during the critical period for visual development in the cat. Despite great progress in our understanding of the regulation and neurobiology of sleep,

as well as the consequences of sleep loss on human performance, why the brain needs sleep remains a mystery (Jones, 1994; Steriade and Amzica, 1998; Carrier and Monk, 1999; Dijk and Edgar, 1999). Our findings now provide strong evidence that one function of sleep is to help consolidate the effects of waking experience on cortical circuitry. These findings raise a number of interesting questions about the role of sleep in cortical plasticity. What are the cellular or molecular mechanisms responsible for the effects of sleep on cortical plasticity, and to what extent are these mechanisms distinct from those governing cortical plasticity during wakefulness? What is the role of REM sleep in cortical plasticity? Does sleep have similar effects on cortical plasticity in the adult brain? Answers to these questions may provide new insights into synaptic remodeling and will reveal important clues about sleep function.

Experimental Procedures

Formation of Groups

Cats from the University of California, San Francisco (UCSF) breeding colony (two toms and four queens purchased from a commercial breeder) were assigned to the experimental groups as they became available. To control for potential litter effects, no more than two cats from a litter were used in any one group. We studied cats only near the peak of the critical period for ocular dominance plasticity, and the mean ages of all groups were similar (MD6 = 31.6 ± 0.9 (SEM) days; MDS = 30.8 ± 1.0 days; MDSD = 31.6 ± 1.7 days; and MD12 = 32.0 ± 0.9 days). Cats were housed with their queens in a 14:10 light–dark cycle (maintained at an ambient temperature of 22°C–25°C) until the start of an experiment.

EEG/EMG Surgeries and Sleep Recording

At postnatal days (P)24–26, cats were anesthetized and prepared for chronic implant surgery according to previously described methods (Reiter et al., 1986). The skull was exposed, and six EEG electrodes (00 stainless-steel screws soldered to teflon-insulated steel wires) were placed bilaterally in frontal and parietal bone. Three EMG electrodes (braided stainless-steel wire) were placed deep in nuchal tissues. All electrodes were soldered to a Micro-Tech electrical socket, which was in turn affixed to the skull with dental acrylic. The wound was sutured with 4-0 vicryl thread. The cats received analgesia immediately upon arousal from anesthesia and were returned to their queens once sternal. Cats received antibiotics (amoxicillin, twice daily) for at least 5 days, at which time they were removed for an experiment. On the day of the experiment, cats were attached to electrical recording cables, which were in turn connected to a counter-balanced, slip-ring commutator. Although adult cats are only weakly circadian (Tobler and Scherschlicht, 1990; Lancel et al., 1991) and P27–P34 cats are aperiodic (Lugazzi et al., 1979) with respect to sleep–wake cycles, we staggered the start times of the experiments to ensure that assays of ocular dominance in all experiments began at approximately the same time of day. The baseline sleep periods began at ~15:00 hr for the MDS, MDSD, and MD12 groups and at ~21:00 hr for the MD6 group. All extracellular and optical imaging physiology began between 09:00 and 10:00 hr the following day. The cats were housed in an illuminated, transparent acrylic, cylindrical chamber enclosed within a grounded Faraday cage (maintained at 22°C–25°C). Cats were fed KMR milk replacement ad lib every 6 hr. Frontal-parietal unihemispheric EEGs (filtered at 0.3 and 35.0 Hz) and nuchal EMGs (filtered at 3.0 and 75.0 Hz) were amplified on a Grass 7PCB polygraph, digitized (at 12 bits), and continuously collected (from baseline to the time of ocular dominance assays) on a personal computer in 10 s epochs. Fourier-transformed EEGs and integrated EMG signals were scored in real time as non-REM sleep (high levels of EEG 0.5–4.0 Hz power, low to intermediate EMG levels), REM sleep (low levels of EEG 0.5–4.0 Hz power, EMG minima), or wake (low levels of EEG 0.5–4.0 Hz power, intermediate to high EMG signals). EEG/EMG state criteria were adjusted to match polygraphic and behavioral signs of sleep before the start of each experiment by an experimenter trained in scoring neonatal sleep patterns (M. G. F.).

Post-Hoc Analyses of Sleep and Wake

We conducted a series of post-hoc analyses of the sleep–wake data at the end of each 12–18 hr sleep–wake data collection period. Digitized polygraphic EEG/EMG records were inspected epoch by epoch, and artifacts (usually less than 5% of the total record) were removed. In the baseline period, we calculated the amount of each vigilance state (as a percentage of total recording time) and non-REM/REM EEG SWA ratios. Since EEG SWA tends to increase in non-REM sleep and decrease in REM sleep during postnatal development (relative to faster EEG frequencies), this ratio provides an estimate of developmental changes in sleep EEGs (Frank and Heller, 1997; Davis et al., 1999). We made additional measurements of sleep and wake in the ad lib sleep period to determine if sleep was altered by 6 hr of MD. We measured REM sleep, non-REM sleep, and wake continuity by calculating the average duration of sleep and wake bouts (in minutes). A bout was defined as a sustained vigilance state of at least 20 s, not interrupted by the occurrence of any other vigilance state. We also measured mean latencies to REM sleep and total sleep continuity in each group. REM sleep latencies were defined as the number of non-REM sleep minutes elapsed before each REM sleep bout of at least 20 s in duration. Sleep continuity was measured by calculating the mean length (in minutes) of an episode of total sleep, not interrupted by a wake period of at least 20 s. Sleep and wake data were statistically evaluated using ANOVAs and protected (Tukey) t tests (SAS statistical software).

MD Procedure

Cats were anesthetized with 2.5% isoflurane in oxygen. The eyelid around the eye to be sutured was cleaned with disinfectant, and chloramphenicol ophthalmic ointment was placed in the eye. The eye was then sutured shut with 4.0 vicryl thread. We used only 6 hr of MD since this was the minimum amount of MD needed to induce measurable changes in ocular dominance, and, when combined with the sleep protocol (see below), it allowed us to keep the total length of sleep deprivation within 12 hr.

Sleep Deprivation Techniques Used in the MD and SD

We determined in a series of pilot studies the maximum amount of wakefulness that critical period cats could comfortably maintain. This amount was ~12 hr—an amount of sleep deprivation that does not perturb cortical responses to visual stimuli in the cat (Livingstone and Hubel, 1981). Wakefulness (during the MD for all groups and during the SD) was maintained by gently moving the cage floor and by playing tape recordings of "meowing" on computer detection of polygraphic signs of sleep onset. The sensitivity of the detection was set such that very small increases in EEG SWA coupled to reductions in the EMG signal (corresponding to sleep onset) triggered the motorized cage floor. These EEG and EMG parameters were periodically adjusted during the course of each experiment to ensure that sleep was prevented at the appropriate times.

Microelectrode Recordings

Cats were anesthetized with barbiturates, paralyzed, artificially ventilated, and prepared for extracellular recording of unit responses and optical imaging according to methods previously described (Crair et al., 1997). Recordings of responses to moving bar stimuli were made with sharp electrodes advanced in 100 μm steps in tangential medial bank penetrations of primary visual cortex. Electrolytic lesions were made at ~1 mm intervals. Two to three penetrations (3–4 mm in length) were made in each hemisphere in all cats in all groups. Pooled responses of the one to three units at each recording site were isolated with a window discriminator, and visually driven activity was assessed using a combination of responses to a light bar produced by a hand-held projector and a computer-based visual stimulation system that presented moving high-contrast bars at eight different orientations (or no stimulus for measurement of spontaneous response) in a pseudorandom sequence. Ocular dominance was categorized at each recording site on a seven-point scale, where 1 = cells driven entirely by the contralateral

eye, 7 = cells driven entirely by the ipsilateral eye, and 4 = cells that are entirely binocular (Hubel and Wiesel, 1970). Scalar measures of unit responses to the two eyes and the effects of MD were calculated using the contralateral bias index (the CBI for a normal cat is 0.55), the shift index (SI), and the monocularity index (MI for normal cats is 0.35) as described previously (Reiter et al., 1986; Issa et al., 1999; Trachtenberg et al., 2000). These scalar measures were statistically evaluated with Mann–Whitney U tests (SAS statistical software). A total of 25 to 40 microelectrode recordings were made in each hemisphere, giving the following totals. MD6: n = 5 cats, recording sites = 349; MDS: n = 6, sites = 387; MDSD: n = 7, sites = 408; and MD12: n = 4, sites = 299. To verify that there was no bias in our original ocular dominance assignments, we also digitally collected unit spike times for each recording (in the MDS, MDSD, and MD12 groups), which were then rescored for ocular dominance by an experimenter blind to the sleep condition of the animal. The results of this blinded analysis were then compared to the original nonblinded results to test for subjective biasing in ocular dominance score assignments.

Trachtenberg et al. (2000) recently showed that MD has more pronounced effects in the extragranular laminae of the cortex. To verify that we sampled equally from granular and extragranular laminae across groups, we reconstructed 30 microelectrode penetrations (MD6: five cats, six penetrations; MDS: five cats, six penetrations; MDSD: six cats, ten penetrations: and MD12: five cats, eight penetrations). Fixed brains were cut at 50 μm on a freezing microtome, mounted on glass slides, defatted, and stained with cresyl violet according to previously described methods (Trachtenberg et al., 2000). Cortical sections containing the electrode penetration were mounted on glass slides, inspected under high magnification, and the electrode path was reconstructed using camera lucida techniques. The number of recording sites obtained in granular (layer IV) and extragranular (layer II–III and V–VI) layers was then tabulated for each penetration.

Optical Imaging

Intrinsic signal optical responses were measured using the ORA 2000 system (Optical Imaging, Inc., Germantown, NY) as previously described (Issa et al., 1999). Reflectance of 610 nm light from the primary visual cortex during monocular presentation of moving high-contrast square wave gratings (0.2 cycles/deg moving at 2 cycles/s) was compared to reflectance during the presentation of a blank (gray) screen. Stimuli were presented to one or the other eye in pseudorandom order at eight different orientations. Stimulus and blank screens were presented 16 times in each stimulus session, and three to four sessions were run for each hemisphere. Images were analyzed using custom software written in IDL (Research Systems, Inc., Boulder, CO). The ocular dominance at each pixel was computed from a comparison of responses to the two eyes at the optimal stimulus orientation, analogous to ocular dominance as studied with unit recordings (Issa et al., 1999). Ocular dominance ratio maps were constructed by dividing each pixel's response to its preferred orientation shown to the nondeprived eye by the response to the same stimulus orientation presented to the other eye. Optical CBIs and SIs were computed in a similar manner as unit CBIs and SIs (Issa et al., 1999). These scalar measures were statistically evaluated with Mann–Whitney U tests (SAS statistical software).

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References

Ambrosini, M.V., Langella, M., Carnevale, U.A.G., and Guiditta, A. (1992). The sequential hypothesis of sleep function. III. The structure of postacquisition sleep in learning and nonlearning rats. Physiol. Behav. *51*, 217–226.

Antonini, A., and Stryker, M.P. (1993). Rapid remodeling of axonal arbors in the visual cortex. Science *260*, 1819–1821.

Benington, J.H., Woudenberg, M.C., and Heller, H.C. (1994). REM-sleep propensity accumulates during 2-h REM-sleep deprivation in the rest period in rats. Neurosci. Lett. *180*, 76–80.

Block, V., and Hennevin, E. (1979). Relationship between paradoxical sleep and memory processes. In Brain Mechanisms in Memory and Learning: From the Single Neuron to Man, M.A.B. Brazier, ed. (New York: Raven Press), pp. 120–135.

Bramham, C.R., Maho, C., and Laroche, S. (1994). Suppression of long-term potentiation induction during alert wakefulness but not during 'enhanced' REM sleep after avoidance learning. Neuroscience *59*, 501–509.

Buzsaki, G. (1998). Memory consolidation during sleep: a neurophysiological perspective. J. Sleep Res. *7*, 17–23.

Carrier, J., and Monk, T.H. (1999). Effects of sleep and circadian rhythms on performance. In Regulation of Sleep and Circadian Rhythms, Volume 133, P.C. Zee and F.W. Turek, eds. (New York: Marcel Dekker, Inc.), pp. 527–551.

Cauter, E.V., and Spiegel, K. (1999). Circadian and sleep control of hormonal secretions. In Regulation of Sleep and Circadian Rhythms, Volume 133, P.C. Zee and F.W. Turek, eds. (New York: Marcel Dekker, Inc.), pp. 397–425.

Cirelli, C., and Tononi, G. (1999). Differences in brain gene expression between sleep and waking as revealed by mRNA differential display and cDNA microarray technology. J. Sleep Res. Suppl. *1*, 44–52.

Crair, M.C., Ruthazer, E.S., Gillespie, D.C., and Stryker, M.P. (1997). Relationship between the ocular dominance and orientation maps in visual cortex of monocularly deprived cats. Neuron *19*, 307–318.

Dave, A.S., and Margoliash, D. (2000). Song replay during sleep and computational rules of sensorimotor vocal learning. Science *290*, 812–816.

Dave, A.S., Yu, A.C., and Margoliash, D. (1998). Behavioral state modulation of auditory activity in a vocal motor system. Science *282*, 2250–2254.

Davis, F.C., Frank, M.G., and Heller, H.C. (1999). Ontogeny of sleep and circadian rhythms. In Regulation of Sleep and Circadian Rhythms. Volume 133, P.C. Zee and F.W. Turek, eds. (New York: Marcel Dekker, Inc.), pp. 19–80.

Daw, N.W., Sato, H., Fox, K., Carmichael, T., and Gingerich, R. (1990). Cortisol reduces plasticity in the kitten visual cortex. J. Neurobiol. *22*, 158–168.

De Kloet, E.R., Oitzl, M.S., and Joels, M. (1999). Stress and cognition: are corticosteroids good or bad guys? TINS *22*, 422–426.

Dijk, D.-J., and Edgar, D.M. (1999). Circadian and homeostatic control of wakefulness and sleep. In Regulation of Sleep and Circadian Rhythms, Volume 133, P.C. Zee and F.W. Turek, eds. (New York: Marcel Dekker, Inc.) pp. 111–148.

Ekstrand, B.R., Barrett, T.R., West, J.N., and Meier, W.G. (1977). The effect of sleep on human long-term memory. In Neurobiology of Sleep and Memory, R.R. Drucker-Colin and J.L. McGaugh, eds. (New York: Academic Press), pp. 419–438.

Ficca, G., Lombardo, P., Rossi, L., and Salzarulo, P. (2000). Morning recall of verbal material depends on prior sleep organization. Behav. Brain Res. *112*, 159–163.

Fishbein, W., and Gutwein, B.M. (1977). Paradoxical sleep and memory storage processes. Behav. Biol. *19*, 425–464.

Frank, M.G., and Heller, H.C. (1997). Development of REM and slow wave sleep in the rat. Am. J. Physiol. *272*, R1792–R1799.

Frank, M.G., Morrissette, R., and Heller, H.C. (1998). Effects of sleep deprivation in neonatal rats. American J. Physiol. *275*, R148–R157.

Freeman, R.D. (1979). Effects of brief uniocular 'patching' on kitten visual cortex. Trans. Ophthal. Soc. U. K. *99*, 382–385.

Freeman, R.D., and Olson, C. (1982). Brief periods of monocular

deprivation in kittens: effects of delay prior to physiological study. J. Neurophysiol. *47*, 139–150.

Freeman, R.D., and Olson, C.R. (1979). Is there a 'consolidation' effect for monocular deprivation? Nature *282*, 404–406.

Gais, S., Plihal, W., Wagner, U., and Born, J. (2000). Early sleep triggers memory for early visual discrimination skills. Nat. Neurosci. *3*, 1335–1339.

Guiditta, A., Ambrosini, M.V., Montangnese, P., Mandile, P., Cotugno, M., Zucconi, G.G., and Vescia, S. (1995). The sequential hypothesis of the function of sleep. Behav. Brain Res. *69*, 157–166.

Hennevin, E., Mahao, C., Hars, B., and Dutrieux, G. (1993). Learning-induced plasticity in the medial geniculate nucleus is expressed during paradoxical sleep. Behav. Neurosci. *107*, 1018–1030.

Hubel, D.H., and Wiesel, T.N. (1970). The period of susceptibility to the physiological effects of unilateral eye closure in kittens. J. Physiol. *206*, 419–436.

Imbert, M., and Buisseret, P. (1975). Receptive field characteristics and plastic properties of visual cortical cells in kittens reared with or without visual experience. Exp. Brain Res. *22*, 25–36.

Irwin, M., Thompson, J., Miller, C., Gillin, J.C., and Ziegler, M. (1999). Effects of sleep and sleep deprivation on catecholamine and interleukin-2 levels in humans: clinical implications. J. Clin. Endocrinol. Metab. *84*, 1979–1985.

Issa, N.P., Trachtenberg, J.T., Chapman, B., Zahs, K.R., and Stryker, M.P. (1999). The critical period for ocular dominance plasticity in the ferret's visual cortex. J. Neurosci. *19*, 6955–6978.

Jones, B.E. (1994). Basic mechanisms of sleep-waking states. In Principles and Practice of Sleep Medicine, M.H. Kryger, T. Roth, W.C. Dement, eds. (Philadelphia: Saunders), pp. 145–162.

Jouvet-Mounier, D., Astic, L., and Lacote, D. (1970). Ontogenesis of the states of sleep in rat, cat and guinea pig during the first postnatal month. Dev. Psychobiol. *2*, 216–239.

Karni, A., Tanne, D., Rubenstein, B.S., Askenasy, J.J.M., and Sagi, D. (1994). Dependence of REM sleep of overnight improvement of a perceptual skill. Science *265*, 679–682.

Kasamatsu, T., Pettigrew, J.D., and Ary, M. (1979). Restoration of visual cortical plasticity by local microperfusion of norepinephrine. J. Comp. Neurol. *185*, 163–182.

Kudrimoti, H.S., Barnes, C.A., and McNaughton, B.L. (1999). Reactivation of hippocampal cell assemblies: effects of behavioral state, experience and EEG dynamics. J. Neurosci. *19*, 4090–4101.

Lancel, M., van Riezen, H., and Glatt, A. (1991). Effects of circadian phase and duration of sleep deprivation on sleep and EEG power spectra in the cat. Brain Res. *548*, 206–214.

Livingstone, M.S., and Hubel, D.H. (1981). Effects of sleep and arousal on the processing of visual information in the cat. Nature *291*, 554–561.

Louie, K., and Wilson, M.A. (2001). Temporally structured replay of awake hippocampal ensemble activity during rapid eye movement sleep. Neuron *29*, 145–156.

Lugazzi, R.F., Adrien, J., Bourgoin, S., and Hamon, M. (1979). Effects of intraventricular injection of 6-hydroxydopamine in the developing kitten. I. On the sleepwaking cycles. Brain Res. *160*, 445–459.

Macquet, P., Laureys, S., Peigneux, P., Fuchs, S., Petiau, C., Phillips, C., and Aerts, J. (2000). Experience-dependent changes in cerebral activation during human REM sleep. Nat. Neurosci. *3*, 831–836.

Malach, R., Ebert, R., and Van Sluyters, R.C. (1984). Recovery from effects of brief monocular deprivation in the kitten. J. Neurophysiol. *54*, 538–551.

Marks, G.A., Shaffery, J.P., Oksenberg, A., Speciale, S.G., and Roffwarg, H.P. (1995). A functional role for REM sleep in brain maturation. Behav. Brain Res. *69*, 1–11.

McEwen, B.S., and Sapolsky, R.M. (1995). Stress and cognitive function. Curr. Opin. Neurobiol. *5*, 205–216.

McGaugh, J.L. (2000). Memory—a century of consolidation. Science *287*, 248–251.

McGrath, M.J., and Cohen, D.B. (1978). REM sleep facilitation of adaptive waking behavior: a review of the literature. Psychol. Bull. *85*, 24–57.

Mioche, L., and Singer, W. (1989). Chronic recordings from single sites of kitten striate cortex during experience-dependent modifications of receptive-field properties. J. Neurophysiol. *62*, 185–197.

Mirmiran, M. (1995). The function of fetal/neonatal rapid eye movement sleep. Behav. Brain Res. *69*, 13–22.

Nakanishi, H., Sun, Y., Nakamura, R.K., Mori, K., Ito, M., Suda, S., Namba, H., Storch, F.I., Dang, T.P., Mendelson, W., et al. (1997). Positive correlations between cerebral protein synthesis rates and deep sleep in *Macaca mulatta*. Eur. J. Neurosci. *9*, 271–279.

Neuner-Jehle, M., Rhynery, T.A., and Borbely, A.A. (1995). Sleep deprivation differentially alters the mRNA and protein levels of neurogranin in the brain. Brain Res. *685*, 143–153.

Neuner-Jehle, M., Denizot, J.-P., Borbely, A.A., and Mallet, J. (1996). Characterization and sleep deprivation-induced expression modulation of dendrin, a novel dendritic protein in rat brain neurons. J. Neurosci. Res. *46*, 138–151.

Oksenberg, A., Shaffery, J.P., Marks, G.A., Speciale, S.G., Mihailoff, G., and Roffwarg, H.P. (1996). Rapid eye movement sleep deprivation in kittens amplifies LGN cell-size disparity induced by monocular deprivation. Dev. Brain Res. *97*, 51–61.

Olson, C.R., and Freeman, R.D. (1980). Cumulative effect of brief daily periods of monocular vision on kitten striate cortex. Exp. Brain Res. *38*, 53–56.

Pascual-Leone, A., Grafman, J., and Hallett, M. (1994). Modulation of cortical motor output maps during development of implicit and explicit knowledge. Science *263*, 1287–1289.

Pavlides, C., and Winson, J. (1989). Influences of hippocampal place cell firing in the awake state on the activity of these cells during subsequent sleep. J. Neurosci. *9*, 2907–2918.

Peck, C.K., and Blakemore, C. (1975). Modification of single neurons in the kitten's visual cortex after brief periods of monocular deprivation. Exp. Brain Res. *22*, 57–68.

Pettigrew, J.D., and Garey, L.J. (1974). Selective modification of single neuron properties in the visual cortex of kittens. Brain Res. *66*, 160–164.

Plihal, W., and Born, J. (1999). Effects of early and late nocturnal sleep on priming and spatial memory. Psychophysiology *36*, 571–582.

Poe, G.R., Nitz, D.A., McNaughton, B.L., and Barnes, C.A. (2000). Experience-dependent phase-reversal of hippocampal neuron firing during REM sleep. Brain Res. *855*, 176–180.

Qin, Y.L., McNaughton, B.L., Skaggs, W.E., and Barnes, C.A. (1997). Memory reprocessing in corticocortical and hippocampocortical neuronal ensembles. Philos. Trans. R. Soc. Lond. B Biol. Sci. *352*, 1525–1533.

Ramachandran, V.S., and Ary, M. (1982). Evidence for a "consolidation" effect during changes in ocular dominance of cortical neurons in kittens. Behav. Neural Biol. *35*, 211–216.

Ramm, P., and Smith, C.T. (1990). Rates of cerebral protein synthesis are linked to slow-wave sleep in the rat. Physiol. Behav. *48*, 749–753.

Rauschecker, J.P., and Hahn, S. (1987). Ketamine-xylazine anesthesia blocks consolidation of ocular dominance changes in kitten visual cortex. Nature *326*, 183–185.

Reiter, H.O., Waitzman, D.M., and Stryker, M.P. (1986). Cortical activity blockade prevents ocular dominance plasticity in the kitten visual cortex. Exp. Brain Res. *65*, 182–188.

Ribeiro, S., Goyal, V., Mello, C.V., and Pavlides, C. (1999). Brain gene expression during REM sleep depends on prior waking experience. Learn. Mem. *6*, 500–508.

Roffwarg, H.P., Muzio, J.N., and Dement, W.C. (1966). Ontogenetic development of the human sleep-dream cycle. Science *152*, 604–619.

Rottenberg, V.S. (1992). Sleep and memory II. Investigations on humans. Neurosci. Biobehav. Rev. *16*, 503–505.

Shaffery, J.P., Roffwarg, H.P., Speciale, S.G., and Marks, G.A. (1999). Ponto-geniculo-occipital wave suppression amplifies lateral genicu-

late nucleus cell-size changes in monocularly deprived kittens. Dev. Brain Res. *114*, 109–119.

Singer, W. (1979). Neuronal mechanisms in experience dependent modification of visual cortex function. In Development and Chemical Sensitivity of Neurons, Volume 31, M. Cuenod, G.W. Kreutzberg, and F.E. Bloom, eds. (Amsterdam: Elsevier/North-Holland Biomedical Press), pp. 457–477.

Singer, W. (1982). The role of attention in developmental plasticity. Hum. Neurobiol. *1*, 41–43.

Skaggs, W.E., and McNaughton, B.L. (1996). Replay of neuronal firing sequences in rat hippocampus during sleep following spatial experience. Science *271*, 1870–1873.

Smith, C. (1985). Sleep states and learning: a review of the animal literature. Neurosci. Biobehav. Rev. *9*, 157–168.

Smith, C. (1995). Sleep states and memory processes. Behav. Brain Res. *69*, 137–145.

Smith, C., and Lapp, L. (1986). Prolonged increases in both PS and number of REMS following a shuttle avoidance task. Physiol. Behav. *36*, 1053–1057.

Steriade, M. (1999). Coherent oscillations and short-term plasticity in corticothalamic networks. TINS *22*, 337–345.

Steriade, M., and Amzica, F. (1998). Coalescence of sleep rhythms and their chronology in corticothalamic networks. Sleep Res. Online *1*, 1–10.

Stickgold, R., LaTanya, J., and Hobson, J.A. (2000a). Visual discrimination learning requires sleep after training. Nat. Neurosci. *3*, 1237–1238.

Stickgold, R., Whifbee, D., Schirmer, B., Patel, V., and Hobson, J.A. (2000b). Visual discrimination task improvement: a multi-step process occurring during sleep. J. Cogn. Neurosci. *12*, 246–254.

Tobler, I., and Scherschlicht, R. (1990). Sleep and EEG slow-wave activity in the domestic cat: effect of sleep deprivation. Behav. Brain Res. *37*, 109–118.

Tobler, I., Murrison, R., Ursin, R., Ursin, H., and Borbely, A.A. (1983). The effect of sleep deprivation and recovery sleep on plasma corticosterone in the rat. Neurosci. Lett. *35*, 297–300.

Trachtenberg, J.T., Trepel, C., and Stryker, M.P. (2000). Rapid extragranular plasticity in the absence of thalamocortical plasticity in the developing primary visual cortex. Science *287*, 2029–2032.

Wilson, M.A., and McNaughton, B.L. (1994). Reactivation of hippocampal ensemble memories during sleep. Science *265*, 676–682.

Yinon, U., and Goshen, S. (1984). Survival of early monocular deprivation effects in cortical cells of kittens following prolonged dark rearing. Dev. Brain Res. *16*, 135–146.